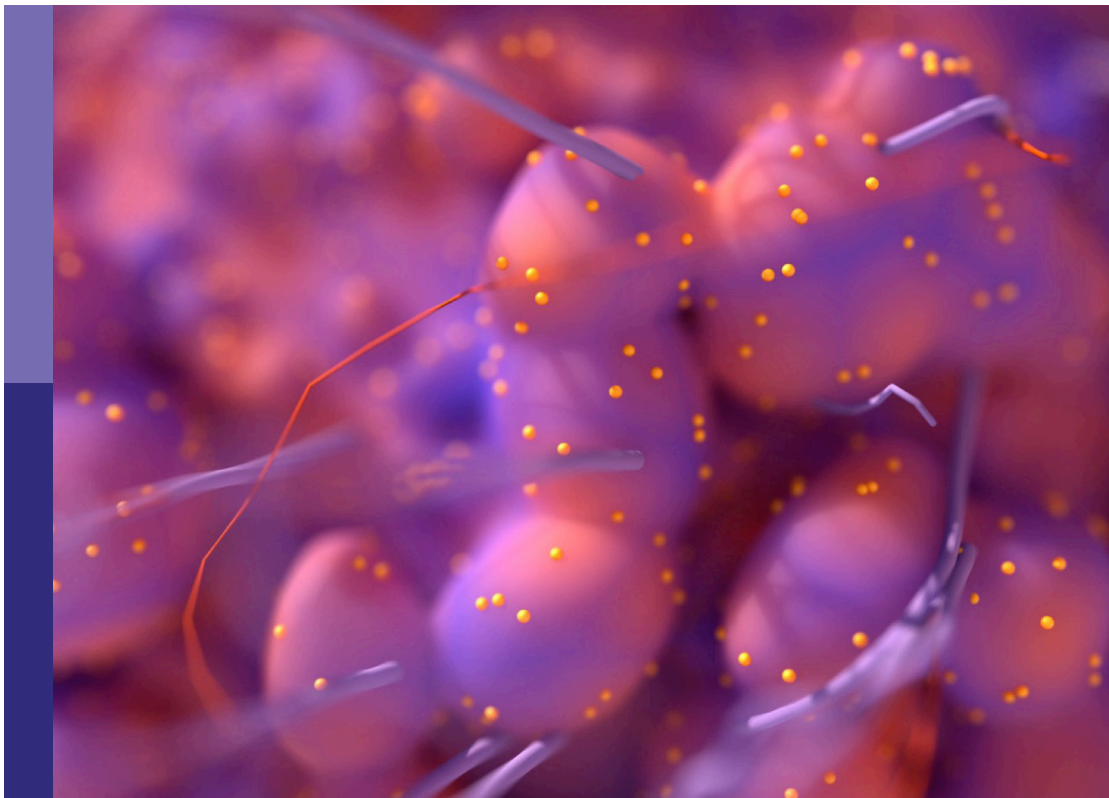
frontiers | Research Topics

Challenges in peripheral T-cell lymphomas: From biological advances to clinical applicability

Edited by
Juliana Pereira and Ryan Wilcox

Coordinated by
Luis Alberto De Pádua Covas Lage

Published in
Frontiers in Oncology
Frontiers in Immunology



ISSN 1664-8714
ISBN 978-2-8325-4208-8
DOI 10.3389/978-2-8325-4208-8

About Frontiers

Frontiers is more than just an open access publisher of scholarly articles: it is a pioneering approach to the world of academia, radically improving the way scholarly research is managed. The grand vision of Frontiers is a world where all people have an equal opportunity to seek, share and generate knowledge. Frontiers provides immediate and permanent online open access to all its publications, but this alone is not enough to realize our grand goals.

Frontiers journal series

The Frontiers journal series is a multi-tier and interdisciplinary set of open-access, online journals, promising a paradigm shift from the current review, selection and dissemination processes in academic publishing. All Frontiers journals are driven by researchers for researchers; therefore, they constitute a service to the scholarly community. At the same time, the *Frontiers journal series* operates on a revolutionary invention, the tiered publishing system, initially addressing specific communities of scholars, and gradually climbing up to broader public understanding, thus serving the interests of the lay society, too.

Dedication to quality

Each Frontiers article is a landmark of the highest quality, thanks to genuinely collaborative interactions between authors and review editors, who include some of the world's best academicians. Research must be certified by peers before entering a stream of knowledge that may eventually reach the public - and shape society; therefore, Frontiers only applies the most rigorous and unbiased reviews. Frontiers revolutionizes research publishing by freely delivering the most outstanding research, evaluated with no bias from both the academic and social point of view. By applying the most advanced information technologies, Frontiers is catapulting scholarly publishing into a new generation.

What are Frontiers Research Topics?

Frontiers Research Topics are very popular trademarks of the *Frontiers journals series*: they are collections of at least ten articles, all centered on a particular subject. With their unique mix of varied contributions from Original Research to Review Articles, Frontiers Research Topics unify the most influential researchers, the latest key findings and historical advances in a hot research area.

Find out more on how to host your own Frontiers Research Topic or contribute to one as an author by contacting the Frontiers editorial office: frontiersin.org/about/contact

Challenges in peripheral T-cell lymphomas: From biological advances to clinical applicability

Topic editors
Juliana Pereira — University of São Paulo, Brazil
Ryan Wilcox — University of Michigan, United States

Topic coordinator
Luis Alberto De Pádua Covas Lage — University of São Paulo, Brazil

Citation
Pereira, J., Wilcox, R., De Pádua Covas Lage, L. A., eds. (2024). *Challenges in peripheral T-cell lymphomas: From biological advances to clinical applicability*. Lausanne: Frontiers Media SA. doi: 10.3389/978-2-8325-4208-8

Table of contents

frontiers | Frontiers in Oncology

TYPE Editorial
PUBLISHED 13 December 2023
DOI 10.3389/fonc.2023.1325170

OPEN ACCESS

EDITED AND REVIEWED BY
Alessandro Isidori,
AORMN Hospital, Italy

*CORRESPONDENCE
Luís Alberto de Pádua Covas Lage
luis.lage@hc.fm.usp.br;
luisalberto_lage@yahoo.com.br

RECEIVED 20 October 2023
ACCEPTED 08 December 2023
PUBLISHED 13 December 2023

CITATION
Lage LAdPC, Pereira J and Wilcox RA (2023) Editorial: Challenges in peripheral T-cell lymphomas: from biological advances to clinical applicability.
Front. Oncol. 13:1325170.
doi: 10.3389/fonc.2023.1325170

Editorial: Challenges in peripheral T-cell lymphomas: from biological advances to clinical applicability

Luís Alberto de Pádua Covas Lage[1,2]*, Juliana Pereira[1,2,3] and Ryan A. Wilcox[4]

[1]Department of Hematology, Hemotherapy & Cell Therapy, Faculty of Medicine – University of São Paulo (FM-USP), São Paulo, Brazil, [2]Laboratory of Medical Investigation in Pathogenesis and Directed Therapy in Onco-Immuno-Hematology (LIM-31), Faculty of Medicine – University of São Paulo (FM-USP), São Paulo, Brazil, [3]Hospital Alemão Oswaldo Cruz (HAOC), Department of Hematology and Oncology, São Paulo, Brazil, [4]Department of Internal Medicine, Division of Hematology and Oncology, University of Michigan, Ann Arbor, MI, United States

KEYWORDS

peripheral T-cell lymphoma, pathogenesis, molecular biomarkers, epigenetic regulation, treatment

Editorial on the Research Topic
Challenges in peripheral T-cell lymphomas: from biological advances to clinical applicability

Peripheral (mature) T-cell lymphomas (PTCL) are a heterogeneous group of rare lymphoid malignancies derived from monoclonal proliferation of CD4+ T-helper cell subsets, CD8+ cytotoxic T-cells or natural-killer cells (NK). Although less prevalent than B-cell non-Hodgkin's lymphomas (NHLs), understanding of their molecular pathogenesis and classification has significantly improved over the last decade. However, these advances have not always translated into therapeutic improvement, and thus treatment of the NK/T-cell lymphomas remain an area of unmet medical need. Therefore, most PTCL patients still have poor clinical outcomes and substantially shortened survival when compared to individuals with aggressive B-cell lymphomas. This Special Edition includes 15 scientific articles that compile the main biological, pathological and therapeutic advances recently obtained in this field of knowledge, aiming to improve the clinical outcomes of PTCL patients.

The classification, risk-stratification and treatment of most PTCL subtypes remain, in the words of those who contributed to this Research Topic, "a challenge" [Murga-Zamalloa and Inamdar; Zain and Kallam]. In this sense, Murga-Zamalloa and Inamdar reviewed the most recent updates implemented by the latest version of the World Health Organization's classification of hematolymphoid tumors (WHO-HAEM 5th.), focusing on the most relevant diagnostic findings of PTCL, centered on histopathological basis and the description of new molecular markers. The discovery of new molecular biomarkers has contributed significantly to the elucidation of pathogenic mechanisms, prognostic stratification and implementation of therapeutic measures adapted to biological risk and directed against classically deregulated

signaling pathways in the setting of different subtypes of PTCL. Thus, Zain and Kallam and Drieux et al. prepared two comprehensive reviews in this Research Topic describing the main recurrent mutations, changes in the gene expression profile (GEP) and deregulated intracellular signaling pathways in PTCL. Additionally, the authors clarify how such molecular advances influence the therapeutic management of individuals with different subtypes of PTCL, highlighting the promising role of new drug classes directed against key molecular targets in these neoplasms, such as epigenetic modifying agents (histone deacetylase inhibitors [HDAi] and DNA methyl transferase antagonists [hypomethylants, such as 5-azacytidine and decitabine]), therapies that target kinases (PI3K, JAK, SYK, ALK and Aurora A kinase inhibitors), immunotherapeutic agents (anti-CD30, anti-CCR4, anti-CD25), and drugs that modulate the tumor microenvironment (TME), such as lenalidomide and immune checkpoint inhibitors.

Notwithstanding well-described geographic differences in PTCL prevalence, largely attributed to differences in the epidemiology of virally-associated, particularly EBV-related [Drieux et al., Barros et al.] NK/T-cell lymphoma subtypes [Costa et al.], the most common PTCL subtype remains "not otherwise specified" [Weiss et al.]. While Weiss et al. described recent advances in our understanding of transcriptionally, genetically, and clinically distinct PTCL, NOS subsets, these advances are discussed in broader histopathologic [Murga-Zamalloa and Inamdar] and clinical [Zain and Kallam] contexts by other notable contributions to this Research Topic. The reviews conducted by Weiss et al., Zain and Kallam and Drieux et al. focuses on the central contribution of GEP to mitigate the diagnostic and prognostic heterogeneity of PTCL, NOS. Therefore, these authors highlight the categorization of two genetic subgroups within this category of PTCL, the TBX21 subtype, associated with Th1/NF-kB deregulation, and the subgroup with overexpression of the Th2/GATA3 transcription factor, related to deregulation of the PI3K/mTOR axis. Such articles reinforce the prognostic differences between both groups of PTCL, NOS and, highlighting the highly ominous prognosis of the GATA3 subgroup, as well as the potential therapeutic application of immunomodulatory agents in the TBX21 group and PI3K/mTOR inhibitors in the GATA3 subtype.

While the molecular advances have not yet led to a formal update in the latest WHO classification [Murga-Zamalloa and Inamdar], improved understanding in the molecular pathogenesis of angioimmunoblastic T-cell lymphomas (AITL), including the role of highly recurrent loss-of-function mutations in TET2 [Carty et al.], have led to significant changes in the classification of these and other highly related T follicular helper (TFH)-derived PTCL [Lage et al.; Marques-Piubelli et al.], and improved therapeutic strategies, with the advent of novel agents targeting the epigenome [Zain and Kallam; Drieux et al.; Carty et al.; Lage et al.; Marques-Piubelli et al.] or the TME [Drieux et al.; Lage et al.]. In this sense, Lage et al. and Marques-Piubelli et al. conducted two extensive reviews summarizing the main clinical-laboratory, pathogenic and histopathological aspects of nodal peripheral T-cell lymphomas with TFH-phenotype (nPTCL-TFH), focusing on AITL. In their article, Lage et al. highlight the main findings that make up the so-called "*immunodysplastic syndrome*", characteristic of AITL, marked by different inflammatory and autoimmune features, as well as summarize the contribution of molecular changes involving the epigenetic machinery (IDH2, TET2, and DNMT3A) in the genesis of AITL [8]. Similarly, Marques-Piubelli et al. characterizes in their article the pathological findings of the main 3 subtypes of nPTCL-TFH in light of recent updates proposed by the WHO-HAEM5th. classification, list the antigenic expression profile of TFH-cells, and describe the main genomic-molecular findings of these malignancies. At the same time, in their review, Carty et al. dissects the biological role of TET2, particularly in nPTCL-TFH biology, highlighting its lymphomagenesis-promoting mechanisms and the potential of epigenetic therapies to improve the clinical outcomes of these neoplasms. Still in the therapeutic field of nodal PTCL, in the relapsed/refractory (R/R) setting, traditional chemotherapeutic agents do not provide an apparent advantage over novel drugs, and hence clinical trial participation is encouraged. However, Fante et al. highlight in their retrospective and single-center experience, the role of the all-oral and palliative regimen TEPIP (trophosphamide, etoposide, procarbazine, idarubicin and prednisolone) in elderly and frail patients with PTCL. In this study, the TEPIP regimen demonstrated competitive efficacy with a tolerable safety profile in a population with difficult-to-treat PTCL, making it a relatively safe and effective alternative in the palliative context of individuals with nodal PTCL.

As noted by Wu and Lim, the genetic landscape is a significant determinant of disease natural history and the response to therapy among anaplastic large cell lymphomas (ALCL). For example, those harboring recurrent *DUSP22* rearrangements are highly curable with the current standard of care based on BV-CHP (brentuximab-vedotin, cyclophosphamide, doxorubicin, and prednisone) regimen, whereas those harboring mutually exclusive *TP63* rearrangements are associated with dismal outcomes. While the molecular and genetic landscape has predictive value in PTCL, NOS [Weiss et al.] and ALCL [Wu and Lim], the natural history associated with AITL and other TFH-derived PTCL is notoriously variable, yet poorly understood. Hu et al. provide data suggesting that metabolic activity (total lesion glycolysis - TLG) by PET-CT may have prognostic implications in these patients, while Chen et al. provide preliminary evidence suggesting that this variable natural history may be explained, at least in part, by differences in tumor immune surveillance within the TME. In their pioneering study, Hu et al. identified that TLG was a strong predictor associated with poor overall survival in a cohort of 40 Chinese patients diagnosed with AITL and followed for more than 30 months. Additionally, these authors developed a new prognostic scoring system including different clinical-laboratory (IPI) and imaging/metabolic variables (TMTV, TLG and SUV max) in patients with AITL. By this score, three different risk-categories were identified, with 3-year overall survival (OS) estimates of 100%, 43%, and 25%, respectively. Interestingly, Chen et al. also conducted an unprecedented translational study, comparing clinicopathological findings, lymphocyte composition of the immune TME (TIL – tumor infiltrating lymphocytes), and gene expression profile between different subsets of AITL. In this study, the authors demonstrated that CD8-predominant AITL presents a very peculiar immune

pattern, characterized by compromised anti-tumor immunity, immunosuppressive microenvironment, more severe clinical manifestations and worse survival, thus clarifying the heterogeneity of this clinicopathological entity.

The TME plays a central role in both the suppression of host anti-tumor immunity, but its constituents also directly promote T-cell lymphomagenesis by providing ligands for corresponding antigen, costimulatory, and cytokine receptors that are variously expressed across the PTCL spectrum. Not surprisingly then, the TME´s role in T-cell lymphomas generally, including CTCL [Miyashiro and Sanches], is a recurring theme across many papers included in this Research Topic, and has significant therapeutic implications in the era of checkpoint blockade [Costa et al.; Weiss et al.] and novel cellular therapies [Couto et al.]. Concerning to this last Research Topic, in their review article, Couto et al. discusses the use of autologous hematopoietic stem cell transplantation (ASCT) and allogeneic transplantation (allo-SCT) as therapeutic strategies for PTCL, as well as approaches based on the advancement of cellular therapy techniques, in addition to their limitations in the PTCL scenario, proposing some approaches to overcome them.

While the papers included in this Research Topic are diverse, a unifying theme is abundantly clear - improved understanding of the transcriptional, genetic, and TME-dependent drivers across the spectrum of NK/T-cell neoplasms has led to improved disease classification and risk-stratification, but has also expanded the menu of novel agents available on the therapeutic smorgasbord, and thus provides hope for a brighter future for new patients afflicted with these aggressive neoplasms.

Author contributions

LL: Writing – review & editing. JP: Writing – review & editing. RW: Writing – review & editing.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

frontiers | Frontiers in Immunology

TYPE Original Research
PUBLISHED 17 October 2022
DOI 10.3389/fimmu.2022.987227

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Igor Bucchwalow,
Institute For Hematopathology
Hamburg, Germany
Kenichi Yoshida,
National Cancer Center Research
Institute, Japan
Kengo Takeuchi,
Japanese Foundation For Cancer
Research, Japan

*CORRESPONDENCE
Sha Zhao
hxblzhaosha@126.com
Yuan Tang
1202ty@163.com

†These authors contributed equally to this work and share first authorship

SPECIALTY SECTION
This article was submitted to Cancer Immunity and Immunotherapy, a section of the journal Frontiers in Immunology

RECEIVED 05 July 2022
ACCEPTED 27 September 2022
PUBLISHED 17 October 2022

CITATION
Chen Z, Zhu Q, Deng X, Yao W, Zhang W, Liu W, Tang Y and Zhao S (2022) Angioimmunoblastic T-cell lymphoma with predominant CD8+ tumor-infiltrating T-cells is a distinct immune pattern with an immunosuppressive microenvironment.
Front. Immunol. 13:987227.
doi: 10.3389/fimmu.2022.987227

Angioimmunoblastic T-cell lymphoma with predominant CD8+ tumor-infiltrating T-cells is a distinct immune pattern with an immunosuppressive microenvironment

Zihang Chen†, Qiqi Zhu†, Xueqin Deng, Wenqing Yao, Wenyan Zhang, Weiping Liu, Yuan Tang* and Sha Zhao*

Department of Pathology, West China Hospital, Sichuan University, Chengdu, China

Background: Angioimmunoblastic T-cell lymphoma (AITL) has a rich tumor microenvironment (TME) that typically harbors plenty of CD4+tumor infiltrating lymphocytes, (TIL)-T-cells (so called common AITL). Nonetheless, AITL with large numbers of CD8+TIL-Ts that outnumber CD4+cells have been observed (CD8-predominant AITL). However, detailed comparison of CD8-predominant AITL and common AITL are still lacking.

Methods: We compared clinicopathological features, TIL subsets, TME T cell receptor-β (TRB), and immunoglobulin heavy chain (IGH) repertoires, and gene expression profiles in six CD8-predominant and 12 common AITLs using case-control matching (2014 to 2019).

Results: Comparing with common AITLs, CD8-predominant AITLs showed more frequent edema (P = 0.011), effusion (P = 0.026), high elevated plasma EBV-DNA (P = 0.008), and shorter survival (P = 0.034). Moreover, they had more pronounced eosinophil increase (P = 0.004) and a higher Ki67 index (P = 0.041). Flow cytometry revealed an inverted CD4/CD8 ratio in TIL-Ts and lower TIL-B proportions (P = 0.041). TRB repertoire metrics deteriorated, including lower productive clones (P = 0.014) and higher clonality score (P = 0.019). The IGH repertoire was also narrowed, showing a higher proportion of the top 10 clones (P = 0.002) and lower entropy (P = 0.027). Gene expression analysis showed significant enrichment for upregulated negative regulation of immune system processes and downregulated T-cell activation and immune cell differentiation.

Conclusion: Our findings demonstrated that CD8-predominant AITL is a distinct immune pattern of AITL characterized by anti-tumor immunity impairment and

10.3389/fimmu.2022.987227

an immunosuppressive microenvironment. These characteristics can interpret its severe clinical manifestations and poor outcomes.

KEYWORDS

angioimmunoblastic T-cell lymphoma, tumor microenvironment, T cell receptor-β repertoire, immunoglobulin heavy chain repertoire, TME function

Introduction

Angioimmunoblastic T-cell lymphoma (AITL) is a subtype of peripheral T-cell lymphoma (PTCL) with a T follicular helper (TFH) phenotype (1). In our previous analysis, it was the fourth most common mature T-cell lymphoma, accounting for 11% of all patients with PTCL (2). Patients with AITL frequently exhibit clinical symptoms, including skin rash, arthritis, effusion, and positive autoimmune tests, indicating an aberrant immune response (3, 4). Finally, patients exhibit immunodeficiency secondary to neoplastic development. Even following an intensive treatment regimen, the prognosis is generally poor, with most studies reporting a median survival of < 3 years (5). As a result, immunological dysregulation characterizes the entire course of AITL and has been identified as a distinguishing feature of this malignancy.

AITL possesses a complex tumor microenvironment (TME) composed of irregularly proliferating follicular dendritic cells in the meshwork, arborizing high endothelial venules (HEVs) and a variety of reactive immune cells, the most obvious of which are B-immunoblasts, plasma cells, and tumor-infiltrating lymphocyte (TIL) -T cells (TIL-Ts) (6). Immunostaining has revealed that AITL TIL-Ts often have a significantly larger proportion of CD4+ cells than CD8+ cells (defined as common AITL in our study), which is due to CD4 expression in neoplastic T-cells and a normal CD4+/CD8+ ratio in TIL-Ts (CD4+TIL-Ts outnumber CD8+TIL-Ts) (7, 8). Nonetheless, we observed a few cases of AITL with an extremely inverted CD4/CD8 ratio (defined as CD8-predominant AITL in our study), indicating the significant increase of CD8+TIL-Ts. However, CD8-predominant AITL was rare, accounting for only 1.5% of AITL cases in daily practice (Time: 2014-2019).

The different CD8+ TIL-T proportions in the TMEs of CD8-predominant AITL and common AITL implied that TME immune function differed between the two AITL groups (7). Furthermore, the immune function of the TME not only correlated with the patient's clinical manifestations related to inflammation and immune response but also reflects anti-tumor immunity, which has a direct impact on tumor development and progression. However, detailed comparison of CD8-predominant AITL and common AITL are still lacking. Therefore, we used a case-control matching approach to include cohorts of CD8-predominant AITL and common AITL cases for clinicopathological analysis, flow cytometry testing, T cell receptor-β (TRB), immunoglobulin heavy chain (IGH) repertoire sequencing, and RNA sequencing. Data were compared to gain a better understanding of the TME immune function of this uncommon CD8-predominant AITL and its connection with clinicopathological findings.

Methods

Case selection

The CD8-predominant AITL cohort was identified from the database of the Department of Pathology of West China Hospital, Sichuan University from January 2014 to October 2019 according to the following criteria: 1) Diagnosed with lymph node sample and fulfilled the WHO-classified diagnostic criteria of AITL (4th edition, 2008/Revised 4th edition, 2017); 2) identification of neoplastic T-cells with aberrant expression of T cell markers by flow cytometry (9); 3) identification of CD8+cells/CD4+cells > 1 by immunostaining; and 4) *de novo* cases. For comparison, we employed case-control matching to randomly choose *de novo* common AITL cases (diagnosed with lymph node sample) from a list of potential matched controls throughout the same period (2014–2019, Supplementary Method in detail). In order to eliminate the influence of confounding factors on the prognosis, we matched the CD8-predominant and common AITLs based on the neoplastic T-cell proportion (detected by flow cytometry) and treatment strategy. Two common AITL cases were matched to each CD8-predominant AITL case. Case-control matching was performed using SPSS (v24.0; SPSS Corp., Chicago, IL, USA). Pathological data were gathered *via* slide review by four expert hematopathologists (Z.C., S. Z., W. Z., and W. L.). The pathological analysis, flow cytometry, and the following molecular analysis were all performed on the same lymph node sample of each case. Detailed clinical data were collected from medical records. Follow-up data were obtained *via* telephone interviews and/or medical records. Overall survival (OS) was calculated from the date of diagnosis to the date of death or the last follow-up. The flowchart of case selection was provided in supplementary

method (Supplementary Material Data Sheet 1). The study was approved by the Ethics Committee on Biomedical Research, West China Hospital of Sichuan University (No. 2021-628).

Histological, immunohistochemical, and Epstein–Barr virus (EBV) status assessment

Sections (4 μm) were cut from paraffin blocks and stained with hematoxylin and eosin for histological examination. Immunohistochemical staining was performed using the following antibodies: cytoplasmic CD3 (cCD3), CD20, CD5, CD4, CD8, CD10, CD21, Bcl-6, CXCL13, PD1, CD30, and Ki-67. The basic information of the antibodies is summarized in the Supplementary Method (Supplementary Material Data Sheet 1). *In situ* hybridization with a digoxin-labeled oligonucleotide probe complementary to EBER-1 and EBER-2 (EBER1/2; Dako, NO. Y520001) was used to assess EBV status.

Flow cytometry

The process for sample preparation, staining, acquisition, data analysis, and identification of neoplastic T-cells and TILs was performed in the same manner as previously described (9–11). The fluorescent-labeled antibodies used in this study are listed in Supplementary Method (Supplementary Material Data Sheet 1). Flow cytometry data were collected using the BD FACSCanto II equipped with two lasers (blue and red) and analyzed with FACSDiva software. The proportion of neoplastic T-cells and various TIL subsets, including TIL-Bs ($CD20^+$), activated TIL-Bs ($CD38^{dim+}CD20^+$), non-activated TIL-Bs ($CD38^-CD20^+$), TIL-Ts ($CD3^+$), CD4+TIL-Ts, CD8+TIL-Ts, and plasma cells ($CD38^{bight+}CD20^-$) were defined as counts of each type of cell/total counts of lymphocytes.

TRB and IGH sequencing

DNA was isolated from formalin-fixed paraffin-embedded tissues using the QIAamp DNA FFPE Tissue Kit (Qiagen Inc., Valencia, CA, USA). For TRB and IGH repertoire sequencing, the LymphoTrack kits (TRB Assay and IGH Assay) were used following the manufacturer's instructions with 50 ng of AITL specimen DNA as a template. Primers in the LymphoTrack assays were designed using Illumina adapters. Each amplicon was purified using AMPure XP beads (Beckman Coulter, Brea, CA, USA) and quantified using an Agilent 2100 Bioanalyzer (Santa Clara, CA, USA). Subsequently, the samples were sequenced on an Ion PGM (Thermo Fisher Scientific, Loughborough, UK). Sequencing reads were then aligned using MiXCR (v3.0.13), and only productive rearrangements were included in further analyses. Lymphoma-derived clone were identified based on TRB clonal rearrangement analysis following the manufacturer's instructions and removed to obtain the TRB repertoire of the AITL TME (Supplementary Method in detail). The proportion of the top 10 clones ["% top 10 maximal frequency clones"; Frequency of each clone in AITL TME = reads of clone/(Total reads – Reads of lymphoma clone)], Shannon's entropy (entropy), and the clonality score were calculated. The numbers of productive clones in the samples were identified by calculating the number of "productive rearrangements". This is a measure of the number of functional T or B cells with a distinct TR or IGH rearrangement which represents the "richness." Entropy measures both the sample richness and the degree of unevenness in clone frequencies. The clonality score describes the "evenness" of the distribution of TR or IGH clones in the repertoire, that is, how much of the TRB or IGH repertoire is composed of expanded clones independent of sample size. The top 10 clones is the frequency of the top 10 dominant clones identified in each sample. The distribution of V-gene segment usage by both TRB and IGH in CD8-predominant AITL and common AITL were compared. Both shared and differential clones with a specific complementarity determining region 3 (CDR3) sequence between the two AITL groups were identified. These analyses were performed on R using the "LymphoSeq" and "tcR" packages (12–15).

RNA sequencing

The procedures of RNA sequencing, quality control, and normalization of data were performed by Shanghai Rightongene Biotechnology Co. Ltd (Shanghai, China), and the differentially expressed genes were identified using the R package "limma" (v3.38.3).

Statistical analysis

Continuous variables were analyzed using the Mann–Whitney test, while categorical variables were compared using Fisher's exact test. Survival data were calculated using the Kaplan–Meier estimator. The log-rank test was used to compare survival functions. Tests were considered significant at two-sided probability values less than 0.05 ($P < 0.05$). All results were analyzed using GraphPad Prism (v8.0.2; GraphPad Software Inc., USA) or SPSS (v24.0; SPSS Corp., Chicago, IL, USA).

Results

Clinical findings

This study included six CD8-predominant AITL cases and 12 matched common AITL cases. The clinical comparison between

CD8-predominant AITL and common AITL is summarized in Table 1. The detailed clinical features of all AITL cases included in this study are summarized in Supplementary Table S1. The CD8-predominant AITL cases included five men and one woman with a median age of 64.5 years (range: 48–83 years). Complete clinical data were not obtained for one patient, and the remaining five patients were classified as advanced stage (stage III/IV) with frequent splenomegaly (5/5, 100%), B symptoms (4/5, 80%), and hepatomegaly (3/5, 60%). In general, these baseline features were comparable to the matched common AITL cases. Edema (6/6,100%, P = 0.011) and serous effusion (5/5,100%, P = 0.026) were more prevalent in CD8-predominant AITL than in common AITL cases. In laboratory studies, CD8-predominant AITL exhibited findings identical to those of common AITL cases in routine blood tests and immunological assays. While lower fibrinogen (2.09 g/L, range: 1.60–2.56 g/L, P = 0.038) and more common decreased C4 (3/3, 100%, P = 0.033) were detected in CD8-predominant AITL than in common AITL cases. Highly elevated plasma EBV-DNA (> 10^4 copies/ml) was detected in all tested CD8-predominant AITL (3/3, 100%) but not in any common AITL cases (0/7, P = 0.008).

Pathological findings

Histopathological comparisons between CD8-predominant AITLs and matched common AITLs are summarized in Table 2. In general, CD8-predominant AITL had characteristic AITL

TABLE 1 Clinical comparison between microenvironment CD8-predominant AITLs and matched normal AITLs.

	CD8-predominant AITL	Common AITL	*P*
Age [median, (range)]	64.5y(48y-83y)	59y(43y-82y)	0.483
Sex (male/female)	5/1	7/5	0.600
Stage			1.000
I/II	0 (0)	1 (17)	
III/IV	5 (100)	5 (83)	
PS ≥2	2 (40)	3 (27)	1.000
B symptoms	4 (80)	7 (58)	0.600
Fever	3 (60)	5 (42)	0.620
Night sweat	0 (0)	3(25)	0.515
Weight loss	1 (20)	4 (33)	1.000
Skin rash	1 (20)	5 (45)	0.588
Edema	6 (100)	3 (30)	***0.011***
Serous effusion	5 (100)	3 (30)	***0.026***
Splenomegaly	5 (100)	4 (44)	0.086
Hepatomegaly	3 (60)	1 (11)	0.095
Anemia	3 (60)	7 (64)	1.000
Thrombocytopenia	2 (40)	0 (0)	0.083
Leukocytosis	3(60)	1 (9)	0.063
EO ($\times10^9$/L) [median, (range)]	0.22 (0.02-5.86)	0.25 (0.01-1.16)	0.827
FIB (g/L) [median, (range)]	2.09 (1.60-2.56)	3.08 (1.84-6.79)	***0.038***
Elevated LDH	3 (75)	5 (71)	1.000
CD4+T-cells in PB (%) [median, (range)]	23.90 (12.00-32.60)	25.2 (19.70-36.30)	1.000
CD8+T-cells in PB (%) [median, (range)]	48.00 (33.40-57.00)	31.00 (18.50-40.70)	0.111
Elevated IgG	2 (50)	4 (57)	1.000
Elevated IgA	2 (50)	4 (57)	1.000
Abnormal IgM	2 (50)	3 (43)	1.000
Elevated IgE	2 (67)	2 (33)	0.524
Decreased C3	3 (100)	3 (43)	0.200
Decreased C4	3 (100)	1 (14)	***0.033***
High plasma EBV-DNA (>10^4 copies/ml)	3 (100)	0 (0)	***0.008***
Chemotherapy	3 (50)	6 (50)	1.000

PS, performance status; EO, Eosinophils; FIB, Fibrinogen; PB, peripheral blood. Bold value indicates statistically significant (P < 0.05).

TABLE 2 Pathological comparison between CD8-predominant AITLs and matched normal AITLs.

	CD8-predominant AITL	Common AITL	*P*
Effaced LN architecture	6 (100)	9 (75)	0.515
Diffuse growth pattern	6 (100)	9 (75)	0.515
Arborizing proliferation of HEVs	6 (100)	12 (100)	1.000
Cell size			0.254
Small	0 (0)	0 (0)	
Small-Medium	0 (0)	2 (16)	
Medium	2 (33)	5 (42)	
Medium-Large	4 (67)	5 (42)	
Large	0 (0)	0(0)	
EO increase (>30/HPF)	5 (83)	1 (8)	***0.004***
TFH cell marker			0.686
2 markers	0 (0)	1 (8)	
3 markers	4 (67)	5 (42)	
4 markers	2 (33)	6(50)	
CD30 (%)	20 (0-30)	5 (0-15)	***0.020***
Ki67 (%)	60 (50-70)	50 (15-80)	***0.041***
EBER/HPF	7.5 (1-150)	3.5 (0-20)	0.263

TFH, T follicular helper; LN, lymph node; EO, eosinophil; HEVs, high endothelial venules; HPF, high power filed. Bold value indicates statistically significant (P < 0.05).

histological features, with effaced lymph node architecture (6/6, 100%, Figure 1A) in a diffuse growth pattern (6/6, 100%, Figure 1B) harboring arborizing proliferation of HEVs (6/6, 100%, Figure 1C). In two cases, the proliferating cells were medium-sized (Figure 1E), and in four cases, they were medium-large sized (Figure 1F). These morphological findings were consistent with those found in the matched common AITL cases. Nonetheless, CD8-predominant AITL exhibited more significant eosinophil increase [eosinophil cell count > 30/high power filed (HPF), 5/6, 83%, Figure 1D] than common AITL cases (1/12, 8%, $P = 0.004$), which is a distinguishing morphological feature. Immunohistochemically, CD8-predominant AITL was diffusely positive for CD3 (Figure 2A) and ≥3 TFH markers (Figures 2D-F). CD8+cells outnumbered CD4+cells (Figures 2B, C). CD8-predominant AITL exhibited a higher CD30 positive rate (median: 20%, range: 0–30%, $P = 0.020$) and Ki67 index (median: 60%, range: 50–70%, $P = 0.041$) than common AITL. All CD8-predominant AITL cases tested positive for EBER with a median positive cell count of 7.5/HPF (range: 1–150/HPF), which is higher than in common AITL (median: 3.5/HPF, range: 0–20/HFP), although not statistically significant ($P = 0.263$).

Flow cytometry evaluated neoplastic T-cell and TIL subset proportions in CD8-predominant AITLs and common AITLs

The neoplastic T-cells in CD8-predominant AITLs had a median proportion of 13% (neoplastic T-cells/total lymphocytes; range: 3.5–21.8%, Table S1). The CD4+TIL-Ts/CD8+TIL-Ts was ranged from 0.09 to 0.41 (Table S1). A typical case (C1) is shown in Figure 3A. The included common AITLs had matched tumor cell proportions with a median of 12.1% (range: 4.3–23.5%, Table S1).

TIL subset proportions in CD8-predominant AITLs differed from those in common AITLs. CD8-predominant AITLs had a significantly higher proportion of total TIL-Ts (CD8-predominant AITL: median: 60.8%, range: 45.4–70.3%, vs. common AITL: median: 37.8%, range: 17.4–65.8%, P = 0.025), CD8+TIL-Ts (CD8-predominant AITL: median: 47.5%, range: 33.6–64.0%, vs. common AITL: median: 16.9%, range: 7.4–35.5%, p < 0.001), and lower CD4+TIL-Ts (CD8-predominant AITL: median: 10.7%, range: 5.7–17.3%, vs. common AITL: median: 19.9%, range: 9.9–35.5%, P = 0.013). Furthermore, total TIL-Bs (median: 9.1%, range: 2.9–36.7%), activated TIL-Bs (median: 0.9%, range: 0–2.9%), and non-activated TIL-Bs (median: 3.9%, range: 0–19.0%) had lower proportions in CD8-predominant AITLs than in common AITLs (total TIL-Bs, median: 30.4%, range: 11.9–67.6%, p=0.041; activated TIL-Bs, median: 4.1%, range: 1.2–16.5%, P = 0.027; non-activated TIL-Bs, median: 17.1%, range: 3.3–64.0%, P = 0.038). However, there was no significant difference between CD8-predominant and common AITLs in the proportion of TIL-plasma cells. Figure 3B depicts the TIL subset comparisons in flow cytometry analysis.

TRB repertoires

TRB sequencing was performed on five of six patients with CD8-predominant AITL and all 12 patients with common

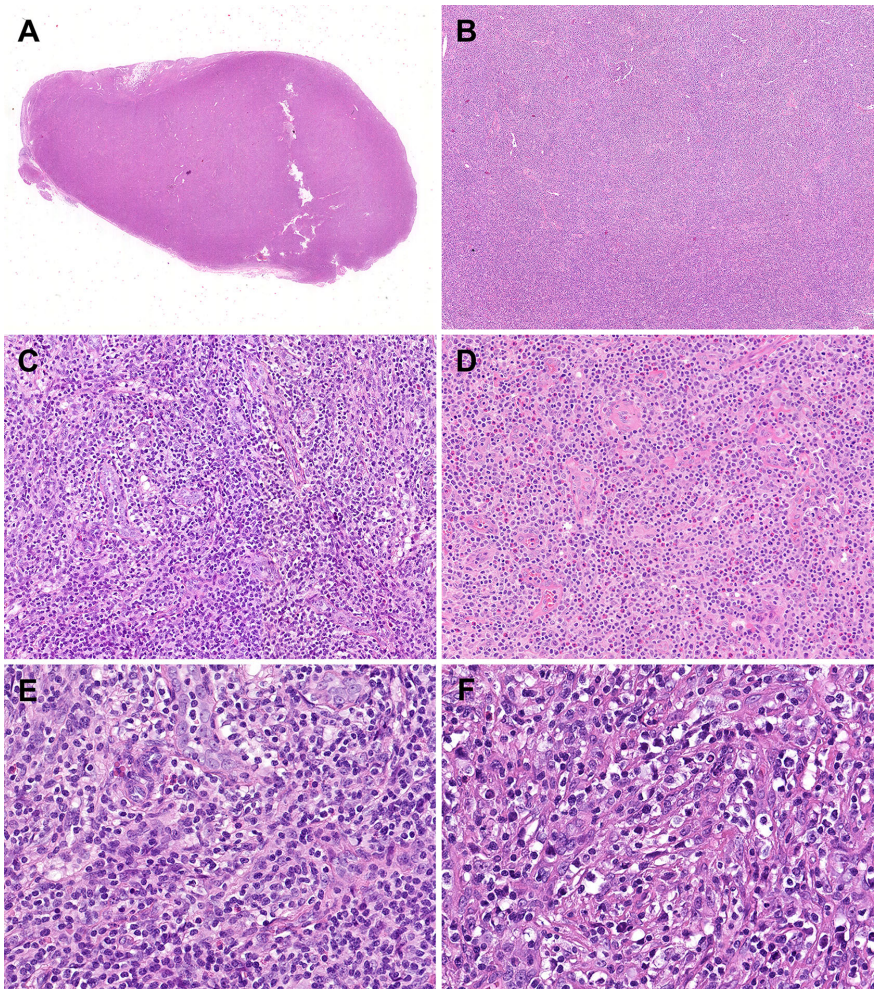


FIGURE 1
Morphological features of CD8-predominant AITL. In the hematoxylin & eosin staining, the CD8-predominant AITL exhibited **(A)** effaced lymph node architecture (×10), **(B)** in a diffuse growth pattern (×40), **(C)** with arborizing HEV proliferation (×200) and **(D)** eosinophil increase (×200). **(E)** Medium-sized (×400) or **(F)** medium-large-sized (×400) neoplastic T-cells with clear to pale cytoplasm surrounding HEVs.

AITL. For the TRB repertoire in the TME, CD8-predominant AITLs exhibited significantly lower productive clones (median: 998, range: 443–1378) than those of common AITLs (median: 1727, range: 997–3444, P = 0.014, Figure 4A). The CD8-predominant AITLs had a significantly higher proportion of the top 10 clones [CD8-predominant AITLs median top 10 clones' proportion of 39.1% (range 26.1–73.2%) vs. common AITL median top 10 clones' proportion of 20.6% (range 6.9–45.1%), P = 0.009], and higher clonality score [CD8-predominant AITLs median clonality score 0.312 (range 0.205–0.472) vs. common AITL median clonality score 0.191 (range 0.091 –0. 347), P = 0.019]. The entropy did not differ between the two groups (Figure 4A).

The global usage of TRBV segments in the TRB repertoire was not different between the two AITL groups (Figure 4B). However, we found significantly increased usage of the TRBV7 family segment (median: 8.8% vs. 5.5%, P = 0.045) and lower usage of the TRBV24 family (median: 0.3% vs. 0.7%%, P = 0.015) in CD8-predominant AITLs than in common AITLs (Figure 4A). A shared clone with the "CASSFSTCSANYGYTF" CDR3 sequence was identified in all 17 tested AITLs. Furthermore, we found that 18 CDR3 sequences were more frequently identified in CD8-predominant AITLs than in common AITLs ($p<0.05$, Figure 4C). All shared and differential TRB CDR3 data are provided in Supplementary Table S2.

IGH repertoires

IGH sequencing was performed on the same samples as the TRB sequencing. The productive clones were not different between CD8-predominant AITLs and common AITLs (Figure 4D). CD8-

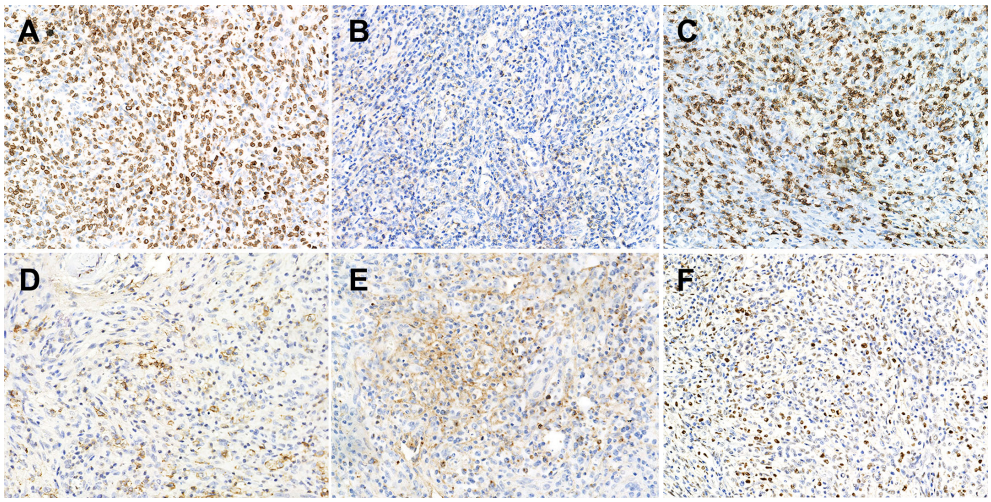


FIGURE 2
Immunophenotype of CD8-predominant AITL. **(A)** CD3 (×200); **(B)** CD4 (×200); **(C)** CD8 (×200); **(D)** PD1 (×200); **(E)** CXCL13 (×200); **(F)** Bcl-6 (×200).

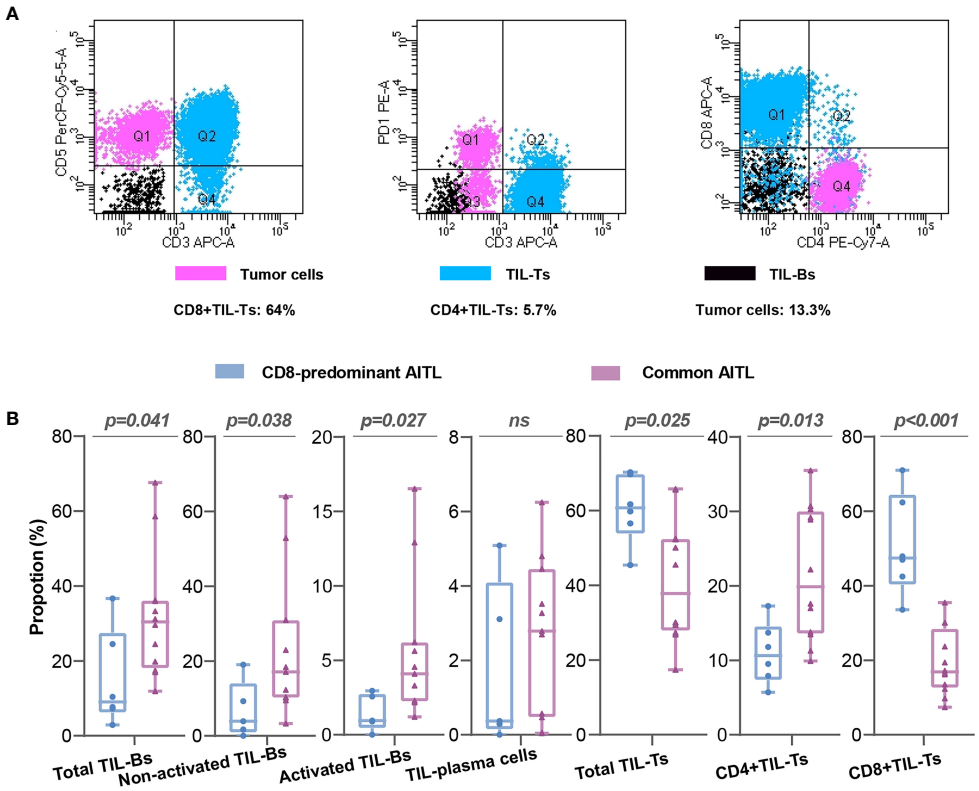


FIGURE 3
Flow cytometry analysis of CD8-predominant AITLs and matched common AITLs. Flow cytometry analysis of a typical case (C1) of CD8-predominant AITL **(A)** Comparisons of TIL subsets in flow cytometry analysis between CD8-predominant AITLs and matched common AITLs **(B)**. "ns" indicates "no significance".

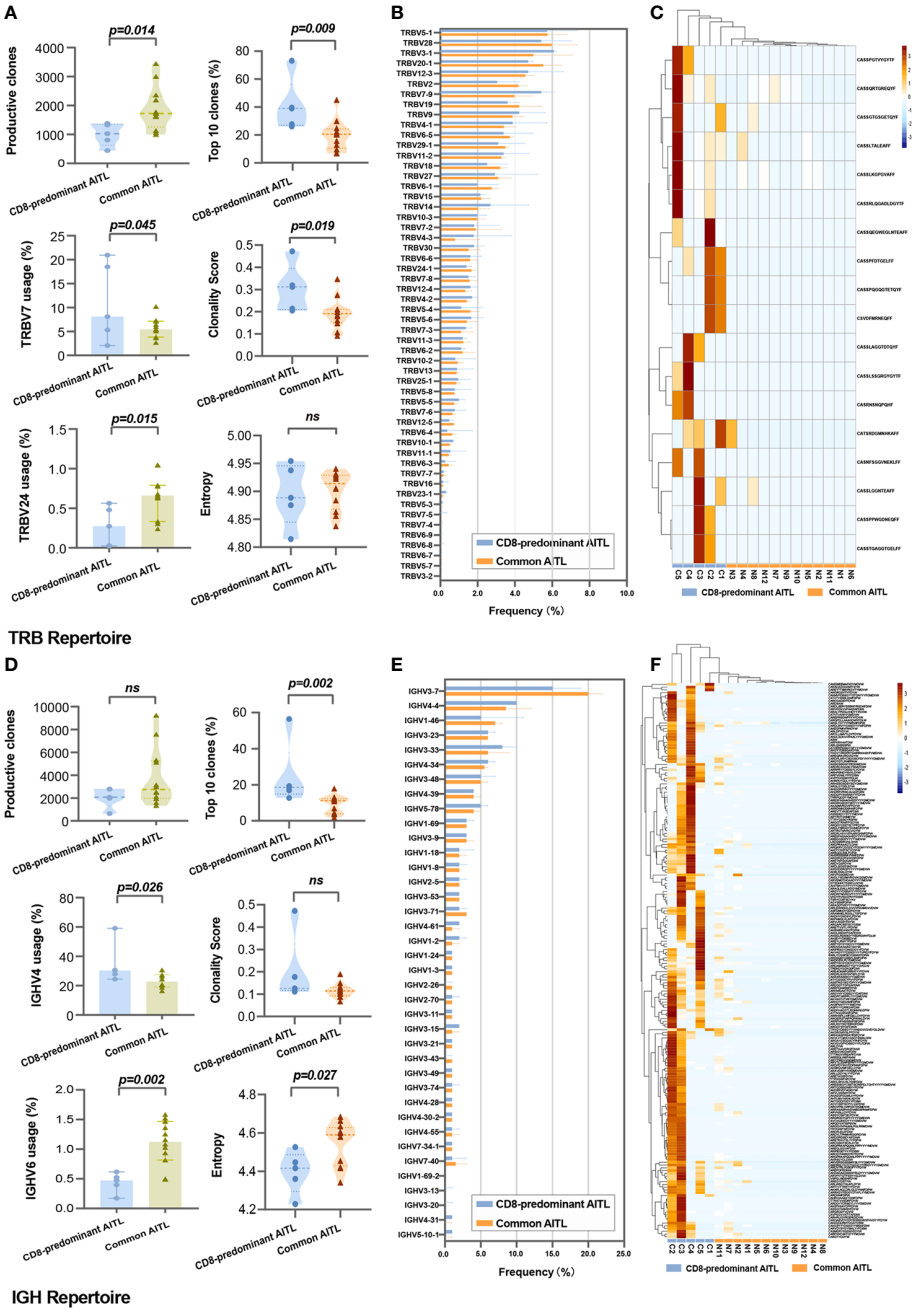


FIGURE 4
Comparison of TRB and IGH repertoires between CD8-predominant AITLs and matched common AITLs. Comparison between two AITL groups TME in statistical metrics including production clones, V-gene family usage, top 10 clones' proportion, clonality score, and entropy of TRB repertoire **(A)** and IGH repertoire **(D)**, in global usage of V-gene of TRB repertoire **(B)** and IGH repertoire **(E)**, and in the CDR3 sequence frequency of the TRB **(C)** and IGH repertoires **(F)**. "ns" indicates "no significance".

predominant AITLs had a higher proportion of the top 10 clones [CD8-predominant AITLs top 10 clones, 18.5% (range 12.7–56.5%) vs. common AITL top 10 clones, 11.1% (range 1.9–17.9%), P = 0.002], lower entropy [CD8-predominant AITLs median entropy 4.415 (range 4.229–4.526) vs. common AITL median entropy 4.589 (range 4.341–4.684), P = 0.027]. The clonality score did not differ between the two groups (Figure 4D).

The global usage of IGHV segments did not differ between CD8-predominant AITLs and common AITLs, with IGHV3-7 being the most frequently used segment in both AITL groups

(Figure 4E). Then, CD8-predominant AITL utilized the IGHV4 family segment more frequently (median: 30.3% vs. 22.7%, P = 0.026) and utilized the IGHV6 family segment less frequently (median: 0.5% vs. 1.1%, P = 0.002, Figure 4D) than common AITLs. We also found a shared IGH CDR3 sequence of "CARPYSYGYGDYVYYYGMDVW" in all tested AITL cases, which was identified more frequently in CD8-predominant AITLs (median: 10.4%, range: 1.1% - 48.9%) than in common AITLs (median: 0.9%, range: 0.01% - 8.6%, P = 0.027). Moreover, we discovered 303 IGH CDR3 sequences that differed in frequency between the two AITL groups (Figure 4F displayed differential IGH CDR3 with p < 0.033, n = 201). Supplementary Table S3 contains all the shared and differential IGH CDR3 sequences.

Comparative gene expression profiling between CD8-predominant and common AITL for inflammation and immune response

RNA sequencing was performed on three CD8-predominant AITLs and four common AITLs. To compare these two groups for inflammation and immune response, we created a panel of 772 genes based on the gene ontology biological process annotation (Supplementary Method in detail, the gene expression profile data were provided in Supplementary Table S4). Comparative analysis identified 36 genes in this panel with significant differential expression between CD8-predominant AITL and common AITL through the linear model fit with p-value correction (Padj < 0.05 and log2fc > 1), of which 20 genes were upregulated and 16 genes were downregulated in CD8-predominant AITLs (Figure 5A). Functional annotation analysis revealed predominant enriched differentially expressed genes in T-cell activation, negative regulation of immune system process, regulation of leukocyte activation, and regulation of lymphocyte activation (Figure 5B). CD8-predominant AITL was considerably enriched for upregulated negative regulation of immune system processes (Figure 5C) and downregulated T-cell activation and immune cell differentiation (Figure 5D) compared with common AITL, which suggested suppressed immune function.

Survival analysis

Survival data were obtained from all six patients with CD8-predominant AITL and 11 of the 12 patients with common AITL. Four patients with CD8-predominant AITL (4/6, 67%) and three patients with common AITL (3/11, 27%) died of the disease. Besides tumor proportion and treatment strategy, the other recognized prognostic factors were also comparable between the two AITL groups (Table 1). The prognosis of patients with CD8-predominant AITL (median overall survival: 5.0 months, 95%CI: 0–14.4 months) was significantly worse than that for those with common AITL (median overall survival not reached, P = 0.034, Figure 5E).

Discussion

AITL is highly heterogeneous, and exhibits complex TMEs (16). Our previous research has discovered that the ratio of the components, particularly TILs, varies in AITL cases (9). When CD8+TIL-Ts proliferated abundantly, they outnumbered CD4+ cells, formatting the CD8-predominant AITL. Because TME is directly associated with immune response, investigating TME immune activity in CD8-predominant AITL may facilitate immune-subtyping and prognostic stratification of AITL cases. We conducted an integrated analysis of the TME of CD8-predominant AITLs and compared them to common AITLs. To reduce the impact of tumor cell proportion on the microenvironment and treatment effect on prognosis, we matched these features in two groups of AITL cases.

To compare TME functions between the two AITL groups, TIL subset proportion, TRB and IGH repertoires, and gene expression profiles were examined. TIL subsets and proportions directly reflected TME immune function and have been shown to be predictive of various malignancies (17–19). TR and IGH repertoires of TME dictate TIL-T and TIL-B variety and represent the ability to identify tumor antigens and combat tumor immunoevasion (13, 14). Furthermore, gene expression profiling may reveal immunological activity by examining the regulation of related pathways. Our findings revealed that CD8-predominant AITL differs significantly from common AITL, which appears to have an immunosuppressive TME.

Although CD8-predominant AITL shared histopathological characteristics with common AITL in diffuse growth patterns and arborizing HEVs, it showed an increase in eosinophil counts. High eosinophil counts have been reported in AITL and have been described to be associated with cytotoxic agents, which may partly imply the cause of our findings (20). Meanwhile, CD8-predominant AITL showed in CD8+ TIL-Ts and a decrease in both CD4+TIL-Ts and TIL-Bs, implying a dysimmune TME. Despite the increase of CD8+TIL-Ts in CD8-predominant AITL, single-cell transcriptome analysis of AITL TME revealed that CD8+TIL-Ts expressed CD45RO, CD27, PD1, and TIGIT, indicating an exhausted phenotype (21). Therefore, CD8-predominant AITL may have reduced cytotoxicity in the TME. Furthermore, CD4+TIL-Ts are required for CD8+ TIL-T activation, and their depletion may result in CD8+ TIL-T impotence (22, 23). Moreover, CD4+TIL-Ts exert anti-tumor effects by recognizing tumor antigens and releasing cytokines, which activate other tumoricidal immune cells (24). Therefore, the low level of CD4+TIL-Ts in CD8-predominant AITL also indicated that anti-tumor immunity was

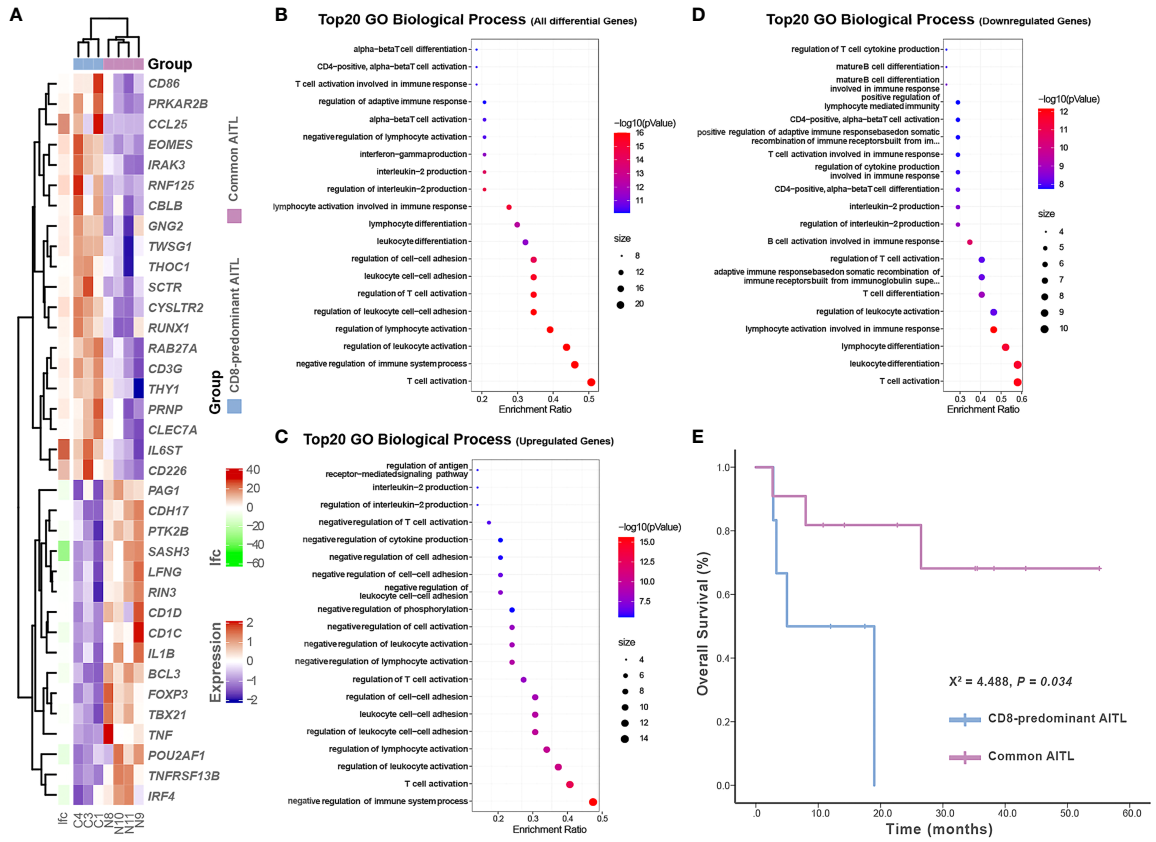


FIGURE 5
Gene expression profiles and survival analysis in CD8-predominant AITLs and matched common AITLs. Heatmap analysis showed differences in inflammation and immune response-related gene expression between CD8-predominant AITLs and common AITLs **(A)**. The top 20 enriched biological processes in gene oncology from all differential genes **(B)**, upregulated differential genes **(C)**, and downregulated differential genes **(D)**. Kaplan–Meier estimates of overall survival rate for CD8-predominant AITL and matched patients with common AITL **(E)**.

downregulated. Additionally, neoplastic T cells in AITL still preserve the function of TFH cells that secrete CXCL13 and other cytokines to promote B cell recruitment, expansion, and activation, generally resulting in a B-cell-rich TME (6, 7, 25, 26). Moreover, our previous study found that the expanded B-cells represent an efficient humoral response and enhance cellular immunity through T-cell and B-cell crosstalk in the TME, leading to good survival (9, 27). As a result, the lower TIL-Bs in CD8-predominant AITL implied suppressed humoral response and immune dysfunction in the TME.

CD8-predominant AITLs displayed narrowed TRB and IGH repertoires in TME, showing a significant drop in productive clones and increased clonality score in the TRB repertoire, a decrease in entropy, and an increase in the proportion of the top 10 clones in the IGH repertoire, which also indicated the immunosuppressive TME and anti-tumor immunity impairment. TR and IGH repertoires, the essential determinants of the TME in both solid tumors and hematological malignancies, reflect the immune activity of the TME (12, 15). Previous investigations have shown that deteriorated TR repertoire metrics could be caused by immunosuppression due to aging or inborn errors (28–31). Accordingly, our findings in the TRB repertoire of the CD8-predominant AITL TME may be interpreted by TME immunosuppression. Additionally, in keeping with knowledge from other tumors (13, 32), the narrowed TR and IGH repertoires in the TME of CD8-predominant AITL may indicate downregulated anti-tumor immunity *via* inadequate immunosurveillance of tumor T-cell neoantigens.

The gene expression profile further supported the findings of flow cytometry analysis and TRB and IGH repertoire sequencing. RNA sequencing data were analyzed focusing on genes related to inflammation and immune response, revealing the increased negative regulation of immune system processes and decreased T-cell activation and immune cell differentiation compared to common AITL. These results directly demonstrate the immunosuppressive TME in CD8-predominant AITL.

Neoplastic T-cells in AITL shape the TME in terms of histological structure and immune pattern by secreting

cytokines and producing tumor antigens (7, 33). Therefore, TME similarity is closely related to tumor cell similarity. Neoplastic AITL cells retain TFH cell functions and harbor a recurrent RHOA^{G17V} mutation (6). Furthermore, in our previous study, AITL tumor cells were found to have a biased usage of TRBV19-1 and a high level of identity in the CDR3 sequence (34). Thus, these commonalities of AITL tumor cells could explain our findings: CD8-predominant AITLs and common AITLs had similar histological structures and shared CDR3 sequences in the TME TRB and IGH repertoires. Neoplastic cell heterogeneity is associated with differences in the TME. We found that CD8-predominant AITLs and common AITLs had different V family segment usage and CDR3 sequence frequency in the TME of both TRB and IGH repertoires. Consistent with the literature on solid tumors and hematopoietic malignancies, these findings reflect the heterogeneity of the tumor antigen portfolio and further imply a heterogeneous mutational landscape of neoplastic cells between the two AITL groups (35–37). Thus, heterogeneity of neoplastic T-cells may be the cause of TME immune function differences between the two AITL groups, resulting in CD8-predominant AITL having an immunosuppressive TME.

Studies from both lymphoma and other solid tumors demonstrated that the TME changes as tumor progresses; in general, the immune response of the TME gradually decreases and eventually leads to immune silence (38, 39). Our results demonstrated that CD8-predominant AITL has an immunosuppressive TME, indicating that CD8-predominant AITL is at a later stage of tumor development and thus has significantly inferior survival compared to common AITLs. Combined with the patient's clinical manifestations, we hypothesized that the immunosuppressed TME of CD8-predominant AITL leads to insufficient anti-tumor immunity, manifesting as an increase in tumor proliferation index, which in turn leads to tumor dissemination (40), causing aggravation of the patient's clinical manifestations such as frequent effusion and edema. Furthermore, the patients' immune system collapses secondary to tumor development, resulting in EBV reactivation and increased plasma EBV-DNA (41). This eventually leads to a reduction in the patient's survival (Figure 6). Therefore, future research should concentrate on enhancing the immune function of the TME of CD8-predominant AITL, thereby improving patient prognosis.

This study has potential limitations. The main point is the small sample size, which is due to the rarity of CD8-predominant AITL. To reduce the confounding factors and sampling error, we introduced the case-control matching. Nevertheless, our findings must be validated through the accumulation of cases.

Conclusions

CD8-predominant AITL is an uncommon disease with more severe clinical manifestations and a shorter OS than that of common AITL. Although CD8-predominant AITL has histological features similar to those of common AITL, it has a lower TIL-B proportion, an inverted CD4+/CD8+ TIL-Ts ratio, deterioration of TR and IGH repertoire metrics, and altered gene expression profiles, indicating the anti-tumor immunity

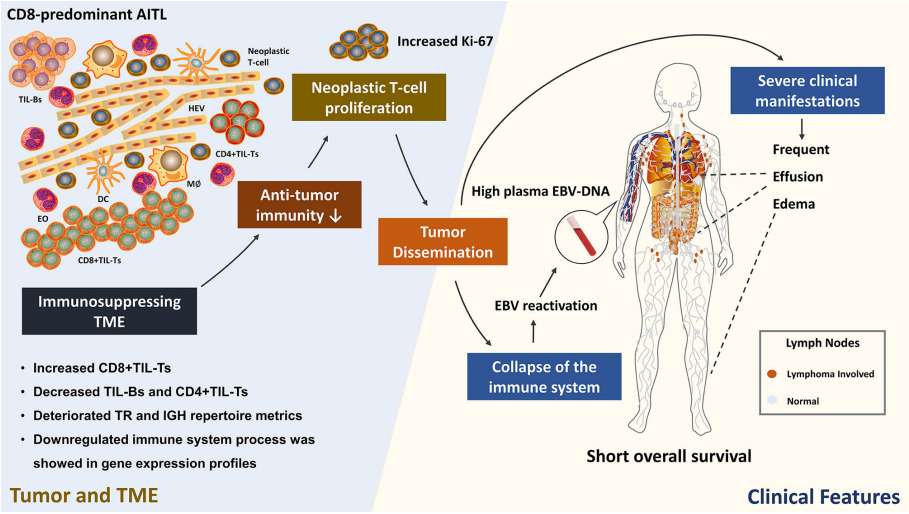


FIGURE 6
Hypothesized correlation between CD8-predominant AITL TME immune function and clinicopathological features. TIL, tumor infiltrating lymphocyte; HEV, high endothelial venule; EO, eosinophil; DC, dendritic cell; MØ, macrophage. This figure was drawn using ScienceSlides 2016, VisiScience, Inc.

impairment in TME. According to the accumulated evidence, CD8-predominant AITL is a distinct immune pattern of AITL with an immunosuppressive TME that must be better identified and investigated to improve patient survival.

Data availability statement

TRB and IGH sequencing data have been deposited in the National Center for Biotechnology Information Sequence Read Archive under accession number PRJNA887199. The RNA-seq data were provided in the Supplementary Material.

Ethics statement

The studies involving human participants were reviewed and approved by Ethics Committee on Biomedical Research, West China Hospital of Sichuan University. Written informed consent for participation was not required for this study in accordance with the national legislation and the institutional requirements.

Author contributions

ZC, YT, and SZ designed and conceived the study. ZC, QZ, WZ, WL, and SZ selected samples. ZC, QZ, and XD performed experiments. ZC, QZ, and WY analyzed the data. ZC wrote the manuscript. All authors contributed to the article and approved the submitted version.

Funding

This study was supported by the National Natural Science Foundation of China (81900195, 30900534), 1·3·5 project for disciplines of excellence–Clinical Research Incubation Project, West China Hospital, Sichuan University (2021HXFH027), and Post-Doctor Research Project, West China Hospital, Sichuan University (2019HXBH067).

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fimmu.2022.987227/full#supplementary-material

References

1. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, et al. The 2016 revision of the world health organization classification of lymphoid neoplasms. *Blood* (2016) 127:2375–90. doi: 10.1182/blood-2016-01-643569

2. Yang QP, Zhang WY, Yu JB, Zhao S, Xu H, Wang WY, et al. Subtype distribution of lymphomas in southwest China: analysis of 6,382 cases using WHO classification in a single institution. *Diagn Pathol* (2011) 6:77. doi: 10.1186/1746-1596-6-77

3. Lunning MA, Vose JM. Angioimmunoblastic T-cell lymphoma: the many-faced lymphoma. *Blood* (2017) 129:1095–102. doi: 10.1182/blood-2016-09-692541

4. Crickx E, Poullot E, Moulis G, Goulabchand R, Fieschi C, Galicier L, et al. Clinical spectrum, evolution, and management of autoimmune cytopenias associated with angioimmunoblastic T-cell lymphoma. *Eur J Haematol* (2019) 103:35–42. doi: 10.1111/ejh.13239

5. Advani RH, Skrypets T, Civallero M, Spinner MA, Manni M, Kim WS, et al. Outcomes and prognostic factors in angioimmunoblastic T-cell lymphoma: final report from the international T-cell project. *Blood* (2021) 138:213–20. doi: 10.1182/blood.2020010387

6. Chiba S, Sakata-Yanagimoto M. Advances in understanding of angioimmunoblastic T-cell lymphoma. *Leukemia* (2020) 34:2592–606. doi: 10.1038/s41375-020-0990-y

7. Boulland ML, Kanavaros P, Wechsler J, Casiraghi O, Gaulard P. Cytotoxic protein expression in natural killer cell lymphomas and in alpha beta and gamma delta peripheral T-cell lymphomas. *J Pathol* (1997) 183:432–9. doi: 10.1002/(SICI)1096-9896(199712)183:4<432::AID-PATH942>3.0.CO;2-4

8. Gaulard P, de Leval L. The microenvironment in T-cell lymphomas: Emerging themes. *Semin Cancer Biol* (2014) 24:49–60. doi: 10.1016/j.semcancer.2013.11.004

9. Zhu Q, Deng X, Yao W, Chen Z, Ye Y, Gao L, et al. Novel tumour-infiltrating lymphocyte-related risk stratification based by flow cytometry for patients with *de novo* angioimmunoblastic T cell lymphoma. *Ann Hematol* (2021) 100:715–23. doi: 10.1007/s00277-020-04389-5

10. Chen Z, Deng X, Ye Y, Gao L, Zhang W, Liu W, et al. Novel risk stratification of *de novo* diffuse large b cell lymphoma based on tumour-infiltrating T lymphocytes evaluated by flow cytometry. *Ann Hematol* (2011) 98:391–9. doi: 10.1007/s00277-018-3534-z

11. Chen Z, Deng X, Ye Y, Zhang W, Liu W, Zhao S. Flow cytometry-assessed PD1/PDL1 status in tumor-infiltrating lymphocytes: A link with the prognosis of diffuse Large b-cell lymphoma. *Front Oncol* (2021) 11:687911. doi: 10.3389/fonc.2021.687911

12. Keane C, Gould C, Jones K, Hamm D, Talaulikar D, Ellis J, et al. The T-cell receptor repertoire influences the tumor microenvironment and is associated with survival in aggressive b-cell lymphoma. *Clin Cancer Res* (2017) 23:1820–8. doi: 10.1158/1078-0432.CCR-16-1576

13. Olschewski V, Witte HM, Bernard V, Steinestel K, Peter W, Merz H, et al. Systemic inflammation and tumour-infiltrating T-cell receptor repertoire diversity are predictive of clinical outcome in high-grade b-cell lymphoma with MYC and BCL2 and/or BCL6 rearrangements. *Cancers (Basel)* (2021) 13(4):887. doi: 10.3390/cancers13040887

14. Rieken J, Bernard V, Witte HM, Peter W, Merz H, Olschewski V, et al. Exhaustion of tumour-infiltrating T-cell receptor repertoire diversity is an age-dependent indicator of immunological fitness independently predictive of clinical outcome in burkitt lymphoma. *Br J Haematol* (2021) 193:138–49. doi: 10.1111/bjh.17083

15. Wu L, Zhu J, Rudqvist NP, Welsh J, Lee P, Liao Z, et al. T-Cell receptor profiling and prognosis after stereotactic body radiation therapy for stage I non-Small-Cell lung cancer. *Front Immunol* (2021) 12:719285. doi: 10.3389/fimmu.2021.719285

16. Xie Y, Jaffe ES. How I diagnose angioimmunoblastic T-cell lymphoma. *Am J Clin Pathol* (2021) 156:1–14. doi: 10.1093/ajcp/aqab090

17. Xu-Monette ZY, Xiao M, Au Q, Padmanabhan R, Xu B, Hoe N, et al. Immune profiling and quantitative analysis decipher the clinical role of immune-checkpoint expression in the tumor immune microenvironment of DLBCL. *Cancer Immunol Res* (2019) 7:644–57. doi: 10.1158/2326-6066.CIR-18-0439

18. Shirasawa M, Yoshida T, Imabayashi T, Okuma K, Matsumoto Y, Masuda K, et al. Baseline PD-L1 expression and tumour-infiltrated lymphocyte status predict the efficacy of durvalumab consolidation therapy after chemoradiotherapy in unresectable locally advanced patients with non-small-cell lung cancer. *Eur J Cancer* (2021) 162:1–10. doi: 10.1016/j.ejca.2021.11.013

19. Loi S, Salgado R, Adams S, Pruneri G, Francis PA, Lacroix-Triki M, et al. Tumor infiltrating lymphocyte stratification of prognostic staging of early-stage triple negative breast cancer. *NPJ Breast Cancer* (2022) 8:3. doi: 10.1038/s41523-021-00362-1

20. Buchwalow I, Atiakshin D, Samoilova V, Boecker W, Tiemann M. Identification of autofluorescent cells in human angioimmunoblastic T-cell lymphoma. *Histochem Cell Biol* (2018) 149:169–77. doi: 10.1007/s00418-017-1624-y

21. Pritchett JC, Yang ZZ, Kim HJ, Villasboas JC, Tang X, Jalali S, et al. High-dimensional and single-cell transcriptome analysis of the tumor microenvironment in angioimmunoblastic T cell lymphoma (AITL). *Leukemia* (2022) 36:165–76. doi: 10.1038/s41375-021-01321-2

22. Huang H, Hao S, Li F, Ye Z, Yang J, Xiang J. CD4+ Th1 cells promote CD8+ Tc1 cell survival, memory response, tumor localization and therapy by targeted delivery of interleukin 2 *via* acquired pMHC I complexes. *Immunology* (2007) 120:148–59. doi: 10.1111/j.1365-2567.2006.02452.x

23. Gu-Trantien C, Loi S, Garaud S, Equeter C, Libin M, de Wind A, et al. $CD4^+$ follicular helper T cell infiltration predicts breast cancer survival. *J Clin Invest* (2013) 123:2873–92. doi: 10.1172/JCI67428

24. Schmidt M, Weyer-Elberich V, Hengstler JG, Heimes AS, Almstedt K, Gerhold-Ay A, et al. Prognostic impact of CD4-positive T cell subsets in early breast cancer: a study based on the FinHer trial patient population. *Breast Cancer Res* (2018) 20:15. doi: 10.1186/s13058-018-0942-x

25. Yu H, Shahsafaei A, Dorfman DM. Germinal-center T-helper-cell markers PD-1 and CXCL13 are both expressed by neoplastic cells in angioimmunoblastic T-cell lymphoma. *Am J Clin Pathol* (2009) 131:33–41. doi: 10.1309/AJCP62WRKERPXDRT

26. Witalis M, Chang J, Zhong MC, Bouklouch Y, Panneton V, Li J, et al. Progression of AITL-like tumors in mice is driven by tfh signature proteins and T-b cross talk. *Blood Adv* (2020) 4:868–79. doi: 10.1182/bloodadvances.2019001114

27. Bouaziz JD, Yanaba K, Venturi GM, Wang Y, Tisch RM, Poe JC, et al. Therapeutic b cell depletion impairs adaptive and autoreactive CD4+ T cell activation in mice. *Proc Natl Acad Sci USA* (2007) 104:20878–83. doi: 10.1073/pnas.0709205105

28. Roskin KM, Simchoni N, Liu Y, Lee JY, Seo K, Hoh RA, et al. IgH sequences in common variable immune deficiency reveal altered b cell development and selection. *Sci Transl Med* (2015) 7:302ra135. doi: 10.1126/scitranslmed.aab1216

29. Somekh I, Marquardt B, Liu Y, Rohlfs M, Hollizeck S, Karakukcu M, et al. Novel mutations in RASGRP1 are associated with immunodeficiency, immune dysregulation, and EBV-induced lymphoma. *J Clin Immunol* (2018) 38:699–710. doi: 10.1007/s10875-018-0533-8

30. Frizinsky S, Rechavi E, Barel O, Najeeb RH, Greenberger S, Lee YN, et al. Novel MALT1 mutation linked to immunodeficiency, immune dysregulation, and an abnormal T cell receptor repertoire. *J Clin Immunol* (2019) 39:401–13. doi: 10.1007/s10875-019-00629-0

31. Simnica D, Akyüz N, Schliffke S, Mohme M, V Wenserski L, Mährle T, et al. T Cell receptor next-generation sequencing reveals cancer-associated repertoire metrics and reconstitution after chemotherapy in patients with hematological and solid tumors. *Oncoimmunology* (2019) 8:e1644110. doi: 10.1080/2162402X.2019.1644110

32. Green MR, Kihira S, Liu CL, Nair RV, Salari R, Gentles AJ, et al. Mutations in early follicular lymphoma progenitors are associated with suppressed antigen presentation. *Proc Natl Acad Sci USA* (2015) 112:E1116–1125. doi: 10.1073/pnas.1501199112

33. Zhang H, Liu L, Zhang J, Chen J, Ye J, Shukla S, et al. Investigation of antigen-specific T-cell receptor clusters in human cancers. *Clin Cancer Res* (2020) 26:1359–71. doi: 10.1158/1078-0432.CCR-19-3249

34. Yao WQ, Wu F, Zhang W, Chuang SS, Thompson JS, Chen Z, et al. Angioimmunoblastic T-cell lymphoma contains multiple clonal T-cell populations derived from a common TET2 mutant progenitor cell. *J Pathol* (2020) 250:346–57. doi: 10.1002/path.5376

35. Reuben A, Gittelman R, Gao J, Zhang J, Yusko EC, Wu CJ, et al. TCR repertoire intratumor heterogeneity in localized lung adenocarcinomas: An association with predicted neoantigen heterogeneity and postsurgical recurrence. *Cancer Discovery* (2017) 7:1088–97. doi: 10.1158/2159-8290.CD-17-0256

36. Joshi K, de Massy MR, Ismail M, Reading JL, Uddin I, Woolston A, et al. Spatial heterogeneity of the T cell receptor repertoire reflects the mutational landscape in lung cancer. *Nat Med* (2019) 25:1549–59. doi: 10.1038/s41591-019-0592-2

37. Roudko V, Greenbaum B, Bhardwaj N. Computational prediction and validation of tumor-associated neoantigens. *Front Immunol* (2020) 11:27. doi: 10.3389/fimmu.2020.00027

38. Cho J, Kim SJ, Park WY, Kim J, Woo J, Kim G, et al. Immune subtyping of extranodal NK/T-cell lymphoma: a new biomarker and an immune shift during disease progression. *Mod Pathol* (2020) 33:603–15. doi: 10.1038/s41379-019-0392-8

39. Galon J, Bruni D. Tumor immunology and tumor evolution: Intertwined histories. *Immunity* (2020) 52:55–81. doi: 10.1016/j.immuni.2019.12.018

40. Ostrand-Rosenberg S, Sinha P, Danna EA, Miller S, Davis C, Dissanayake SK. Antagonists of tumor-specific immunity: tumor-induced immune suppression and host genes that co-opt the anti-tumor immune response. *Breast Dis* (2004) 20:127–35. doi: 10.3233/BD-2004-20113

41. Eladl AE, Satou A, Elsayed AA, Suzuki Y, Kato S, Asano N, et al. Clinicopathological study of 30 cases of peripheral T-cell lymphoma with Hodgkin and reed-sternberg-like b-cells from Japan. *Am J Surg Pathol* (2017) 41:506–16. doi: 10.1097/PAS.0000000000000813

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 20 December 2022
DOI 10.3389/fonc.2022.1099265

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Anamarija Perry,
University of Michigan, United States
Luis Carrillo,
University of Arkansas System,
United States

*CORRESPONDENCE
Carlos Murga-Zamalloa
catto@uic.edu

SPECIALTY SECTION
This article was submitted to Hematologic Malignancies, a section of the journal Frontiers in Oncology

RECEIVED 15 November 2022
ACCEPTED 05 December 2022
PUBLISHED 20 December 2022

CITATION
Murga-Zamalloa C and Inamdar K (2022) Classification and challenges in the histopathological diagnosis of peripheral T-cell lymphomas, emphasis on the WHO-HAEM5 updates.
Front. Oncol. 12:1099265.
doi: 10.3389/fonc.2022.1099265

Classification and challenges in the histopathological diagnosis of peripheral T-cell lymphomas, emphasis on the WHO-HAEM5 updates

Carlos Murga-Zamalloa[1]* and Kedar Inamdar[2]

[1]Department of Pathology, University of Illinois at Chicago, Chicago, IL, United States, [2]Department of Pathology, Henry Ford Hospital, Detroit, MI, United States

Mature T-cell lymphomas represent neoplastic expansions of T-cell lymphocytes with a post-thymic derivation. Most of these tumors feature aggressive clinical behavior and challenging histopathological diagnosis and classification. Novel findings in the genomic landscape of T-cell lymphomas are helping to improve the understanding of the biology and the molecular mechanisms that underly its clinical behavior. The most recent WHO-HAEM5 classification of hematolymphoid tumors introduced novel molecular and histopathological findings that will aid in the diagnostic classification of this group of neoplasms. The current review article summarizes the most relevant diagnostic features of peripheral T-cell lymphomas with an emphasis on the updates that are incorporated at the WHO-HAEM5.

KEYWORDS

T-cell lymphoma, WHO, classification, diagnosis, molecular diagnosis, biology, pathology

1 Introduction

Mature T and NK cell lymphomas represent approximately 5-10% of all non-Hodgkin lymphomas globally. With few exceptions, these groups of neoplasms are usually diagnosed at an advanced stage in adult patients. They feature an aggressive clinical course, with overall survival of 3 to 5 years after initial diagnosis (1–6). Currently, more than 20 different types of mature T-cell lymphomas (excluding primary cutaneous T-cell lymphomas) exist (Figure 1). The diagnosis is usually challenging due to the lack of specific molecular markers and overlapping morphological features.

The continuous discovery of the genomic landscape and mutational signatures of T-cell lymphomas is helping to identify novel molecular biomarkers that will improve the

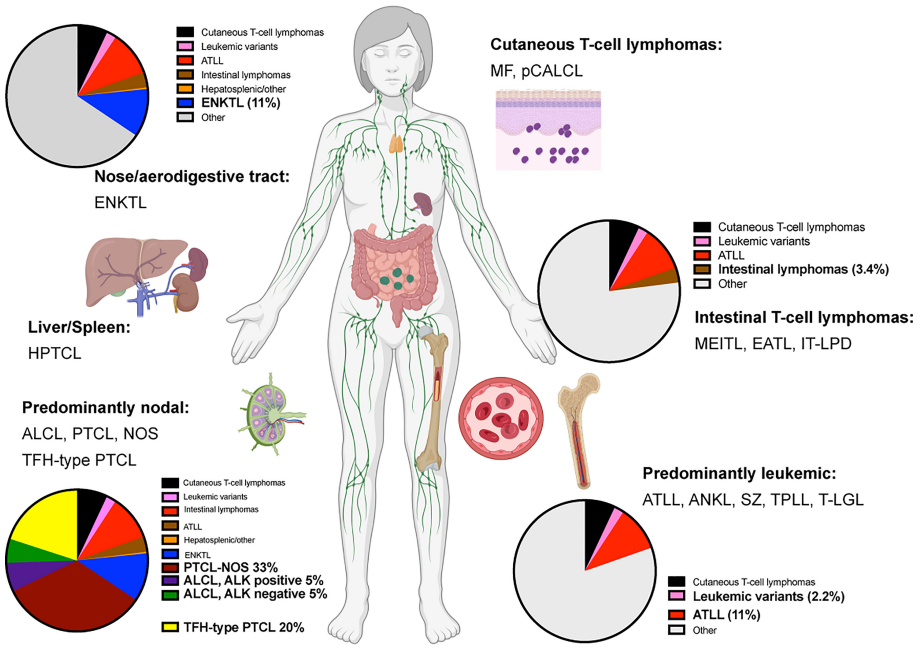


FIGURE 1
Predominant anatomical presentation and relative frequency of mature T-cell lymphomas. Abbreviations: Extranodal T/NK-cell lymphoma, nasal type (ENKTL); Hepatosplenic T-cell lymphoma (HPTCL); Anaplastic large cell lymphoma (ALCL); Peripheral T-cell lymphoma, non-otherwise specified (PTCL, NOS); T-follicular helper type peripheral T-cell lymphoma (TFH-PTCL); Mycosis Fungoides (MF); Primary cutaneous anaplastic large cell lymphoma (pCALCL); Monomorphic epitheliotropic intestinal T-cell lymphoma (MEITL), Enteropathy-associated T-cell lymphoma (EATL); Indolent T-cell lymphoproliferative disease of the gastrointestinal tract (IT-LPD); Adult T-cell leukemia/lymphoma (ATLL); Aggressive NK-cell leukemia (ANKL); Sèzary Syndrome (SZ); T-cell prolymphocytic lymphoma (T-PLL); T-cell large granular lymphoma (T-LGL).

classification, patient risk stratification and introduce novel tailored therapies. The current review focuses on the relevant pathological and molecular findings that were incorporated in the upcoming World Health Organization classification of Haematolymphoid Tumours (WHO-HAEM5) (7). The updated WHO-HAEM5 has introduced organizational changes that aid in the differential diagnosis of mature T-cell lymphomas with a leukemic presentation. Modifications in the terminology of entities such as indolent T-cell lymphoproliferative disease of the GI tract and peripheral T-cell lymphoma of T-follicular helper origin are also included. Finally, the WHO-HAEM5 included novel genomic findings to help recognize aggressive forms in entities such as T-cell large granular leukemia (T-LGL) and Adult T-cell leukemia/lymphoma (ATLL).

2 Mature T-cell and NK-cell leukemias

This section encompasses the mature T-cell neoplasms characterized by predominant leukemic presentation (Table 1). This section also included Sèzary Syndrome to emphasize the primary site of presentation.

2.1 T-cell prolymphocytic leukemia

Is characterized by the leukemic expansion of neoplastic post-thymic T-cell lymphocytes. More than 90% of the cases feature chromosomal abnormalities that involve the 14q32 or Xq28 loci associated with the increased cytoplasmic expression of TCL1A, TCL1B, or MTCP1. Immunophenotypically, the tumor cells are characterized by CD4 expression in 75% of the cases and positive expression of both CD4 and CD8 markers in 25% of the cases (8, 9). The WHO-HAEM5 incorporated

TABLE 1 List of disease entities with a predominant leukemic presentation.

Mature T-cell and NK-cell leukemias
T-cell prolymphocytic leukemia (T-PLL)
T-cell large granular lymphocytic leukemia
NK-large granular lymphocytic leukemia
Adult T-cell leukemia/lymphoma
Sezary Syndrome
Aggressive NK-cell leukemia

diagnostic recommendations from the T-PLL international study group (TPLL-ISG) assembled in 2017 (10). The proposed guidelines indicate that at least 2 (out of 3) major criteria must be met to establish a diagnosis of T-PLL (Table 2). The major criteria include defining T-PLL characteristics, including peripheral blood/bone marrow involvement, specific genetic aberrancies (*TCL1A/B* and *MTCP*), and T-cell clonality (Table 2). Importantly, less than 10% of the cases will feature characteristic clinical features of T-PLL, yet genetic aberrancies of *TCL1A*, *TCL1B*, or *MTCP* are not identified. This group of patients is recognized as TCL1-family negative T-PLL. In these scenarios, the proposed diagnostic criteria indicate that at least one chromosomal abnormality specific to T-PLL or the involvement of T-PLL-specific sites (spleen, effusions, skin, and CNS) must be present to establish a definitive diagnosis of T-PLL (1 minor criteria, Table 2).

2.2 T-cell large granular lymphocytic leukemia

Is defined by a persistent (at least 6 months) increase in the number of circulating large granular lymphocytes (2-20 x 10^9/L). About 40-50% of the patients will develop neutropenia, 30-40% anemias, and 10-25% splenomegaly. The updated WHO-HAEM5 incorporated novel molecular findings from two studies that established a correlation between the genomic landscape and clinical characteristics.

The first study (Sannikommu et al.) conducted a retrospective analysis of 224 patients with T-LGL (median follow-up of 36 months). It evaluated the clinical outcomes, therapy responses, and the mutational status of *STAT3* (11). In this study, mutations in *STAT3* were identified in 36% of the patients, and the most frequent change was Y640F-STAT3. The study demonstrated that patients with *STAT3* mutations are more likely to feature neutropenia and anemias. However, no differences in overall survival were identified when patients were stratified according to the mutational status of *STAT3*.

The second (Barila et al.) is a retrospective study that evaluated 169 patients diagnosed with T-LGL between 1992 and 2018 (12). *STAT3* mutations were identified in 60% of the patients with CD8+ T-LGL (n=65). However, mutations in *STAT3* were not detected in patients with CD4+/$CD8^{neg/dim}$ T-LGL. *STAT5b* mutations were detected in 34% of CD4+/$CD8^{neg/dim}$ T-LGL patients and not in CD8+ T-LGL patients. Correlation with clinical characteristics showed that patients with *STAT3* mutations were characterized by a higher frequency of neutropenia, anemia, and transfusion-dependent anemia. Multivariate analysis demonstrated that mutations in *STAT3* were independently associated with reduced overall survival (267 months vs. not-reached).

TABLE 2 Proposed diagnostic criteria for T-PLL by the T-PLL international study group (TPLL-ISG). *Adapted from Staber et al.* (10).

Diagnostic criteria for T-PLL
Major criteria
≥ 5 x 10^9/L cells of with a T-PLL phenotype in the peripheral blood or bone marrow
Evidence of T-cell clonality
Abnormalities of 14q32 or Xq28 or expression of TCL1A, TCL1B or MTCP1
Minor criteria
Abnormalities involving the chromosome 11 (11q22.3; ATM)
Abnormalities in chromosome 8: idic (8)(p11), t(8:8), trisomy 8q
Abnormalities in chromosome 5, del12p, 13, 22 or a complex karyotype
Involvement of a T-PLL specific site (splenomegaly, effusions, skin and CNS)

2.3 Chronic lymphoproliferative disorder of natural killer cells

The study by Barila et al. also included 36 patients with CLPD-NK (12). The clinical characteristics of CLPD-NK were like the T-LGL counterparts. Approximately 26% of the patients featured anemia, 48% neutropenia, and 13% splenomegaly. However, mutations in *STAT3* were identified only in 6% of the patients, and *STAT5b* mutations were not detected. Expression of CD94 was identified in 94% of the CLPD-NK cases. In contrast, CD94 expression was detected in 14% of Tα/β T-LGL and 52% of Tγ/δ T-LGL cases. Due to the clinical similarities with T-LGL, the WHO-HAEM5 has renamed this entity NK-large granular lymphocytic leukemia (NK-LGL).

2.4 Adult T-cell leukemia/lymphoma

Is an aggressive T-cell neoplasm caused by infection of the human T-cell leukemia virus type 1 (HTLV-1). Therefore, it is predominantly diagnosed in HTLV-1 endemic areas including Japan, Central America, South America, and intertropical Africa (13). The neoplastic lymphocytes show variable morphology and range from small to large forms, with irregular nuclear contours, 'flower-like cells,' and occasionally vacuolated cytoplasm (14). CD3/CD4 positive, mature T-cells characterize the immunophenotype with co-expression of CD25, FoxP3, and lack of CD7 expression (14). The most common presentation (acute type, ~60%) and the chronic variant (~15% of cases) are characterized by leukemic involvement with hepatosplenomegaly. Predominantly nodal and cutaneous manifestations occur in the smoldering and lymphomatous variants (~25% of cases). Those two variants are more frequent in geographical areas where HTLV-1 is nonendemic, and the diagnosis is usually problematic because those subtypes can mimic cutaneous T-cell lymphomas or, more rarely, anaplastic

large cell lymphoma (15). The prognosis is poor, regardless of the clinical presentation, and ranges from 1 year to 2 years after diagnosis. HTLV-1 serology testing is non-diagnostic of ATLL, and direct detection of viral transcripts in the neoplastic cells is critical for a definitive diagnosis in cases where the clinical and histopathological presentation is suggestive of different subtypes of T-cell lymphomas (15, 16).

The updated version of WHO-HAEM5 has incorporated novel genomic findings from two recent studies. The first study (Kogure et al.) identified recurrent loss-of-function mutations in the CIC-L/ATXN1 complex after performing whole-exome sequencing in 150 ATLL patients (17). The *CIC-L* gene encodes a transcriptional repressor that complexes with the ATXN1 protein. Mutations or structural variations in *CIC-L* and *ATXN1* were mutually exclusive in ATLL patients, and in combination, alterations in the ATXN1 and CIC-L complex were identified in 53% of the cases (17). To support the oncogenic role of CIC-L/ATXN1 during the neoplastic expansion of ATLL, murine models demonstrated that conditional deletion of *CIC-L* in CD4+ T-cell lymphocytes was associated with the proliferation of CD4+CD25+CD127-FoxP3+ regulatory T-cells.

A second study evaluated the clinical outcomes and the genetic landscape of 463 ATLL patients (18). Patients with aggressive forms of ATLL displayed a higher number of mutations. The 3-year overall survival rate was 54% for patients with 0 to 1 mutation, 39% for those with 2-5 mutations, and 19% for those with more than 6 mutations (18). Multivariate analysis demonstrated that older age (more than 70 years), *PRKCB* mutations, and *PD-L1* amplifications were independent prognostic factors for poor overall survival. Copy number amplifications of *PD-L1* were more frequent in the aggressive group and were predictive of poor outcomes in patients with indolent and aggressive forms of the disease. Among the patients with indolent clinical behavior, *IRF4* mutations, *PD-L1* amplifications, and *CDKN2A* deletions predicted poor outcomes (18). Overall, these findings highlight the relevance of including genomic profiling for the classification and prognosis of ATLL.

2.5 Aggressive NK-cell leukemia

Is a rare NK-cell neoplasm characterized by a fulminant clinical course with a median overall survival of 2 months or less after diagnosis. A leukemic presentation and occasional skin and CNS involvement characterize this entity. The WHO-HAEM5 included under this category the cases of NK/T cell lymphoma with an intravascular presentation that were previously considered a variant of extranodal NK/T-cell type lymphoma. In addition, novel clinical findings and mutational analysis are included under this category in the updated WHO-HAEM5.

A recent large retrospective evaluated the molecular profile and clinical features of 161 individuals diagnosed with ANKL in China (19). The peak incidence was 21 and 30 years old, with a median overall survival of 55 days. A group of the patients (16%) demonstrated a prolonged prodromal disease (subacute ANKL) that was characterized by fever, lymphocytosis, generalized lymphadenopathy, and hepatosplenomegaly (infectious mononucleosis-like symptoms). The prodromal phase has a median duration of 115 days, precedes the fulminant onset of ANKL, and the overall mean survival in this group of patients is 213 days. In contrast, patients without a prodromal phase ('classic' ANKL) have a median overall survival of 44 days. A targeted sequencing panel identified similar frequencies of mutations in the JAK-STAT pathway between the two groups. Importantly, mutations in *TP53* were detected in 38% of the 'classic' ANKL patients, and no mutations in *TP53* were identified in the subacute group (19).

A second study evaluated the genomic landscape of 10 ANKL patients utilizing whole-exome sequencing. The mutational spectrum of ANKL clustered separately from other related mature T-cell lymphomas with leukemic presentation (T-LGLL, T-PLL, and CPLD-NK) (20). The most recurrently mutated genes were *DDX3X* (29%) and *STAT3* (21%), and mutations in TP53 were detected in a single patient. The most frequent mutations in ANKL were also seen in NK-T-cell lymphoma, nasal type (NKTCL); therefore, no specific mutations were identified in the ANKL that can help distinguish those cases from NKTCL (20).

3 Intestinal T-cell lymphomas

Primary intestinal T-cell lymphomas constitute a diverse group of neoplasms predominantly derived from resident intestinal T-cell lymphocytes. Apart from NK-cell enteropathy, these neoplasms feature an aggressive clinical course and a poor prognosis. Within this group, the neoplastic proliferations that arise secondary to celiac disease are defined as enteropathy-associated T-cell lymphoma (EATL). EATL is an aggressive disease predominantly composed of CD8+ T-cells with the expression of TCR-α/β (21, 22). Monomorphic epitheliotropic intestinal T-cell lymphoma (MEITL) is more frequent in Asia and is composed of medium-sized lymphocytes with a homogeneous appearance, epitheliotropism, and frequent expression of CD56 (23–27) (Figure 2). The category of intestinal T-cell lymphomas non-otherwise specified (ITCL-NOS) remains as an entity that fails classification with the current schemes (Table 3; Figure 3), with very few case reports available (28–30). The WHO-HAEM5 has modified the designation of indolent-type T-cell

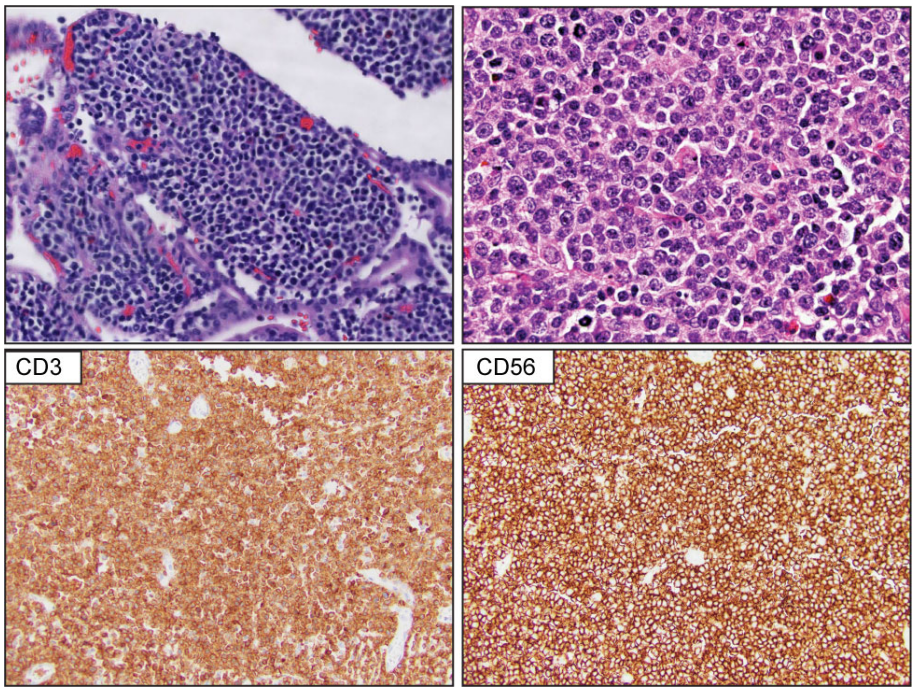


FIGURE 2
Monomorphic epitheliotropic intestinal T-cell lymphoma (MEITL). Upper panel show representative H&E images that highlight extensive epitheliotropism by medium size lymphocytes with a homogeneous appearance. The lower panel demonstrates that the atypical lymphocytes are composed of CD3-positive and CD56-positive T-cell lymphocytes.

lymphoproliferative disease of the GI tract and has incorporated novel molecular findings in NK-cell enteropathy.

3.1 Indolent T-cell lymphoproliferative disease of the gastrointestinal tract

Is morphologically characterized by superficial infiltrates composed of small lymphocytes with minimal cytological atypia that exhibit a Ki-67 proliferation index that is less than 10% (31). This entity is characterized by chronic and relapsing gastrointestinal symptoms, including diarrhea, dyspepsia, and vomiting (32, 33). In some cases, disease progression with an aggressive clinical course has been reported (34, 35). Therefore, the term 'lymphoproliferative' has been replaced by 'lymphoma' to highlight that this entity represents a clonal disease with a risk of transformation into more aggressive forms.

3.2 NK-cell enteropathy

Has been renamed indolent NK-cell lymphoproliferative disorder of the GI tract (iNKLPD). This entity is characterized by non-specific gastrointestinal symptoms, including abdominal pain, constipation, and reflux, with a chronic/relapsing course (36). The tumor cells feature mild cytological atypia, and the Ki-67 proliferation index is usually in the 25%-50% range; in contrast to ENKTCL, these tumors are negative for EBV expression. Additional case series reports have confirmed the indolent clinical course, with no reported cases of progression to more advanced stages (37). A recent study in 7 patients identified somatic mutations in *JAK3* in 30% of the patients; additional mutations in other genes, including *IFG1R* and *AURKB*, were also detected at lower frequencies (38). The same study demonstrated increased expression of phosphorylated STAT5 in 100% of cases (n = 7) (38), supporting the idea that this process constitutes a neoplastic lymphoproliferative disease.

4 Nodal T-cell lymphomas

Nodal involvement is the most frequent mode of presentation of T-cell lymphomas, and the three entities discussed below account for more than 60% of all mature T-cell neoplasms. Secondary nodal involvement by other T-cell lymphomas, such as ENKTL (discussed in 5.1) or cutaneous T-cell lymphomas, is not uncommon. However, their primary

TABLE 3 Histological and molecular features of intestinal T-cell lymphomas. *Adapted from Osmani et al.* (27).

	NK-cell lymphoproliferative disorder of the GI tract	Indolent T cell lymphoma of the GI tract	EATL	MEITL
Epidemiology	*Unknown*	*Unknown*	Northern Europe	Asian, Hispanic
Associations	*Unknown*	*Unknown*	Celiac, HLA-DQ2, HLA-DQ8	*Unknown*
Location	Stomach, small intestine, colon	Small intestine, colon, others	Small intestine	Small intestine
Histology	Medium to large in size, with mild cytological atypia. Destruction of adjacent glands may be present at advanced stages. Epitheliotropism is usually absent.	Small and monotonous lymphocytes, with none to mild cytological atypia. Non-destructive. Occasional epitheliotropism.	Pleomorphic. Medium to large with cytological atypia. Epitheliotropism is usually present. Angiodestruction may be present.	Monomorphic medium cells. Epitheliotropism usually present.
Phenotype	cCD3+* CD8- CD5- CD7 + CD4- CD56+ TIA1+ EBER(ish)- Granzyme B+ Low Ki-67(<25%). Increase levels of pSTAT5.	CD2+ CD3+ CD5+/- CD7+/- CD8+>CD4+*** CD56- TIA1-/+ Granzyme B- EBER(ish) - Low Ki-67 (<10%)	CD3+ CD4- CD5- CD7+ CD8-/+**** CD56- CD103+ CD30+/- TIA-1+, Granzyme B+ High Ki-67 (>50%)	CD3+ CD5- CD8+>CD4+ CD56 +** CD30- CD103+ TIA1+ Granzyme B+ EBER(ish)- High Ki-67 (>50%). MATK+#
TCR expression	Negative	Alpha beta (αβ)	Alpha beta (αβ) > gamma delta (γδ)	Gamma delta (γδ) > Alpha beta (αβ)
Molecular	TCR polyclonal. JAK3 mutations.	STAT3-JAK2 fusion	STAT5B, JAK3, GNAI2 Gains of 1q and 5q	SETD2, STAT5B, JAK3, GNAI2 Gains of MYC

Differential diagnosis to Intestinal T cell lymphomas. *Concurrent flow cytometry analysis demonstrates that CD3 expression is cytoplasmic. **Majority of tumors (~80%) are CD8+, a small subset of cases (9-18%) can be negative for CD56. *** Majority of the cases are CD8+ (~80%), a minority of the cases are either CD4+ or double CD4/CD8 negative in similar proportions. **** Approximately 30% of the cases are CD8+, majority of the cases are CD4-/CD8-. #MATK/Lsk nuclear expression is detected in majority of MEITL cases and in NK/T cell lymphomas, whereas EATL cases only feature cytoplasmic staining.

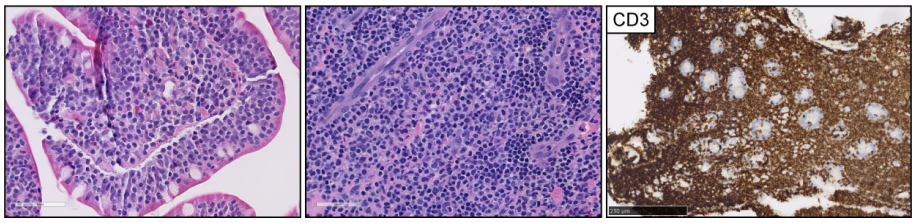


FIGURE 3
Intestinal T-cell lymphoma, non-otherwise specified (ITCL-NOS). H&E pictures demonstrate dense infiltrates of medium to large, atypical lymphocytes with epitheliotropism. The infiltrates were composed of CD3-positive T-cell lymphocytes with (not shown) aberrant CD7 and CD5 loss and negative expression of CD56.

presentation site is different from nodal, and the involvement of lymph nodes can occur during the evolution of the disease.

4.1 Anaplastic large cell lymphoma

ALCL is the third most common nodal T-cell lymphoma and accounts for approximately 11-13% of peripheral T-cell lymphomas (39, 40). Morphologically is characterized by large anaplastic cells, 'hallmark cells,' organized in a cohesive pattern, with uniform and strong expression of CD30 in more than 80% of the tumor cells (41) (Figure 4). Translocations involving the tyrosine kinase ALK are present in approximately 50% of ALCL cases. The uncontrolled activation of the ALK-kinase defines a phosphoproteomic and transcriptional signature that drives the oncogenic program of ALK+ ALCL cases (42–44), and ALK+ ALCL constitutes a specified entity. ALK-negative ALCL comprises a heterogenous group and is characterized by

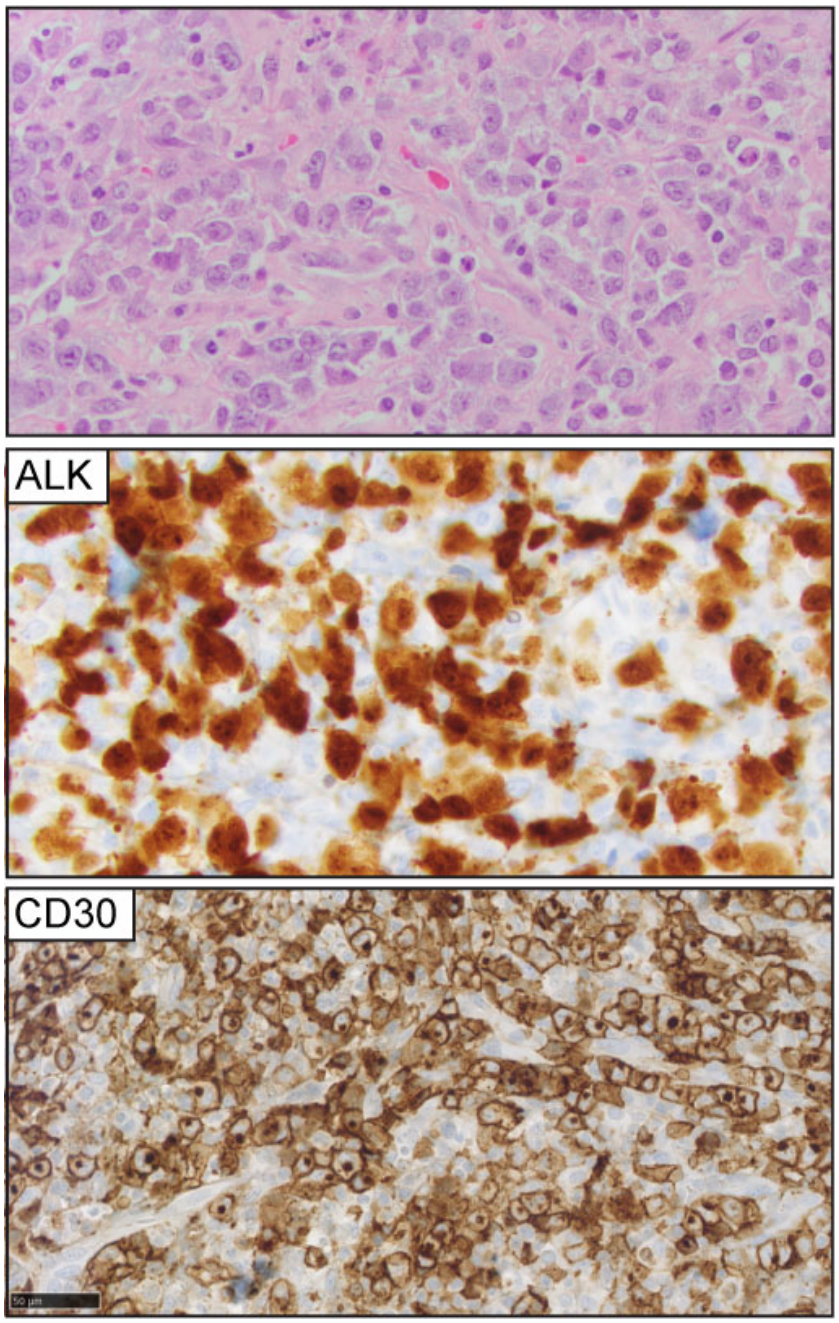


FIGURE 4
Anaplastic large cell lymphoma (ALCL). Representative (H&E) pictures of ALCL highlight the clustering of large, atypical lymphocytes with a variable amount of cytoplasm, open chromatin, and 'kidney-shaped' nuclei. ALK expression is detected in 50% of the cases (ALK+ ALCL). CD30 expression is homogeneously positive in at least 80% of the tumor cells.

inferior clinical outcomes compared to ALK+ ALCL. Specific genomic rearrangements within this group, including *DUSP22*, and *TP63* fusions, are associated with differential clinical outcomes (45–48). The WHO-HAEM5 has included novel genomic findings from two studies that will help understand the biology of ALCL.

Hapgood et al. (48) correlated the outcomes and molecular features of 62 ALK-negative ALCL patients. The findings demonstrated inferior clinical outcomes in patients with *DUSP22* rearrangements (n = 12, 44% five-year OS) in comparison to ≥80% 5-year overall survival (OS) that was described in the previous case series (47). These findings

indicate that additional molecular mechanisms can modify the outcomes of these patients. Consistent with this, a recent cohort study in 82 ALCL patients with systemic ALCL (ALK+ and ALK-) identified that mutations in *TP53* and *STAT3* are associated with a worse prognosis independent of ALK status (49).

The second study demonstrated the presence of specific super-enhancer (SE) regions enriched with *BATF3* sites in ALCL cell lines and patient samples. The findings further revealed that the transcriptional activity of *BATF3* can mediate the expression of IL2R. Importantly, increased expression of IL2R was predominantly detected in ALCL cases compared to different peripheral T-cell lymphomas, including AITL and PTCL-NOS. The findings indicate that chromatin remodeling may play a critical role during the growth of these groups of tumors and that IL2-dependent signaling plays a pivotal role in substituting T-cell receptor signaling in ALCL tumors (50).

Finally, the WHO-HAEM5 incorporated findings from a recent study by Fitzpatrick et al. that identified *JAK2* fusions in 6% of systemic CD30+ ALK-negative T-cell lymphomas (n=97) (51). Among the cases with JAK2 translocations, three fulfilled the diagnostic criteria of ALK-negative ALCL, and the other three were diagnosed as CD30+ PTCL-NOS. All the cases with *JAK2* translocations featured at least a proportion of ALCL-like large anaplastic tumor cells, in addition to Reed-Sternberg (RS)-like cells. Co-expression of CD30 and CD15 was present in the large anaplastic component in 80% of the cases. In all instances, the tumor cells were negative for PAX-5, and T-cell clonality was established. Due to the limited number of cases, a comparison of differential clinical outcomes was not possible (51). The presence of RS-like cells with co-expression of CD30 and CD15 can be a diagnostic pitfall, and recognition of this subset of cases is critical during diagnosis.

4.1.1 Breast implant-associated ALCL

Although morphologically and immunophenotypically indistinguishable from ALK-negative ALCL, is a distinct clinical entity primarily associated with textured breast implants, generally associated with an indolent clinical course. It is a rare disease with an estimated risk of 1 case in 4000-30,000 women undergoing breast implant surgeries. The median interval from surgery to the development of BIA-ALCL is 8-11 years (52). Diagnosis is often made in the peri-implant effusion fluid as the initial specimen. Cytospin preparations from the fluid show large, pleomorphic tumor cells with prominent nucleoli, abundant vacuolated cytoplasm, and irregular cytoplasmic membranes. Hallmark cells seen in other ALCL types are frequently present. Capsulectomy specimens show varying degrees of infiltration by the tumor cells. In the early stage, they often line the capsule, whereas, in the most advanced stage, they infiltrate through the fibrous capsule to form a mass. Axillary lymph nodes can be involved in approximately 20-30% of cases (53). Capsular invasion, mass formation, and lymph node involvement are adverse prognostic factors (53, 54).

In the updated WHO-HAEM5, BIA-ALCL was upgraded to a definite entity based on its unique clinical, genomic, and molecular features. A comprehensive study of the genetic subtype of BIA-ALCL cases by Feldman et al. (55) shows that there is oncogenic activation of the *JAK-STAT3* signaling pathway caused by mutations of somatic mutations of *STAT3, JAK1*, and *JAK2*. Laurent et al. (56) also demonstrated that the genomic alterations in BIA-ALCL include loss-of-function modifications of the epigenetic modifiers *KMT2C, KMT2D, CHD2*, and *CREBBP*, in up to 74% of cases.

The WHO-HAEM5 highlighted the role of allergic inflammation mediated by the secretion of IL-13 in the pathogenesis of BIA-ALCL (57). The frequent presence of eosinophils and mast cells in the tumor microenvironment of BIA-ALCL further supports the role of allergic inflammation during the pathogenesis of the disease (58, 59).

Finally, WHO-HAEM5 incorporated the role of immune evasion during the disease progression of BIA-ALCL. Tabanelli et al. (60) evaluated PD-L1 and pSTAT expression and PD-L1 copy number alterations (CNAs) in a cohort of 9 BIA-ALCL cases. The findings indicated that 56% of BIA-ALCL overexpressed PD-L1, and 33% of cases harbored CNAs at 9p24.1. Consistent with this, the results also demonstrated variable proportions of PD1+ T-cells and PD-L1+ tumor-associated macrophages (TAMs) in both PD-L1+ and PD-L1-negative BIA-ALCLs, suggesting the presence of an active PD-1/PD-L1 axis (60).

4.2 Nodal T-follicular helper type PTCLs

Peripheral T-cell lymphomas derived from T-follicular helper (T-FH) lymphocytes encompass a spectrum of aggressive neoplasms with poor clinical outcomes (61). Two entities in this group are characterized by specific morphological features: angioimmunoblastic T-cell lymphoma (AITL) and Follicular T-cell lymphoma (F-TH). A third group comprises neoplastic expansions of T-follicular helper T-cell lymphocytes that do not fit the characteristic morphological patterns of AITL and T-FH lymphoma. This group was defined in the latest WHO-HAEMR4 as Nodal Peripheral T-cell lymphoma with TFH phenotype (PTCL-TFH).

AITL is the second most common type of nodal T-cell lymphoma and accounts for 15-20% of peripheral T-cell lymphomas. Morphologically is characterized by effacement of the nodal architecture, with an expansion of arborizing post-endothelial venules with clusters of neoplastic lymphocytes with clear cytoplasm (Figure 5). These areas characteristically feature an expansion of follicular dendritic cell meshworks. The diagnosis

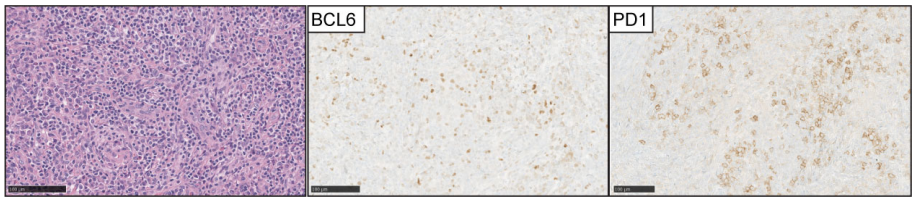


FIGURE 5
Angioimmunoblastic T-cell lymphoma (AITL). (Left panel) H&E images show characteristic branching of post-endothelial venules and adjacent lymphocytes with clear cell cytoplasm. Expression of BCL-6 and PD1 is detected in the neoplastic T-cell lymphocytes in the majority of cases.

can be difficult because the tumor cells represent a minority of the infiltrates, and the tumor microenvironment features positive RS-like cells for EBV with eosinophilic infiltrates (62).

4.2.1 Follicular T-cell lymphoma

Is morphologically distinct from AITL as it doesn't feature expanded post-endothelial venules surrounded by T-cell lymphocytes with clear cytoplasm. In contrast, two main architectural patterns are described; the most common pattern resembles progressive transformation of the germinal centers (PTGC-L), where the atypical lymphocytes are expanded in poorly defined nodules that are surrounded by numerous B-cell lymphocytes. The second pattern is characterized by well-demarcated nodular expansion of atypical lymphocytes, resembling B-cell follicular lymphoma. Consistent with a T-FH immunophenotype, the atypical cells are CD4 positive and feature co-expression of ICOS, PD1, CD10, BCL-6, and CXCL13; importantly, the expression of one or two of the T-FH markers may be absent. CD21 and CD23 highlight distorted follicular dendritic cell (FDC) meshwork's overlapping the atypical cells; in contrast to AITL, the FDCs are not expanded or associated with proliferating post-endothelial cells (63–66).

4.2.2 Nodal Peripheral T-cell lymphoma with follicular helper phenotype

Was a new disease entity introduced in WHO-HAEM4R. This group of cases does not feature the characteristic morphological features of AITL or F-TH lymphoma (Figure 6). However, the expression of at least two (preferentially 3) T-FH markers in addition to CD4 is required for classification. ICOS and PD-1 are expressed in most cases. However, ICOS is expressed in 43-52% of the PTCL-NOS cases, and PD-1 is expressed in approximately 60% of PTCL-NOS cases (67, 68). CD10, BCL6, and CXCL13 are the most specific markers and rarely are expressed in PTCL-NOS cases (68).

The family of peripheral T-cell lymphomas with a T-follicular helper (T-FH) origin share similar clinical features and are characterized by disseminated disease at diagnosis with a 5-year overall survival of 30-35% (61). The morphological features can transition between T-FH subtypes over subsequent biopsies or transform into PTCL-NOS. The genetic profile includes similar frequencies of mutations in *DNMT3A*, *RHOA*, and *TET2* (61, 69, 70), and *IDH2* mutations are predominantly detected in AITL. The gene expression profiles are similar, and these groups of neoplasms cluster in proximity

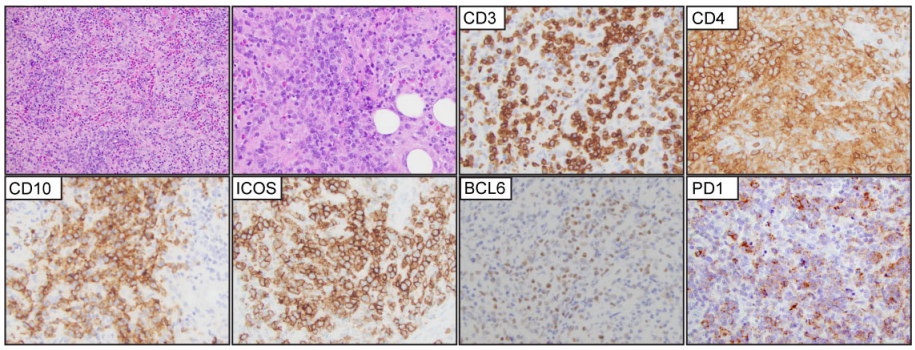


FIGURE 6
T-follicular helper type peripheral T-cell lymphoma (PTCL-TFH). Representative (H&E) images demonstrate effacement of the nodal architecture by medium to large, atypical lymphocytes that are organized in small clusters with numerous background histiocytes and eosinophils. The tumor cells are positive for CD3, CD4, CD10, ICOS, BCL6, and PD1.

and apart from other peripheral T-cell lymphomas, including ALCL and PTCL-NOS (69). However, recent studies show that a subset of PTCL-TFH cases features gene expression profiles closer to the PTCL-NOS group (69). To emphasize that this group of neoplasms constitutes a spectrum with morphological plasticity, this family of neoplasms has been designated nodal T-follicular helper lymphomas (nTFH) in the updated WHO-HAEM5. Therefore, the previous designations of 'angioimmunoblastic T-cell lymphoma,' 'follicular T-cell lymphoma,' and 'nodal peripheral T-cell lymphoma with TFH phenotype' will be renamed nTFHL angioimmunoblastic type (nTFHL-AI), nTFHL follicular-type (nTFHL-F) and nTFHL non-otherwise specified (nTFHL-NOS).

4.3 Peripheral T-cell lymphoma, non-otherwise specified

This group remains the most prevalent in the United States and constitutes a 'waste-basket' category when other disease entities are excluded from the current classification schemes. Recent study series that correlated clinicopathological features with genomic/molecular data has helped to identify distinct groups that are no longer included within this classification, such as Nodal EBV+ T/NK-cell lymphomas and Nodal T-follicular helper lymphomas, NOS.

Previous studies demonstrated that the expression of the transcription factor GATA-3 is enriched in a subset of PTCL-NOS cases with characteristic transcriptional profiles, worse overall survival, and resistance to chemotherapy (71–74). A second, albeit more heterogeneous subtype, highly expresses the transcription factor TBX21 and is similarly enriched for its gene targets. An immunohistochemistry algorithm that includes CD183, CCR4, GATA-3, and TBX21 has been proposed to identify these cases (75) (Figure 7). The upregulated expression of GATA-3 is secondary to the engagement of T-cell receptors in the neoplastic cells, and this is likely secondary to specific interactions with the tumor microenvironment (76–78). Due to insufficient clinicopathological and prognostic findings, the current WHO-HAEM5 did not recognize this group of tumors as a specific subtype of PTCL.

5 Epstein-Barr virus positive T-cell lymphomas

The contribution of EBV during the development of lymphoproliferative diseases is well-recognized (79). Importantly, EBV-infected B-cell or T-cells can be identified in the tumor microenvironment of several T-cell lymphomas, including AITL, ATLL, or PTCL-NOS. However, those are likely the result of dysregulated immunosurveillance derived from the primary lymphoma rather than a primary EBV-driven mechanism.

In the setting of immunosuppression, EBV+ T-cell lymphomas are not as common as their B-cell counterparts. However, T-cell lymphoproliferative disorders that occur secondary to EBV infection in immunocompetent hosts are characterized by markedly aggressive behavior and many of those feature dismal outcomes. This category includes T and NK-cell-derived lymphoproliferative disorders in the setting of chronic active EBV infection (T/NK type CAEBV). T/NK type CAEBV will feature EBV+ T or NK-cell expansions that do not feature definitive morphological or immunophenotypic features

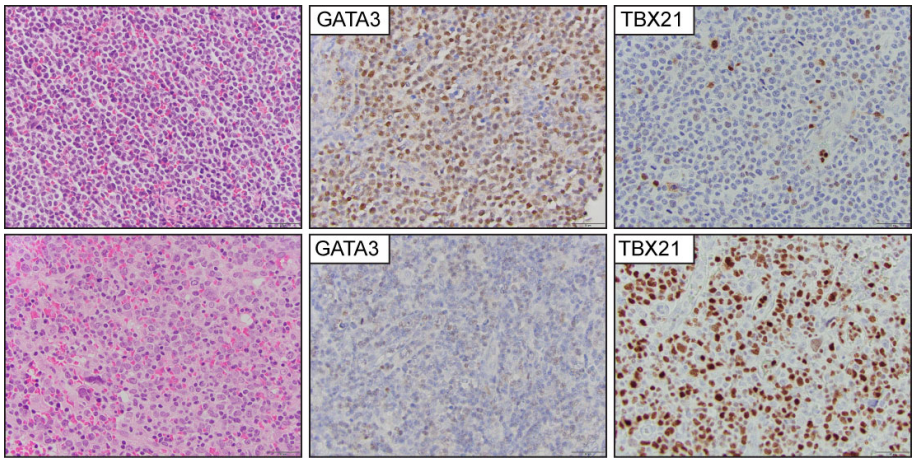


FIGURE 7
Peripheral T-cell lymphoma, non-otherwise specified (PTCL-NOS). Representative images from two PTCL-NOS cases. The upper panel shows a case classified in the GATA3 positive group, and (the lower panel) highlights a case within the TBX21 group.

compatible with a T or NK-cell type lymphoma diagnosis. However, the clinical course is very similar to T-cell lymphomas and is characterized by hepatitis, vasculitis, hemophagocytic syndrome, and end-organ damage (80–84). Aggressive NK-cell leukemia (ANKL) also features EBV+ lymphoma cells and is discussed in a different section (2.5). The WHO-HAEM5 has incorporated novel molecular findings in extranodal T/NK type lymphoma (ENKTL) and included nodal EBV+ T/NK-cell lymphoma as a new disease entity.

5.1 Extranodal NK/T-cell lymphoma, nasal type

ENKTL is an aggressive type of lymphoproliferative disorder predominantly derived from NK cells, with a minority of cases from cytotoxic T-cell origin. Approximately 80% of the cases are localized in the nasal cavity or nasopharynx (85). The remaining 20% of the cases will involve other sites that include skin, testicles, and salivary glands. Morphologically the neoplastic infiltrate can show a broad spectrum of appearances ranging from small cells with minimal atypia to medium to large pleomorphic cells with marked cellular atypia (86, 87) (Figure 8). Necrosis is observed in approximately 60%-80% of the cases, and angiocentricity is present in nearly 50% of the cases (85, 86, 88). Virtually all the cases are EBV+ in more than 80% of the tumor cells (85, 87, 88). An NK-cell immunophenotype is the most frequently observed and is characterized by negative expression of surface CD3, with positive expression of cytoplasmic CD3ε, CD56, and the cytotoxic markers TIA-1 and granzyme B. Approximately 20% of true NK-cell origin tumors (germline TCRγ) will feature negative expression of CD56, however, expression of cytotoxic markers is always present (87–89). Cases with a T-cell origin defined by monoclonal TCRγ rearrangement can account for 20%-40%, and approximately 40% of those will lack CD56 expression (85, 89). However, EBV negative cases with negative expression of CD56 should be designated as PTCL-NOS (87). In recognition that a subset of these cases involves extra-nodal sites other than the nasal cavity, the WHO-HAEM5 has removed the term 'nasal-type' from this designation. In addition, the WHO-HAEM5 mentioned novel genomic findings that highlight the relevance of *STAT3* mutations in ENKTL and the therapeutic role of PD-L1.

A recent study by Song et al. conducted targeted gene capture sequencing in 171 PTCLs, and *JAK/STAT* mutations were identified in 78% of the ENKTL cases (n=109) (90). Mutations in *STAT3* were predominantly detected in ENKTL cases (21%), in contrast to ALCL (3.7%) and PTCL-NOS cases (3.8%). The mutations identified in *STAT3* were associated with increased transcriptional activity and higher expression of PD-L1 in-vitro. However, the evaluation of primary ENKTL samples showed that 93% of the cases (n=30) featured increased expression of PD-L1, suggesting that additional molecular mechanisms are involved in PD-L1 expression (90). Consistent with this, Bi et al. demonstrated that the expression of the EBV latent membrane protein 1 (LMP1) promotes PD-L1 expression in ENKTL cell lines in-vitro (91).

Finally, the therapeutic potential of PD-L1 in ENKTL has been evaluated in a recent phase 2 clinical trial that tested the efficacy of the IgG1-PD-L1 antibody avelumab as a single agent in refractory/relapsed (R/R) ENKTL (92). This study by Jin et al. demonstrated a 24% complete response rate in R/R patients that received avelumab (92). The patient responses to avelumab were associated with higher expression of PD-L1 in the tumor cells. However, response rates were not related to the levels of serum soluble PD-L1 (92).

5.2 Nodal EBV-positive T and NK-lymphomas

This novel designation encompasses EBV+ NK/T cell lymphomas with a predominant nodal presentation. These tumors are characteristically diagnosed at an advanced stage and feature dismal clinical outcomes with overall survival of 1.5-3.5 months after diagnosis (93–95). This new entity differs from ENKTCL with secondary nodal involvement, as the former will

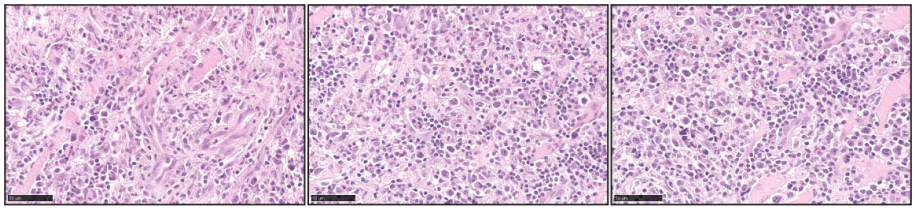


FIGURE 8
Extranodal T/NK-cell lymphoma, nasal type (ENKTL). Three representative pictures of ENTKL (H&E) highlight the pleomorphism of the atypical infiltrates. The atypical infiltrates can be predominantly composed of small lymphocytes or medium to large atypical forms. Angiocentricity is commonly identified.

primarily be present at extranodal sites. Cases of chronic active EBV infection of T-cell or NK-cell type (T/NK- type CAEBV) are also excluded from this category, as those do not feature morphological features consistent with a lymphoproliferative neoplasm. Finally, the presence of leukemic involvement by neoplastic cells with an NK-cell immunophenotype will be best classified as ANKL.

Other nodal T-cell lymphomas can feature a subset of tumor cells positive for EBV. Therefore, the minimal threshold for the number of EBV+ neoplastic cells varies among the published case series and ranges between 40%-50% (93–95). Immunophenotypically, approximately 60% of the cases will show tumor cells with a T-cell immunophenotype that are positive for CD8 and TβF1. The tumor cells will be less frequently positive for CD4 (93–95). Tumor cells with a characteristic NK-cell immunophenotype have been described. However, those account for less than 10% of the cases described (93, 94, 96).

This group of neoplasms features gene-expression profiles distinct from ENKTL with frequent loss of 14q11.2 and upregulation of PD-L1 (CD174) (93). Functional analysis of the gene-expression profiles demonstrates enrichment for NF-κB signaling (97). Consistent with this and in contrast to PTCL-NOS cases, predominant expression of BIRC3 and p50 is detected (97). Frequent mutations in *TET2* (64%, n=14), followed by *PIK3CD* (33%, n=14) and *STAT3* (19%, n=14) (97) characterize the genomic landscape of EBV+ NTNKCL.

6 Hepatosplenic T-cell lymphoma

Hepatosplenic T-cell lymphoma (HSTCL) is a rare but aggressive type of extranodal T-cell lymphoma that accounts for 1-2% of all peripheral T-cell lymphomas. The tumor cells involve the liver, spleen, and bone marrow in an exclusive intrasinusoidal distribution, often engorging and distending the cords and sinuses in the involved organs (39, 98). The morphology of the atypical infiltrates is characterized by monotonous, medium-sized lymphocytes with irregular nuclear contours, a moderate amount of agranular pale cytoplasm, and inconspicuous nucleoli. In a subset of the cases, the neoplastic cells feature blastoid chromatin, especially at advanced stages (99–101). Immunophenotypically, the neoplastic cells express pan T-cell markers CD2, CD3, and CD7, with frequent co-expression of CD56 and aberrant loss of CD5. A subset of cases is CD8+. However, the majority are negative for both CD4 and CD8. Expression of the cytotoxic granule-associated proteins, TIA1, and granzyme M is commonly observed. Expression of TCR-γ/δ is frequently observed (~75%), while ~25% of cases are TCR-α/β, and in a small number of instances (~5%), TCR-silent (102).

Several cytogenetic abnormalities are reported in HSTCL. The most common are isochromosome 7q [i(7q)] and trisomy 8 and are found in approximately 63% and 50% of cases, respectively (103). A unique molecular signature is identified in HSTCL cases. This is characterized by overexpression of genes encoding NK-cell-associated molecules (*FOS* and *VAV3*), the sphingosine-1-phosphatase receptor 5 (*S1PR5*) involved in cell trafficking, the tyrosine kinase *SYK*, with down-regulation of tumor suppressor gene *AIM1* (104).

While HSTCL predominantly occurs in immunocompetent individuals, 20-30% of cases occur in chronic immune suppression or immune dysregulation associated with autoimmune disorders (99, 100, 105). The WHO-HAEM4 reported that this disease is mainly diagnosed in adolescents and young adults. However, a recent report from the prospective T-cell lymphoma project identified that more than 50% of the patients in their cohort were older than 60 years old. Thus, the WHO-HAEM5 acknowledged that the disease is diagnosed in adolescents, young adults, and older individuals (98).

The WHO-HAEM5 also incorporated findings from a recent study by Mckinney et al. (106) that analyzed the genomic landscape of a large cohort of HSTCL cases with whole-exome sequencing. The results demonstrated frequent mutations in genes involving the JAK/STAT signaling pathway, with *STAT3* and *STAT5B* being the most frequently mutated. In addition, mutations in *PI3KCD* and the epigenetic regulators *SETD2*, *INO80*, *TET3*, and *SMARCA2* were observed in up to 62% of the cases (52, 106, 107).

7 Conclusions

The diagnosis and classification of T-cell lymphomas require a correlation between the histomorphology, molecular studies, and the site and manner of presentation. To date, few entities such as ALK+ ALCL or T-PLL display specific genetic aberrancies that are the predominant driving force of lymphoma progression and therefore constitute a distinctive feature for classification. In contrast, most T-cell lymphomas share morphological similarities and feature different frequencies of genetic alterations that either amplify the T-cell receptor signaling (e.g., NF-kB, RhoA, GATA3) or bypass it entirely (e.g., JAK/STAT, Pi3K). Therefore, a more significant proportion of mature T-cell lymphomas remain diagnosed in the unclassifiable (NOS) group. Currently, there is a limited understanding of the biology of T-cell neoplasms, which translates into limited successful therapeutic approaches, and these tumors remain an area of unmet need.

Author contributions

KI and CMZ wrote the manuscript. All the authors contributed to the article and approved the submitted version.

Funding

CMZ is supported by the American Society of Hematology/ Robert Wood Johnson Foundation (Harold Amos Medical Faculty Development Program).

Conflict of interest

The authors declare that the research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

The handling editor RW declared a past collaboration with the authors.

Publisher's note

References

1. Oluwasanjo A, Kartan S, Johnson W, Alpdogan O, Gru A, Mishra A, et al. Peripheral T-cell lymphoma, not otherwise specified (PTCL-NOS). *Cancer Treat Res* (2019) 176:83–98. doi: 10.1007/978-3-319-99716-2_4

2. Broccoli A, Zinzani PL. Peripheral T-cell lymphoma, not otherwise specified. *Blood* (2017) 129(9):1103–12. doi: 10.1182/blood-2016-08-692566

3. Vega F. Pathology and pathogenesis of T-cell lymphoma. *Clin Lymphoma Myeloma Leuk* (2020) 20 Suppl 1:S89–93. doi: 10.1016/S2152-2650(20)30474-2

4. Phan A, Veldman R, Lechowicz MJ. T-Cell lymphoma epidemiology: the known and unknown. *Curr Hematol Malig Rep* (2016) 11(6):492–503. doi: 10.1007/s11899-016-0353-y

5. Lansigan F, Horwitz SM, Pinter-Brown LC, Rosen ST, Pro B, Hsi ED, et al. Outcomes for relapsed and refractory peripheral T-cell lymphoma patients after front-line therapy from the COMPLETE registry. *Acta Haematol* (2020) 143 (1):40–50. doi: 10.1159/000500666

6. Armitage JO. The aggressive peripheral T-cell lymphomas: 2017. *Am J Hematol* (2017) 92(7):706–15. doi: 10.1002/ajh.24791

7. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: Lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

8. Herling M, Khoury JD, Washington LT, Duvic M, Keating MJ, Jones D. A systematic approach to diagnosis of mature T-cell leukemias reveals heterogeneity among WHO categories. *Blood* (2004) 104(2):328–35. doi: 10.1182/blood-2004-01-0002

9. Dearden C. How I treat prolymphocytic leukemia. *Blood* (2012) 120(3):538–51. doi: 10.1182/blood-2012-01-380139

10. Staber PB, Herling M, Bellido M, Jacobsen ED, Davids MS, Kadia TM, et al. Consensus criteria for diagnosis, staging, and treatment response assessment of T-cell prolymphocytic leukemia. *Blood* (2019) 134(14):1132–43. doi: 10.1182/blood.2019000402

11. Sanikommu SR, Clemente MJ, Chomczynski P, Afable MG, 2nd, Jerez A, Thota S, et al. Clinical features and treatment outcomes in large granular lymphocytic leukemia (LGLL). *Leuk Lymphoma* (2018) 59(2):416–22. doi: 10.1080/10428194.2017.1339880

12. Barila G, Teramo A, Calabretto G, Vicenzetto C, Gasparini VR, Pavan L, et al. Stat3 mutations impact on overall survival in large granular lymphocyte leukemia: a single-center experience of 205 patients. *Leukemia* (2020) 34(4):1116–24. doi: 10.1038/s41375-019-0644-0

13. Malpica L, Enriquez DJ, Castro DA, Pena C, Idrobo H, Fiad L, et al. Real-world data on adult T-cell Leukemia/Lymphoma in Latin America: A study from the grupo de estudio latinoamericano de linfoproliferativos. *JCO Glob Oncol* (2021) 7:1151–66. doi: 10.1200/GO.21.00084

14. Dahmoush L, Hijazi Y, Barnes E, Stetler-Stevenson M, Abati A. Adult T-cell leukemia/lymphoma: a cytopathologic, immunocytochemical, and flow cytometric study. *Cancer* (2002) 96(2):110–6. doi: 10.1002/cncr.10480

15. Khanlari M, Ramos JC, Sanchez SP, Cho-Vega JH, Amador A, Campuzano-Zuluaga G, et al. Adult T-cell leukemia/lymphoma can be indistinguishable from other more common T-cell lymphomas. the university of Miami experience with a large cohort of cases. *Mod Pathol* (2018) 31(7):1046–63. 10.1038/s41379-018-0037-3

16. Takatori M, Sakihama S, Miyara M, Imaizumi N, Miyagi T, Ohshiro K, et al. A new diagnostic algorithm using biopsy specimens in adult T-cell leukemia/lymphoma: combination of RNA *in situ* hybridization and quantitative PCR for HTLV-1. *Mod Pathol* (2021) 34(1):51–8. doi: 10.1038/s41379-020-0635-8

17. Kogure Y, Kameda T, Koya J, Yoshimitsu M, Nosaka K, Yasunaga JI, et al. Whole-genome landscape of adult T-cell leukemia/lymphoma. *Blood* (2022) 139 (7):967–82. doi: 10.1182/blood.2021013568

18. Kataoka K, Iwanaga M, Yasunaga JI, Nagata Y, Kitanaka A, Kameda T, et al. Prognostic relevance of integrated genetic profiling in adult T-cell leukemia/lymphoma. *Blood* (2018) 131(2):215–25. doi: 10.1182/blood-2017-01-761874

19. Tang YT, Wang D, Luo H, Xiao M, Zhou HS, Liu D, et al. Aggressive NK-cell leukemia: clinical subtypes, molecular features, and treatment outcomes. *Blood Cancer J* (2017) 7(12):660. doi: 10.1038/s41408-017-0021-z

20. Dufva O, Kankainen M, Kelkka T, Sekiguchi N, Awad SA, Eldfors S, et al. Aggressive natural killer-cell leukemia mutational landscape and drug profiling highlight JAK-STAT signaling as therapeutic target. *Nat Commun* (2018) 9 (1):1567. doi: 10.1038/s41467-018-03987-2

21. Di Sabatino A, Biagi F, Gobbi PG, Corazza GR. How I treat enteropathy-associated T-cell lymphoma. *Blood* (2012) 119(11):2458–68. doi: 10.1182/blood-2011-10-385559

22. Sieniawski M, Angamuthu N, Boyd K, Chasty R, Davies J, Forsyth P, et al. Evaluation of enteropathy-associated T-cell lymphoma comparing standard therapies with a novel regimen including autologous stem cell transplantation. *Blood* (2010) 115(18):3664–70. doi: 10.1182/blood-2009-07-231324

23. Tan SY, Ooi AS, Ang MK, Koh M, Wong JC, Dykema K, et al. Nuclear expression of MATK is a novel marker of type II enteropathy-associated T-cell lymphoma. *Leukemia* (2011) 25(3):555–7. doi: 10.1038/leu.2010.295

24. Tian S, Xiao SY, Chen Q, Liu H, Ping J. Monomorphic epitheliotropic intestinal T-cell lymphoma may mimic intestinal inflammatory disorders. *Int J Immunopathol Pharmacol* (2019) 33:2058738419829387. doi: 10.1177/2058738419829387

25. Tan SY, Chuang SS, Tang T, Tan L, Ko YH, Chuah KL, et al. (epitheliotropic intestinal T-cell lymphoma): A neoplasm of intra-epithelial T-cells with predominant CD8alphaalpha phenotype. *Leukemia* (2013) 27(8):1688–96. doi: 10.1038/leu.2013.41

26. Yi JH, Lee GW, Do YR, Jung HR, Hong JY, Yoon DH, et al. Multicenter retrospective analysis of the clinicopathologic features of monomorphic epitheliotropic intestinal T-cell lymphoma. *Ann Hematol* (2019) 98(11):2541–50. doi: 10.1007/s00277-019-03791-y

27. Osmani K, Shah E, Drumheller B, Webb S, Singh M, Rubinstein P, et al. CD30 + primary intestinal T-cell lymphoma (unclassified) masquerading as chronic inflammation: a case report. *Diagn Pathol* (2022) 17(1):53. doi: 10.1186/s13000-022-01237-0

28. Watanabe-Okochi N, Imai Y, Kimura H, Yamashita D, Hara S, Takahashi T. Intestinal T-cell lymphoma, NOS, presenting with sole peritoneal and mucosal lymphomatosis throughout abdominal cavity. *J Clin Exp Hematop* (2020) 60 (3):117–20. doi: 10.3960/jslrt.20020

29. Sun J, Lu Z, Yang D, Chen J. Primary intestinal T-cell and NK-cell lymphomas: A clinicopathological and molecular study from China focused on

type II enteropathy-associated T-cell lymphoma and primary intestinal NK-cell lymphoma. *Mod Pathol* (2011) 24(7):983–92. doi: 10.1038/modpathol.2011.45

30. Tang XF, Yang L, Duan S, Guo H, Guo QN. Intestinal T-cell and NK/T-cell lymphomas: A clinicopathological study of 27 Chinese patients. *Ann Diagn Pathol* (2018) 37:107–17. doi: 10.1016/j.anndiagpath.2018.10.004

31. Perry AM, Warnke RA, Hu Q, Gaulard P, Copie-Bergman C, Alkan S, et al. Indolent T-cell lymphoproliferative disease of the gastrointestinal tract. *Blood* (2013) 122(22):3599–606. doi: 10.1182/blood-2013-07-512830

32. van Vliet C, Spagnolo DV. T- and NK-cell lymphoproliferative disorders of the gastrointestinal tract: Review and update. *Pathology* (2020) 52(1):128–41. doi: 10.1016/j.pathol.2019.10.001

33. Matnani R, Ganapathi KA, Lewis SK, Green PH, Alobeid B, Bhagat G. Indolent T- and NK-cell lymphoproliferative disorders of the gastrointestinal tract: A review and update. *Hematol Oncol* (2017) 35(1):3–16. doi: 10.1002/hon.2317

34. Perry AM, Bailey NG, Bonnett M, Jaffe ES, Chan WC. Disease progression in a patient with indolent T-cell lymphoproliferative disease of the gastrointestinal tract. *Int J Surg Pathol* (2019) 27(1):102–7. doi: 10.1177/1066896918785985

35. Soderquist CR, Patel N, Murty VV, Betman S, Aggarwal N, Young KH, et al. Genetic and phenotypic characterization of indolent T-cell lymphoproliferative disorders of the gastrointestinal tract. *Haematologica* (2020) 105(7):1895–906. doi: 10.3324/haematol.2019.230961

36. Mansoor A, Pittaluga S, Beck PL, Wilson WH, Ferry JA, Jaffe ES. NK-cell enteropathy: a benign NK-cell lymphoproliferative disease mimicking intestinal lymphoma: clinicopathologic features and follow-up in a unique case series. *Blood* (2011) 117(5):1447–52. doi: 10.1182/blood-2010-08-302737

37. Xia D, Morgan EA, Berger D, Pinkus GS, Ferry JA, Zukerberg LR. NK-cell enteropathy and similar indolent lymphoproliferative disorders: A case series with literature review. *Am J Clin Pathol* (2019) 151(1):75–85. doi: 10.1093/ajcp/aqy108

38. Xiao W, Gupta GK, Yao J, Jang YJ, Xi L, Baik J, et al. Recurrent somatic JAK3 mutations in NK-cell enteropathy. *Blood* (2019) 134(12):986–91. doi: 10.1182/blood.2019001443

39. Vose J, Armitage J, Weisenburger D, International TCLP. International peripheral T-cell and natural killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26(25):4124–30. doi: 10.1200/JCO.2008.16.4558

40. Yoon SE, Song Y, Kim SJ, Yoon DH, Chen TY, Koh Y, et al. Comprehensive analysis of peripheral T-cell and natural killer/T-cell lymphoma in Asian patients: A multinational, multicenter, prospective registry study in Asia. *Lancet Reg Health West Pac* (2021) 10:100126. doi: 10.1016/j.lanwpc.2021.100126

41. Vega F, Medeiros LJ. A suggested immunohistochemical algorithm for the classification of T-cell lymphomas involving lymph nodes. *Hum Pathol* (2020) 102:104–16. doi: 10.1016/j.humpath.2020.05.006

42. Murga-Zamalloa CA, Mendoza-Reinoso V, Sahasrabuddhe AA, Rolland D, Hwang SR, McDonnell SR, et al. NPM-ALK phosphorylates WASp Y102 and contributes to oncogenesis of anaplastic large cell lymphoma. *Oncogene* (2017) 36(15):2085–94. doi: 10.1038/onc.2016.366

43. McDonnell SR, Hwang SR, Rolland D, Murga-Zamalloa C, Basrur V, Conlon KP, et al. Integrated phosphoproteomic and metabolomic profiling reveals NPM-ALK-mediated phosphorylation of PKM2 and metabolic reprogramming in anaplastic large cell lymphoma. *Blood* (2013) 122(6):958–68. doi: 10.1182/blood-2013-01-482026

44. Murga-Zamalloa C, Lim MS. ALK-driven tumors and targeted therapy: focus on crizotinib. *Pharmgenomics Pers Med* (2014) 7:87–94. doi: 10.2147/PGPM.S37504

45. Agnelli L, Mereu E, Pellegrino E, Limongi T, Kwee I, Bergaggio E, et al. Identification of a 3-gene model as a powerful diagnostic tool for the recognition of ALK-negative anaplastic large-cell lymphoma. *Blood* (2012) 120(6):1274–81. doi: 10.1182/blood-2012-01-405555

46. Parrilla Castellar ER, Jaffe ES, Said JW, Swerdlow SH, Ketterling RP, Knudson RA, et al. ALK-negative anaplastic large cell lymphoma is a genetically heterogeneous disease with widely disparate clinical outcomes. *Blood* (2014) 124(9):1473–80. doi: 10.1182/blood-2014-04-571091

47. Pedersen MB, Hamilton-Dutoit SJ, Bendix K, Ketterling RP, Bedroske PP, Luoma IM, et al. DUSP22 and TP63 rearrangements predict outcome of ALK-negative anaplastic large cell lymphoma: A Danish cohort study. *Blood* (2017) 130(4):554–7. doi: 10.1182/blood-2016-12-755496

48. Hapgood G, Ben-Neriah S, Mottok A, Lee DG, Robert K, Villa D, et al. Identification of high-risk DUSP22-rearranged ALK-negative anaplastic large cell lymphoma. *Br J Haematol* (2019) 186(3):e28–31. doi: 10.1111/bjh.15860

49. Lobello C, Tichy B, Bystry V, Radova L, Filip D, Mraz M, et al. STAT3 and TP53 mutations associate with poor prognosis in anaplastic large cell lymphoma. *Leukemia* (2021) 35(5):1500–5. doi: 10.1038/s41375-020-01093-1

50. Liang HC, Costanza M, Prutsch N, Zimmerman MW, Gurnhofer E, Montes-Mojarro IA, et al. Super-enhancer-based identification of a BATF3/IL-2R-module reveals vulnerabilities in anaplastic large cell lymphoma. *Nat Commun* (2021) 12(1):5577. doi: 10.1038/s41467-021-25379-9

51. Fitzpatrick MJ, Massoth LR, Marcus C, Vergilio JA, Severson E, Duncan D, et al. JAK2 rearrangements are a recurrent alteration in CD30+ systemic T-cell lymphomas with anaplastic morphology. *Am J Surg Pathol* (2021) 45(7):895–904. doi: 10.1097/PAS.0000000000001708

52. Kucuk C, Jiang B, Hu X, Zhang W, Chan JK, Xiao W, et al. Activating mutations of STAT5B and STAT3 in lymphomas derived from gammadelta-T or NK cells. *Nat Commun* (2015) 6:6025. doi: 10.1038/ncomms7025

53. Ferrufino-Schmidt MC, Medeiros LJ, Liu H, Clemens MW, Hunt KK, Laurent C, et al. Clinicopathologic features and prognostic impact of lymph node involvement in patients with breast implant-associated anaplastic Large cell lymphoma. *Am J Surg Pathol* (2018) 42(3):293–305. doi: 10.1097/PAS.0000000000000985

54. Clemens MW, Medeiros LJ, Butler CE, Hunt KK, Fanale MA, Horwitz S, et al. Complete surgical excision is essential for the management of patients with breast implant-associated anaplastic Large-cell lymphoma. *J Clin Oncol* (2016) 34(2):160–8. doi: 10.1200/JCO.2015.63.3412

55. Oishi N, Brody GS, Ketterling RP, Viswanatha DS, He R, Dasari S, et al. Genetic subtyping of breast implant-associated anaplastic large cell lymphoma. *Blood* (2018) 132(5):544–7. doi: 10.1182/blood-2017-12-821868

56. Laurent C, Nicolae A, Laurent C, Le Bras F, Haioun C, Fataccioli V, et al. Gene alterations in epigenetic modifiers and JAK-STAT signaling are frequent in breast implant-associated ALCL. *Blood* (2020) 135(5):360–70. doi: 10.1182/blood.2019001904

57. Kadin ME, Morgan J, Xu H, Epstein AL, Sieber D, Hubbard BA, et al. IL-13 is produced by tumor cells in breast implant-associated anaplastic large cell lymphoma: implications for pathogenesis. *Hum Pathol* (2018) 78:54–62. doi: 10.1016/j.humpath.2018.04.007

58. Chen CY, Lee JB, Liu B, Ohta S, Wang PY, Kartashov AV, et al. Induction of interleukin-9-Producing mucosal mast cells promotes susceptibility to IgE-mediated experimental food allergy. *Immunity* (2015) 43(4):788–802. doi: 10.1016/j.immuni.2015.08.020

59. Burd PR, Thompson WC, Max EE, Mills FC. Activated mast cells produce interleukin 13. *J Exp Med* (1995) 181(4):1373–80. doi: 10.1084/jem.181.4.1373

60. Tabanelli V, Corsini C, Fiori S, Agostinelli C, Calleri A, Orecchioni S, et al. Recurrent PDL1 expression and PDL1 (CD274) copy number alterations in breast implant-associated anaplastic large cell lymphomas. *Hum Pathol* (2019) 90:60–9. doi: 10.1016/j.humpath.2019.05.007

61. Dobay MP, Lemonnier F, Missiaglia E, Bastard C, Vallois D, Jais JP, et al. Integrative clinicopathological and molecular analyses of angioimmunoblastic T-cell lymphoma and other nodal lymphomas of follicular helper T-cell origin. *Haematologica* (2017) 102(4):e148–e51. doi: 10.3324/haematol.2016.158428

62. Xie Y, Jaffe ES. How I diagnose angioimmunoblastic T-cell lymphoma. *Am J Clin Pathol* (2021) 156(1):1–14. doi: 10.1093/ajcp/aqab090

63. Ruiz SJ, Cotta CV. Follicular helper T-cell lymphoma: a b-cell-rich variant of T-cell lymphoma. *Ann Diagn Pathol* (2015) 19(4):187–92. doi: 10.1016/j.anndiagpath.2015.03.008

64. Huang Y, Moreau A, Dupuis J, Streubel B, Petit B, Le Gouill S, et al. Peripheral T-cell lymphomas with a follicular growth pattern are derived from follicular helper T cells (TFH) and may show overlapping features with angioimmunoblastic T-cell lymphomas. *Am J Surg Pathol* (2009) 33(5):682–90. doi: 10.1097/PAS.0b013e3181971591

65. Kiiskila J, Uotila P, Haapasaari KM, Kauppila S, Teppo HR, Kuusisto MEL, et al. Incidence and clinicopathological features of follicular T-cell lymphoma in Finland: a population-based immunohistochemical study. *Hum Pathol* (2021) 117:79–87. doi: 10.1016/j.humpath.2021.07.012

66. Lisa M, Verma PK, Nishi, Shuchismita, Hussain MA. Follicular T-cell lymphoma. *J Cancer Res Ther* (2021) 17(6):1568–71. doi: 10.4103/jcrt.JCRT_486_19

67. Marafioti T, Paterson JC, Ballabio E, Chott A, Natkunam Y, Rodriguez-Justo M, et al. The inducible T-cell co-stimulator molecule is expressed on subsets of T cells and is a new marker of lymphomas of T follicular helper cell-derivation. *Haematologica* (2010) 95(3):432–9. doi: 10.3324/haematol.2009.010991

68. Basha BM, Bryant SC, Rech KL, Feldman AL, Vrana JA, Shi M, et al. Application of a 5 marker panel to the routine diagnosis of peripheral T-cell lymphoma with T-follicular helper phenotype. *Am J Surg Pathol* (2019) 43(9):1282–90. doi: 10.1097/PAS.0000000000001315

69. Amador C, Bouska A, Wright G, Weisenburger DD, Feldman AL, Greiner TC, et al. Gene expression signatures for the accurate diagnosis of peripheral T-cell lymphoma entities in the routine clinical practice. *J Clin Oncol* (2022), JCO2102707. doi: 10.1200/JCO.21.02707

70. Miyoshi H, Sakata-Yanagimoto M, Shimono J, Yoshida N, Hattori K, Arakawa F, et al. RHOA mutation in follicular T-cell lymphoma:

Clinicopathological analysis of 16 cases. *Pathol Int* (2020) 70(9):653–60. doi: 10.1111/pin.12981

71. Zhang W, Wang Z, Luo Y, Zhong D, Luo Y, Zhou D. GATA3 expression correlates with poor prognosis and tumor-associated macrophage infiltration in peripheral T cell lymphoma. *Oncotarget* (2016) 7(40):65284–94. doi: 10.18632/oncotarget.11673

72. Wang T, Lu Y, Polk A, Chowdhury P, Murga-Zamalloa C, Fujiwara H, et al. T-Cell receptor signaling activates an ITK/NF-kappaB/GATA-3 axis in T-cell lymphomas facilitating resistance to chemotherapy. *Clin Cancer Res* (2017) 23 (10):2506–15. doi: 10.1158/1078-0432.CCR-16-1996

73. Wang T, Feldman AL, Wada DA, Lu Y, Polk A, Briski R, et al. GATA-3 expression identifies a high-risk subset of PTCL, NOS with distinct molecular and clinical features. *Blood* (2014) 123(19):3007–15. doi: 10.1182/blood-2013-12-544809

74. Geng X, Wang C, Gao X, Chowdhury P, Weiss J, Villegas JA, et al. GATA-3 is a proto-oncogene in T-cell lymphoproliferative neoplasms. *Blood Cancer J* (2022) 12(11):149. doi: 10.1038/s41408-022-00745-y

75. Amador C, Greiner TC, Heavican TB, Smith LM, Galvis KT, Lone W, et al. Reproducing the molecular subclassification of peripheral T-cell lymphoma-NOS by immunohistochemistry. *Blood* (2019) 134(24):2159–70. doi: 10.1182/blood.2019000779

76. Murga-Zamalloa C, Wilcox RA. GATA-3 in T-cell lymphoproliferative disorders. *IUBMB Life* (2020) 72(1):170–7. doi: 10.1002/iub.2130

77. Wilcox RA. A three-signal model of T-cell lymphoma pathogenesis. *Am J Hematol* (2016) 91(1):113–22. doi: 10.1002/ajh.24203

78. Wilcox RA, Wada DA, Ziesmer SC, Elsawa SF, Comfere NI, Dietz AB, et al. Monocytes promote tumor cell survival in T-cell lymphoproliferative disorders and are impaired in their ability to differentiate into mature dendritic cells. *Blood* (2009) 114(14):2936–44. doi: 10.1182/blood-2009-05-220111

79. Montes-Mojarro IA, Fend F, Quintanilla-Martinez L. EBV and the pathogenesis of NK/T cell lymphoma. *Cancers (Basel)* (2021) 13(6). doi: 10.3390/cancers13061414

80. Suzuki K, Ohshima K, Karube K, Suzumiya J, Ohga S, Ishihara S, et al. Clinicopathological states of Epstein-Barr virus-associated T/NK-cell lymphoproliferative disorders (severe chronic active EBV infection) of children and young adults. *Int J Oncol* (2004) 24(5):1165–74. doi: 10.3892/ijo.24.5.1165

81. Okano M, Kawa K, Kimura H, Yachie A, Wakiguchi H, Maeda A, et al. Proposed guidelines for diagnosing chronic active Epstein-Barr virus infection. *Am J Hematol* (2005) 80(1):64–9. doi: 10.1002/ajh.20398

82. Montes-Mojarro IA, Kim WY, Fend F, Quintanilla-Martinez L. Epstein - Barr virus positive T and NK-cell lymphoproliferations: Morphological features and differential diagnosis. *Semin Diagn Pathol* (2020) 37(1):32–46. doi: 10.1053/j.semdp.2019.12.004

83. Bollard CM, Cohen JI. How I treat T-cell chronic active Epstein-Barr virus disease. *Blood* (2018) 131(26):2899–905. doi: 10.1182/blood-2018-03-785931

84. Yonese I, Sakashita C, Imadome KI, Kobayashi T, Yamamoto M, Sawada A, et al. Nationwide survey of systemic chronic active EBV infection in Japan in accordance with the new WHO classification. *Blood Adv* (2020) 4(13):2918–26. doi: 10.1182/bloodadvances.2020001451

85. Li S, Feng X, Li T, Zhang S, Zuo Z, Lin P, et al. Extranodal NK/T-cell lymphoma, nasal type: a report of 73 cases at MD Anderson cancer center. *Am J Surg Pathol* (2013) 37(1):14–23. doi: 10.1097/PAS.0b013e31826731b5

86. Kuo TT, Shih LY, Tsang NM. Nasal NK/T cell lymphoma in Taiwan: a clinicopathologic study of 22 cases, with analysis of histologic subtypes, Epstein-Barr virus LMP-1 gene association, and treatment modalities. *Int J Surg Pathol* (2004) 12(4):375–87. doi: 10.1177/106689690401200410

87. Ng SB, Lai KW, Murugaya S, Lee KM, Loong SL, Fook-Chong S, et al. Nasal-type extranodal natural killer/T-cell lymphomas: a clinicopathologic and genotypic study of 42 cases in Singapore. *Mod Pathol* (2004) 17(9):1097–107. doi: 10.1038/modpathol.3800157

88. Ko YH, Ree HJ, Kim WS, Choi WH, Moon WS, Kim SW. Clinicopathologic and genotypic study of extranodal nasal-type natural killer/T-cell lymphoma and natural killer precursor lymphoma among koreans. *Cancer* (2000) 89(10):2106–16. doi: 10.1002/1097-0142(20001115)89:10<2106::AID-CNCR11>3.0.CO;2-G

89. Pongpruttipan T, Sukpanichnant S, Assanasen T, Wannakrairot P, Boonsakan P, Kanoksil W, et al. Extranodal NK/T-cell lymphoma, nasal type, includes cases of natural killer cell and alphabeta, gammadelta, and alphabeta/gammadelta T-cell origin: a comprehensive clinicopathologic and phenotypic study. *Am J Surg Pathol* (2012) 36(4):481–99. doi: 10.1097/PAS.0b013e31824433d8

90. Song TL, Nairismagi ML, Laurensia Y, Lim JQ, Tan J, Li ZM, et al. Oncogenic activation of the STAT3 pathway drives PD-L1 expression in natural killer/T-cell lymphoma. *Blood* (2018) 132(11):1146–58. doi: 10.1182/blood-2018-01-829424

91. Bi XW, Wang H, Zhang WW, Wang JH, Liu WJ, Xia ZJ, et al. PD-L1 is upregulated by EBV-driven LMP1 through NF-kappaB pathway and correlates with poor prognosis in natural killer/T-cell lymphoma. *J Hematol Oncol* (2016) 9 (1):109. doi: 10.1186/s13045-016-0341-7

92. Kim SJ, Lim JQ, Laurensia Y, Cho J, Yoon SE, Lee JY, et al. Avelumab for the treatment of relapsed or refractory extranodal NK/T-cell lymphoma: an open-label phase 2 study. *Blood* (2020) 136(24):2754–63. doi: 10.1182/blood.2020007247

93. Ng SB, Chung TH, Kato S, Nakamura S, Takahashi E, Ko YH, et al. Epstein-Barr Virus-associated primary nodal T/NK-cell lymphoma shows a distinct molecular signature and copy number changes. *Haematologica* (2018) 103 (2):278–87. doi: 10.3324/haematol.2017.180430

94. Jung KS, Cho SH, Kim SJ, Ko YH, Kim WS. Clinical features and treatment outcome of Epstein-Barr virus-positive nodal T-cell lymphoma. *Int J Hematol* (2016) 104(5):591–5. doi: 10.1007/s12185-016-2068-1

95. Jeon YK, Kim JH, Sung JY, Han JH, Ko YH. Hematopathology study group of the Korean society of p. Epstein-Barr virus-positive nodal T/NK-cell lymphoma: an analysis of 15 cases with distinct clinicopathological features. *Hum Pathol* (2015) 46(7):981–90. doi: 10.1016/j.humpath.2015.03.002

96. Takahashi E, Asano N, Li C, Tanaka T, Shimada K, Shimada S, et al. Nodal T/NK-cell lymphoma of nasal type: a clinicopathological study of six cases. *Histopathology* (2008) 52(5):585–96. doi: 10.1111/j.1365-2559.2008.02997.x

97. Wai CMM, Chen S, Phyu T, Fan S, Leong SM, Zheng W, et al. Immune pathway upregulation and lower genomic instability distinguish EBV-positive nodal T/NK-cell lymphoma from ENKTL and PTCL-NOS. *Haematologica* (2022) 107(8):1864–79. doi: 10.3324/haematol.2021.280003

98. Foss FM, Horwitz SM, Civallero M, Bellei M, Marcheselli L, Kim WS, et al. Incidence and outcomes of rare T cell lymphomas from the T cell project: hepatosplenic, enteropathy associated and peripheral gamma delta T cell lymphomas. *Am J Hematol* (2020) 95(2):151–5. doi: 10.1002/ajh.25674

99. Belhadj K, Reyes F, Farcet JP, Tilly H, Bastard C, Angonin R, et al. Hepatosplenic gammadelta T-cell lymphoma is a rare clinicopathologic entity with poor outcome: Report on a series of 21 patients. *Blood* (2003) 102(13):4261–9. doi: 10.1182/blood-2003-05-1675

100. Wu H, Wasik MA, Przybylski G, Finan J, Haynes B, Moore H, et al. Hepatosplenic gamma-delta T-cell lymphoma as a late-onset posttransplant lymphoproliferative disorder in renal transplant recipients. *Am J Clin Pathol* (2000) 113(4):487–96. doi: 10.1309/YTTC-F55W-K9CP-EPX5

101. Vega F, Medeiros LJ, Bueso-Ramos C, Jones D, Lai R, Luthra R, et al. Hepatosplenic gamma/delta T-cell lymphoma in bone marrow. A sinusoidal neoplasm with blastic cytologic features. *Am J Clin Pathol* (2001) 116(3):410–9. doi: 10.1309/BM40-YM6J-9T3X-MH8H

102. Yabe M, Medeiros LJ, Tang G, Wang SA, Ahmed S, Nieto Y, et al. Prognostic factors of hepatosplenic T-cell lymphoma: Clinicopathologic study of 28 cases. *Am J Surg Pathol* (2016) 40(5):676–88. doi: 10.1097/PAS.0000000000000614

103. Weidmann E. Hepatosplenic T cell lymphoma. a review on 45 cases since the first report describing the disease as a distinct lymphoma entity in 1990. *Leukemia* (2000) 14(6):991–7. doi: 10.1038/sj.leu.2401784

104. Travert M, Huang Y, de Leval L, Martin-Garcia N, Delfau-Larue MH, Berger F, et al. Molecular features of hepatosplenic T-cell lymphoma unravels potential novel therapeutic targets. *Blood* (2012) 119(24):5795–806. doi: 10.1182/blood-2011-12-396150

105. Vega F, Medeiros LJ, Gaulard P. Hepatosplenic and other gammadelta T-cell lymphomas. *Am J Clin Pathol* (2007) 127(6):869–80. doi: 10.1309/LRKX8CE7GVPCR1FT

106. McKinney M, Moffitt AB, Gaulard P, Travert M, De Leval L, Nicolae A, et al. The genetic basis of hepatosplenic T-cell lymphoma. *Cancer Discovery* (2017) 7(4):369–79. doi: 10.1158/2159-8290.CD-16-0330

107. Nicolae A, Xi L, Pittaluga S, Abdullaev Z, Pack SD, Chen J, et al. Frequent STAT5B mutations in gammadelta hepatosplenic T-cell lymphomas. *Leukemia* (2014) 28(11):2244–8. doi: 10.1038/leu.2014.200

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 30 January 2023
DOI 10.3389/fonc.2023.1105651

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Carlos Murga-Zamalloa,
University of Illinois at Chicago, United States
Shannon Carty,
University of Michigan, United States

*CORRESPONDENCE
Mario L. Marques-Piubelli
MLPiubelli@mdanderson.org

†These authors have contributed equally to this work

SPECIALTY SECTION
This article was submitted to Hematologic Malignancies, a section of the journal Frontiers in Oncology

RECEIVED 22 November 2022
ACCEPTED 13 January 2023
PUBLISHED 30 January 2023

CITATION
Marques-Piubelli ML, Amador C and Vega F (2023) Pathologic and molecular insights in nodal T-follicular helper cell lymphomas. *Front. Oncol.* 13:1105651. doi: 10.3389/fonc.2023.1105651

Pathologic and molecular insights in nodal T-follicular helper cell lymphomas

Mario L. Marques-Piubelli[1*], Catalina Amador[2†] and Francisco Vega[3†]

[1]Department of Translational Molecular Pathology, The University of Texas MD Anderson Cancer Center, Houston, TX, United States, [2]Division of Hematopathology, Department of Pathology and Laboratory Medicine, University of Miami, Miami, FL, United States, [3]Department of Hematopathology, The University of Texas MD Anderson Cancer Center, Houston, TX, United States

T-follicular helper (TFH) cells are one of the T-cell subsets with a critical role in the regulation of germinal center (GC) reactions. TFH cells contribute to the positive selection of GC B-cells and promote plasma cell differentiation and antibody production. TFH cells express a unique phenotype characterized by *PD-1*^{hi}, *ICOS*^{hi}, *CD40L*^{hi}, *CD95*^{hi}, *CTLA*^{hi}, *CCR7*^{lo}, *and CXCR5*^{hi}. Three main subtypes of nodal TFH lymphomas have been described: 1) angioimmunoblastic-type, 2) follicular-type, and 3) not otherwise specified (NOS). The diagnosis of these neoplasms can be challenging, and it is rendered based on a combination of clinical, laboratory, histopathologic, immunophenotypic, and molecular findings. The markers most frequently used to identify a TFH immunophenotype in paraffin-embedded tissue sections include PD-1, CXCL13, CXCR5, ICOS, BCL6, and CD10. These neoplasms feature a characteristic and similar, but not identical, mutational landscape with mutations in epigenetic modifiers (*TET2*, *DNMT3A*, *IDH2*), *RHOA*, and T-cell receptor signaling genes. Here, we briefly review the biology of TFH cells and present a summary of the current pathologic, molecular, and genetic features of nodal lymphomas. We want to highlight the importance of performing a consistent panel of TFH immunostains and mutational studies in TCLs to identify TFH lymphomas.

KEYWORDS

peripheral T cell lymphomas, angioimmunoblastic T cell lymphoma, follicular T helper, next-generation sequencing, molecular and genetic profiling

Introduction

Nodal T-follicular helper (TFH) lymphomas represent a group of mature peripheral T-cell lymphomas (TCLs) with a gene expression profile and immunophenotype similar to that of nodal TFH cells (1, 2). This concept was initially described in angioimmunoblastic T-cell lymphoma (AITL) (3, 4) and more recently in a subset of peripheral T-cell lymphomas not otherwise specified (PTCL, NOS), which also has a gene expression profile, and immunophenotype suggestive of a TFH derivation (5, 6).

Here we present a brief summary of the biology of TFH cells and our current understanding of the clinicopathologic, molecular, and genetic features of nodal TFH lymphomas. Diagnostic considerations for this subgroup of lymphomas include distinguishing TFH lymphomas from other entities, particularly PTCL, NOS. For this, it is paramount to highlight the importance of performing a consistent set of at least 5 TFH immunomarkers and mutational studies in the work up of mature TCLs. Currently, we recommend using a five-marker panel that includes CD10, CXCL13, PD-1, ICOS and BCL6. This panel is used by other major academic centers in US and Europe. Per the WHO classification, the minimum criteria for assigning a TFH phenotype is 2 (but ideally 3 or more) markers in addition to CD4.

T-Follicular helper cells

The differentiation of naïve CD4+ T-cells through the stimulation of antigen-presenting cells (APC) is an essential step for the maintenance of adaptive immunity homeostasis. The CD4+ helper T (Th) subtype is essential for the effector functions of the T-cells, and it can be divided into four major subsets of effector cells that produce distinct cytokines to help in the recruitment and activation of different cell types. These subsets include Th1 and Th2 cells (for type 1 and type 2 helper T-cells, respectively), Th17 cells (due to their IL-17 signature), and follicular helper T (TFH) cells. The latter is a fundamental component of the germinal center (GC) reaction and B-cell specialization (7, 8). Other T-cell subsets include T regulatory cells (Treg), Th9, and Th22 (9).

Phenotypically, TFH cells are characterized by the expression of BCL6 and CXC chemokine receptors type 5 (CXCR5), which allow them to reach the GCs (10, 11). TFH cells also express the inducible T-cell costimulator (ICOS), the programmed cell death protein (PD-1), and the signaling lymphocytic activation molecule (SLAM)-associated protein (SAP) that contribute to the crosstalk between T- and B-cells. Inherited mutations in the *ICOS* gene cause some antibody deficiencies (12).

TFH cells also provide crucial signaling for the normal ontogeny of B-cells. Engagements of MHC class II, CD40, and ICOS-ligand on GC B-cells with the TCR, CD40L, and ICOS on TFH cells may generate the production of IL-21, CD40L, and IL-4 and support the formation and maintenance of GCs (13). These signals allow antigen-specific B-cells to survive, proliferate, undergo affinity maturation, and, ultimately, differentiate into memory B-cells or long-lived plasma cells.

BCL6 is the master transcriptional regulator in the differentiation of TFH cells. BLIMP1 is a transcription factor that antagonizes BCL6 function and prevents TFH differentiation (14). BCL-6 also inhibits transcription factors important to other T-cell subsets, such as T-bet, Gata 3, and RORyt needed for Th1, Th2, and Th17 differentiation, respectively (15–17). BCL-6 also prompts the upregulation of CXCR5, a required step for the relocation of these cells toward the GCs (18).

TET2-mediated demethylation of DNA at specific regulatory regions is required to balance the differentiation of CD4+ T-cells towards Th1 and TFH lineages. In the absence of TET2, CD4+ T-cell differentiation is skewed toward the generation of highly functional TFH cells (19). Note that *TET2* loss-of-function mutations are frequently found in follicular helper T-cell lymphomas (see below).

Although TFH cells are needed for the GC reaction and therefore are located inside the GCs, they can also be found in other compartments outside the GCs. Circulating memory TFH cells appear to derive from GC TFH cells following downregulation of BCL6, ICOS, and PD-1 and upregulation of CCR7 (20–22). An activated subset of circulating TFH cells with expression of P-D1 and ICOS but with low expression of CCR7 has also been described (23). These circulating activated subsets of TFH cells are functional and associated with autoimmune processes (23, 24). T-follicular regulatory (Tfr) cells are another subset of TFH cells that share many characteristics of TFH cells, including the expression of BCL6, CXCR5, ICOS, and PD-1 but also express FOXP3, as conventional Tregs (25). Importantly, Tfr cells seem to have a suppressive function inside the GCs.

Nodal T-Follicular helper cell lymphomas

In the revised 4th edition of the Classification of Hematolymphoid Neoplasms of the World Health Organization (WHO), three lymphoma entities with a TFH gene expression signature were included and designated as AITL, nodal PTCL with a TFH phenotype and follicular T-cell lymphoma (FTCL) and grouped under the provisional umbrella category of nodal lymphomas of TFH origin (Table 1) (26). In the updated classification (5th Edition), these lymphomas are currently unified as nodal follicular helper T-cell lymphomas (TFH lymphoma) with three diseases, AITL-type, follicular-type, and not otherwise specified (NOS) (1). A similar approach and terminology have been adopted in the International Consensus Classification (ICC) (2). While the nomenclature is almost identical, there are minor differences, including the term nodal in the WHO, as implied in the ICC (Table 1) (1, 2). The immunomarkers most frequently used to establish a TFH immunophenotype in clinical practice include PD-1, CXCL13, CXCR5, ICOS, BCL6, and CD10.

Nodal TFH cell lymphoma, angioimmunoblastic-type

For simplicity, we refer to this lymphoma as the historically used abbreviation AITL. AITL is the prototype of nodal TFH lymphomas, and it is one of the most common PTCL subtypes. There are some geographical variations in the frequency of AITL, with reported higher incidences in Europe than in the US; AITL in Europe represents about 35% of non-cutaneous T-cell lymphomas (TCLs) and ~16% in the US (27, 28). The diagnosis of AITL is based on a combination of clinical and laboratory findings, distinctive histopathologic features, and expression of TFH immunophenotype by the neoplastic cells. The neoplastic cells are characterized by the expression in variable rates of TFH markers such as CD10, Bcl-6, PD-1/CD279, ICOS, and CXCL13 (6, 26, 29). Other reported TFH markers that are not routinely used in clinical practice, include CXCR5, SAP, c-MAF, and CD200 (30–32).

TABLE 1 Different designations for nodal T cell lymphomas with a TFH immunophenotype according to the different classifications schemas.

WHO 2008	WHO 2017	ICC 2022	WHO 2022
	Nodal lymphomas of T-follicular helper cell origin	Follicular helper T-cell lymphoma	Nodal TFH cell lymphoma
Angioimmunoblastic T-cell lymphoma	*Angioimmunoblastic T-cell lymphoma*	Follicular helper T-cell lymphoma, angioimmunoblastic type	Nodal TFH lymphoma, angioimmunoblastic-type
Follicular variant of PTCL, NOS	Follicular T-cell lymphoma	Follicular helper T-cell lymphoma, follicular type	Nodal TFH lymphoma, follicular-type
Subset of PTCL, NOS including T-zone variant	Nodal peripheral T-cell lymphoma with TFH phenotype	Follicular helper T-cell lymphoma, NOS	Nodal TFH lymphoma, NOS

WHO, World Health Organization; ICC, International Consensus Classification; PTCL, peripheral T cell lymphoma, not otherwise specified; TFH, T-follicular helper.

Epidemiology and clinical features

AITL commonly affects middle-aged patients in the fifth to sixth decades, and there is a nearly equal incidence between genders and ethnicity. No racial predisposition is recognized. The most common clinical presentation is generalized lymphadenopathy (typically less than 3 cm), hepatosplenomegaly, constitutional symptoms, and skin rashes secondary to either neoplastic T-cell infiltration or as an autoimmune paraneoplastic manifestation (33). At diagnosis, most patients present with advanced stage and extranodal involvement. The most common sites of extranodal involvement are the spleen, bone marrow, skin, and lungs. Bone marrow involvement tends to occur early in the disease course, and its diagnosis can be extremely challenging (34). AITL is often associated with immune dysregulation, resulting in autoimmune complications and opportunistic infections (35, 36). The autoimmune and immunologic manifestations in TFH lymphomas are likely related to the functional and key role of TFH cells in B-cell activation, differentiation to plasma cells and B-cell recruitment. There are anecdotal reports of patients presenting with a smoldering course with waxing and waning lymphadenopathy (37, 38). While AITL is a systemic disease, rare cases of AITL and PTCL with TFH phenotype with localized disease and more indolent behavior have been reported, for example cases localized to the Waldeyer's ring at presentation (39).

Morphologic features

Lymph nodes involved by AITL typically show partial or complete effacement with usually a diffuse or paracortical growth pattern and frequent perinodal extension but sparing of the subcortical sinuses (Figure 1). The neoplastic T-cells often constitute a minor part of a polymorphic inflammatory cellular infiltrate. The tumor cells are usually small- to medium-sized with mild nuclear atypia and clear cytoplasm. While they can be difficult to identify, they usually cluster around high endothelial venules (HEV) and are entrapped by the follicular dendritic cell (FDC) meshworks. Scattered or small groups of medium to large tumor cells with clear cytoplasm have been associated with $IDH2^{R172}$ mutations (40). Additional distinctive pathologic features include the proliferation of arborizing high endothelial venules (HEV) surrounded by expanded networks of follicular dendritic cells (FDCs), and an inflammatory background with plasma cells, eosinophils, histiocytes, and scattered B-cell immunoblasts. Plasma cells may be abundant, in rare cases obscuring the neoplastic T-cells, and are usually polyclonal but may be monoclonal in a few cases (41).

Three overlapping architectural patterns (types I, II, and III) have been described by Attygale and colls (29). Pattern III is the most frequent (~80%) and is characterized by a total effacement of the nodal architecture without follicles. Pattern II shows multiple regressive "burnout"/atretic germinal centers, and pattern I (reactive hyperplasia-like) is characterized by reactive hyperplastic germinal centers with the lymphoma located in the interfollicular regions and associated with minimal expansion of FDC. A tumor cell-rich pattern has been recognized and likely represents a histological progression in patients with previous AITL (42). This pattern is enriched with tumor cells and is associated with minimal inflammatory background and limited or no HEV proliferation simulating PTCL-NOS. Other histologic variants of AITL include epithelioid-rich AITL, plasma cell-rich AITL, and AITL with Hodgkin Reed-Sternberg (HRS)-like cells. The epithelioid-rich AITL is characterized by a high content of epithelioid cells simulating lymphoepitheloid lymphoma/Lennert lymphoma or a granulomatous reaction (43, 44).

Frequently, AITL contains a variable number of B immunoblasts, which can be EBV-positive or negative. In some cases, B-immunoblasts can be monoclonal and form confluent sheets of large B-cells that morphologically meet the diagnostic criteria of large B cell lymphoma (27, 42, 45, 46). More rarely, the associated clonal B-cell proliferations are characterized by small lymphocytes and/or plasma cells, which may resemble nodal or extranodal marginal zone lymphoma (47).

HRS-like B-cells are not unusual in AITL, which are more frequently EBV-positive (48). These HRS-like cells can be rosetted by neoplastic T-cells, express B-cell markers such as CD20, or occasionally have overlapping immunophenotype with classical Hodgkin lymphoma (CHL), posing a diagnostic difficulty (42, 48).

Several reactive lymph node conditions with predominant paracortical and/or interfollicular patterns may morphologically resemble TCL. These include viral-induced lymphadenopathies, drug reactions in patients receiving anticonvulsant therapy (most commonly diphenylhydantoin), antibiotics or antivirals, and vaccination-induced reactions, as well as other non-specific etiologies. Drug-induced hypersensitivity syndrome/drug reaction with eosinophilia and systemic symptoms (DRESS) syndrome (also called Dilantin-associated lymphadenopathy) is a drug-induced severe adverse reaction that can be associated with lymphadenopathy, mimicking AITL (49).

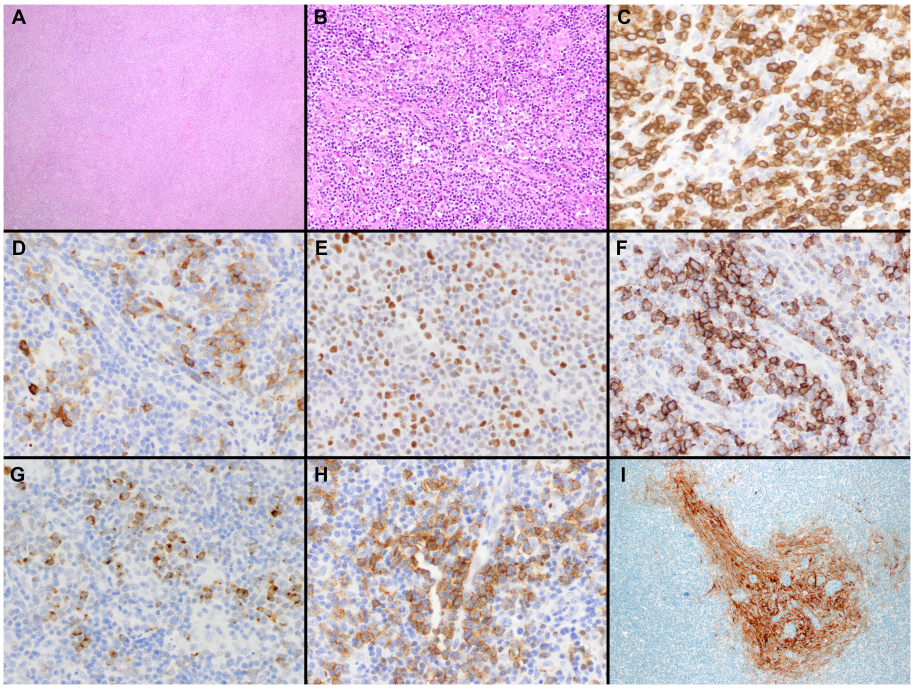


FIGURE 1
Histopathologic features of nodal T-follicular helper (TFH) cell lymphoma, angioimmunoblastic-type. **(A, B)** Hematoxylin & Eosin (H&E) shows that the neoplasm completely effaces the nodal architecture **(A)**; the neoplasm is diffuse and composed of a heterogeneous cell infiltrate associated with numerous high endothelial venules (HEVs), inflammatory cells, and clusters of small lymphocytes with clear cytoplasm **(B)** (2x and 40x); **(C)** CD3 shows that most of the cells in the infiltrate are T-cells. Some of the T-cells are intermediate in size with irregular nuclear contours (40x); **(D-H)** The tumor cells are positive for CD10 **(D)**, BCL6 **(E)**, ICOS **(F)**, CXCL13 **(G)** and PD-1 **(H)** supporting a TFH immunophenotype (all 40x). Note the clustering of the tumor cells around the HEVs. **(I)** CD21 highlights focally expanded follicular dendritic cell meshworks surrounding HEVs (20x).

Immunophenotypic features

The neoplastic T-cells are positive for alpha/beta TCR and CD4 and typically express pan-T cell markers, including CD2, CD3, and CD5. The aberrant loss or downregulation of one or more T-cell markers is frequently observed (50). The markers used to identify a TFH immunophenotype in paraffin-embedded tissue sections include PD-1, CXCL13, CXCR5, ICOS, BCL6, and CD10. PD-1 and ICOS are more sensitive than CXCL13 or CD10; conversely, CXCL13 and CD10 are more specific (51, 52). Partial expression of CD30 by the tumor cells is not unusual, and aberrant expression of CD20 by the lymphoma cells has also been reported (53). High expression of CD20 has been reported to be associated with a better overall survival (54–56). The use of follicular dendritic cell markers (e.g., CD21, CD23, or CD35) is helpful to assess for perivascular expansion of FDCs. CD23 has been recommended when staining for CD21 fails to show perivenular expansion of the FDCs (42). AITL with *IDH2* mutations has stronger expression of CD10 and CXCL13. An antibody against $IDH2^{R172K}$ is available and can detect most cases with this mutation (57).

Flow cytometry frequently reveals decreased or absent expression of surface CD3 (58, 59). Also, the detection of a T-cell population by flow cytometry coexpressing CD4/CD10 or CD4/PD-1 (bright) on lymph nodes, bone marrow, or peripheral blood samples may help to support the diagnosis (60).

There have been occasional reports of AITL with cytotoxic phenotypes (61) and the possibility of a florid CD8+ cytotoxic cell proliferation obscuring a neoplastic TFH cell population has also been reported (62).

Molecular features

Mutational profile

AITL frequently shows a distinctive mutational profile with mutations involving $RHOA^{G17V}$ (50-70%), *TET2* (40-80%), $IDH2^{R172}$ (20-45%) and *DNMT3A* (20-30%) (Table 2) (5, 55, 56).

RHOA is a small GTPase involved in T-cell migration, polarization, and antigen recognition by cycling between GDP-bound (inactive) and GTP-bound (active states). The c.50G>T (p.Gly17Val) is the most frequent missense mutation of *RHOA* in AITL (56, 63). Mutations in $RHOA^{G17V}$ act as dominant negatives interfering with the signaling initiated by wild-type *RHOA* (64) and seem to be a secondary event contributing to the differentiation toward TFH phenotype (65). These mutations frequently co-occur with mutations in epigenetic regulators, especially *TET2* mutations. Mutated *RHOA* acquires a novel function, binding and phosphorylating VAV1 protein (66). $RHOA^{G17V}$ mutation seems to increase TCR signaling through enhanced VAV1 resulting in stronger TCR signaling and preferential commitment to TFH rather than non-TFH lineage through enhancing ICOS signaling (67).

IDH2 is a metabolic mitochondrial enzyme involved in the generation of 2-oxoglutarate (2-HG). Mutant *IDH2* forms have a neomorphic enzymatic activity leading instead to the generation of 2-hydroxyglutarate (2-HG), an oncometabolite that antagonizes the activity of α-KG-dependent dioxygenases (histone demethylases and the TET family of 5mC hydroxylases) (68, 69). They are described as a secondary event and might refine the differentiation of the premalignant clones towards a TFH signature (70). $IDH2^{R172}$

TABLE 2 Mutations associated with nodal follicular helper T cell lymphomas.

Genes	Frequency			
	AITL	nTFH-NOS	nTFH-FL	
GTPase				
RHOA^{G17V}	50-70	25-50	60	• G17V specific to AITL/PTCL-TFH
				• Not associated with prognosis
Epigenetic regulators				
TET2	40-80	50-75	75	• Found in other neoplasms (myeloid)
DNMT3A	20-30	7-18	25	• *TET2* co-occur w/ *DNMT3* and *IDH2* mutations is specific to TFH lymphomas
IDH2^{R172}	20-45	0	0	• *IDH2* mutations mostly restricted to AITL • Presence of clear cells • More pronounced TFH signature • Strong CD10 and CXCL13 expression • Chr 5 and 21 gains • More aberrant genome than IDH2 negative cases • Clinical trial: enasidenib
TCR signaling pathway				
PLCγ	8-14	6.25	N/A	• Not specific (PTCL-NOS)
CD28	10-12	0	N/A	• Worse prognosis in AILT

N/A, Not available.

mutations are associated with cases with a more pronounced TFH signature, strong CD10 and CXCL13 expression, gains of chromosomes 5 and 21, and more aberrant genome than the cases without *IDH2*^{R172} (40, 71). The more aberrant genome seen in cases with *IDH2*^{R172} mutations is likely due to the inhibitory effect of 2-HG oncometabolite on DNA repair enzymes (72). AITL with wild-type *IDH2* show significant enrichment of PI3K-AKT activation pathways and have focal losses of negative regulators (phosphatases) of the PI3K-AKT pathway (71).

TET2, also known as ten-eleven translocation 2 (TET2), encodes a 2-oxoglutarate/Fe2+-dependent oxygease that participates in the epigenetic control of gene expression by catalyzing the oxidation of DNA 5-methylcytosine to 5-hydroxymethylcytosine (73, 74). Its loss-of function is an initial event in the neoplastic transformation and is associated with a worse outcome (75). *DNMT3A* encodes a DNA methyltransferase that controls cytosine methylation. Loss-of-function mutations in *DNMT3A* are also considered an initial event in the transformation process and frequently co-occur with *TET2* mutations (69).

Other recurrent mutations frequently seen in AITL include TCR signaling genes, such as *VAV1, PLCG1, CD28*, and *FYN* (76). Those mutations, except for specific mutation and fusions of CD28, are not specific to AITL or TFH lymphomas. Mutations in the *CD28* gene appear to show implications in outcomes since CD28-mutated AITL patients have inferior survival compared to patients with wild-type CD28 (77). Alterations in *RHOA* and *VAV1* are mutually exclusive.

Relationship with clonal hematopoiesis

TET2 and *DNMT3A* mutations are not specific to AITL. However, different from other neoplasms these two mutations frequently co-occur in AITL. *TET2* and *DNMT3a* mutations are the most frequent mutations in clonal hematopoiesis (CH). Many studies have now established that up to 80% of patients with TFH lymphomas carry the same *TET2* and/or *DNMT3A* mutations identified in the T-cells and the myeloid cells (78–80). Furthermore, *TET2* and *DNMT3A* mutations are not restricted to T-cells and myeloid cells but can also be identified in the admixed mature B-cells in the lymph nodes. On the contrary, *RHOA* and *IDH2* mutations appear to be confined to the neoplastic T-cells and represent the "second" hit that contribute to the T-cell lymphomagenesis (65, 79, 81).

The background of clonal hematopoiesis of indetermined potential (CHIP) appears to be the source of myeloid neoplasm seen in TCL with a TFH phenotype, particularly after cytotoxic therapy (78, 79, 82). The occurrence of AML and other myeloid neoplasms after the diagnosis of AITL is significantly high (79). From a practical perspective, it is important to remember that in a patient being followed for AITL the presence of CHIP mutations does not necessary imply the presence of residual lymphoma.

Gene expression profile

In AITL, the expression profiling signatures are enriched in genes typically expressed by TFH cells (55, 56, 71). This demonstration of molecular similarities between AITL cells and TFH cells at a genome-wide level established the cellular "derivation" of AITL from TFH cells, initially suspected based on the expression of single TFH markers in AITL cells (3, 4, 6).The molecular profile of AITL is also dominated by a strong microenvironment imprint, including overexpression of B-cell and FDC-related genes, chemokines/chemokine receptors, and genes related to the extracellular matrix and vascular biology (52, 83, 84). Gene expression studies identified oncogenic pathways, including the NF-κB pathway, IL-6 signaling, and the TGFβ pathway enriched in AITL compared to other PTCLs (3, 4, 6, 55), but the genetic etiology of these activated pathways has not been completely elucidated.

Recently, Amador and colls have developed a digital gene expression classifier using specific signatures in nodal T-cell lymphomas and included a specific signature for nodal AITL (54). This assay can be used in paraffin-embedded tissue sections and thus can easily be translated to routine clinical practice to complement our conventional pathology approaches to better classify nodal TCLs.

Nodal TFH cell lymphoma, follicular type

For simplicity, we refer to this lymphoma as the historically used abbreviation FTCL. These are nodal TCLs with a nodular growth pattern that show significant histologic, immunophenotypic, transcriptomic, and genetic overlap with AITL (5). Isolated FTCL is very rare and usually occurs in association with AITL. Some patients at disease presentation have typical histologic and clinical features of AITL but, at relapse, show features of FTCL or vice versa (42, 85). Therefore it has been proposed that these two entities may constitute different morphologic representations of the same biological process (1, 2).

Epidemiology and clinical features

The median age of onset is 60-65 years with a slight male predominance (2). The clinical syndrome resembles AITL and other TFH lymphomas and is characterized by advanced-stage disease, generalized lymphadenopathy, splenomegaly, B symptoms, skin rash, and occasionally immune manifestations (85). Cases with localized disease have also been reported (86).

Morphologic features

Two patterns have been described. In the classic pathology description, FTCL has a follicular growth pattern mimicking follicular lymphoma (FL), where the follicles are populated by aberrant T-cells that express TFH markers (Figure 2) (85). Residual B-cells can be seen and are usually pushed to the periphery of the follicles by the neoplastic T-cells. Hodgkin and Reed-Sternberg (HRS)-like cells are also frequently noted. Alternatively, it can also mimic progressive transformation of germinal centers (PTGC). In this pattern, the nodules display a 'moth-eaten' appearance with aggregates of the neoplastic T-cells surrounded by small B-cells (85). Mixed FL-like and PTGC-like patterns can be seen. Focal paracortical hyperplasia is present with a polymorphic infiltrate of eosinophils and plasma cells and hyperplastic high endothelial venules.

Immunophenotypic features

The neoplastic T-cells have a TFH phenotype and typically express pan-T cell markers, including CD2, CD3, and CD5, although aberrant loss or downregulation of CD7 can be seen. By flow cytometry, dim expression of surface CD3 is frequently observed. CD4 is positive in most cases, with few instances double negative for CD4 and CD8 (85). The HRS-like cells are of B-cell lineage, positive for CD30 and positive for CD15 and EBER in some cases. This phenomenon raises concern for classic Hodgkin lymphoma. Neoplastic rosetting by T-cells around the HRS-like cells is seen in virtually all cases of FTCL, and a retained network of FDCs is seen underlying the neoplastic follicles. FTCL usually lacks the other typical features of AITL, including the characteristic expanded FDC networks surrounding HEVs (85).

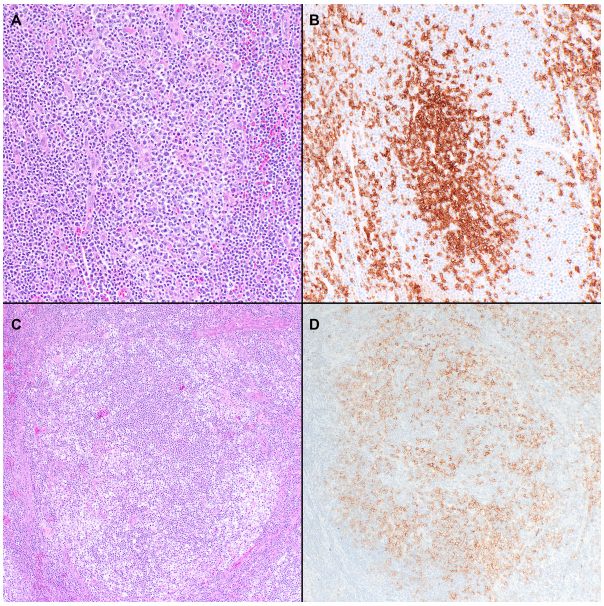


FIGURE 2
Histopathologic features of nodal T-follicular helper (TFH) cell lymphoma, follicular-type. **(A)** This case shows a predominantly follicular growth pattern, simulating follicular lymphoma, with the neoplastic follicles composed of small to medium-sized atypical lymphocytes admixed with scattered large cells (20x); **(B)** PD-1 shows that the tumor cells form solid clusters inside the nodules with residual B-cells pushed to the periphery of the follicles (not shown) (20x); **(C)** This other case shows that the neoplastic nodules have features of progressive transformation of germinal centers (PTGC). The nodules display a 'moth-eaten' appearance with aggregates of neoplastic T-cells surrounded by small B-cells (20x); **(D)** The neoplastic cells are positive for PD-1 in addition to other TFH markers (not shown) (20x).

Molecular features

The few cases included in gene expression profiling studies have shown that FTCL clusters closer to AITL than PTCL-NOS (5). Similarly, the mutational profile of FTCL seems to be similar to other nodal TFH lymphomas with mutations in *TET2, DNMT3A*, and $RHOA^{G17V}$, but not in *IDH2* which are usually restricted to AITL (5). FTCL can harbor a characteristic t(5;9)(q33;q22) resulting in an *ITK-SYK* fusion in approximately 40% of cases (87). This fusion acts as a constitutively active SYK tyrosine kinase and drives lymphomagenesis by triggering antigen-independent activation of TCR signaling. Translocations involving *FER* and *FES* have been recently described, including *ITK-FER* and *RLTPR-FES*, that result in the activation of the STAT3 signaling (88).

Nodal TFH cell lymphoma, not otherwise specified

Nodal TFH lymphoma, NOS includes those TCLs with TFH phenotype, confirmed by the expression of CD4 and at least 2 TFH markers that lack the morphologic features of AITL and FTCL. This group includes cases previously categorized as PTCL-NOS that have shown significant molecular and genetic overlap with other TFH lymphomas (5, 56, 71). It is possible that TFH lymphoma, NOS, represents more than a single entity as currently defined.

Epidemiology and clinical features

The frequency of this neoplasm is unknown. However, it is our personal experience that up to 30% of the PTCL-NOS are reclassified as nodal TFH, NOS when a panel of TFH immunomarkers is analyzed. The patients usually present with disseminated lymphadenopathy associated, which can be associated with autoimmune manifestations (5).

Morphologic features

The overall morphologic spectrum of these tumors has not been completely elucidated. Most cases are characterized by a diffuse tumor-cell-rich infiltrate of variably-sized lymphoid cells without the typical HEC and FDC proliferations (Figure 3) (1, 2). Additionally, cases previously described as lymphoepithelioid lymphomas with TFH marker expression are currently included in this group (89). A systematic evaluation of these cases shows that they can frequently have one or two AITL-like features not commonly seen in PTCL-NOS cases (51). There are some overlapping features between nodal TFH lymphoma, NOS and the tumor cell-rich pattern of AITL. However, it has been mentioned that the tumor cell-rich pattern of AITL still maintains the focal perivenular FDC expansions (1).

Immunophenotypic features

By definition, the tumor cells are positive for CD4 and express at least 2 TFH makers. Some cases show focal positivity for TFH markers and positivity in a small subset of the tumor cells, being difficult to classify. Currently, there is no standard percentage of TFH positivity and the intensity of expression in tumor cells needed to establish a reproducible diagnosis of TFH lymphoma, NOS over PTCL, NOS.

Molecular features

Similarly to FTCL, gene expression studies show that TFH lymphoma, NOS clusters closer to AITL than PTCL-NOS (5, 76). The mutational profile of TFH lymphoma, NOS, seems similar to AITL mutations in *TET2, DNMT3A*, and $RHOA^{G17V}$ (5, 56, 76). $IDH2^{R172}$ mutation is characteristic of AITL, although it can rarely be seen in TFH lymphoma, NOS (90). $RHOA^{G17V}$ mutations were identified in 60% of the cases (76). *TET2* mutations seem slightly more frequent in TFH lymphoma, NOS (5). Mutations in genes of the

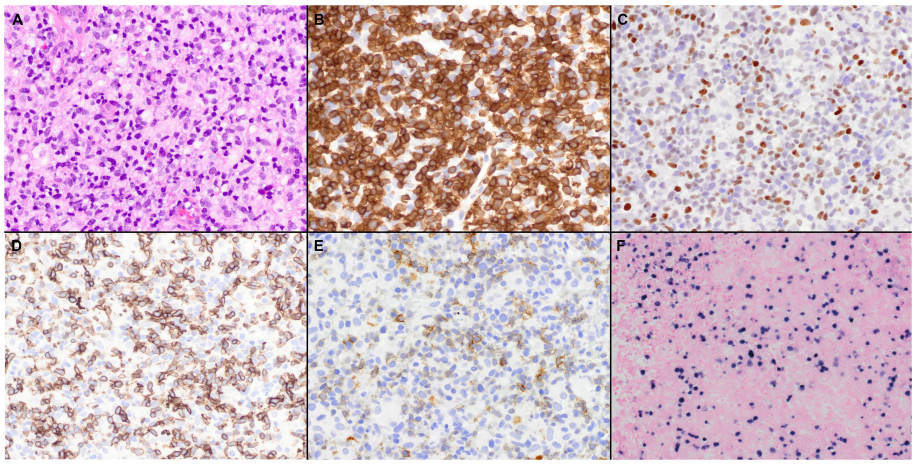


FIGURE 3
Histopathologic features of nodal TFH cell lymphoma, not otherwise specified (NOS). **(A)** In the case shown, the neoplasm is diffuse and composed predominantly of intermediate atypical lymphoid cells (40x); Features of AITL were not seen. **(B)** The tumor cells are positive for CD3 (40x); **(C-E)** BCL6 **(C)**, ICOS **(D)**, and PD-1 **(E)** are also variably positive in the neoplastic cells (40x); **(F)** Epstein-Barr virus-encoded small RNAs (EBER) shows positivity in scattered cells (20x).

TCR signaling pathway (including CD28) can be seen in a subset of cases (76, 90). It has been reported that the presence of TCR-signaling-related mutations correlated with early disease progression (76).

Conclusions

TFH lymphomas are a group of mature peripheral T-cell lymphomas with distinctive clinicopathologic and molecular features. Although AITL is well-characterized and has unique morphologic features that facilitate its diagnosis, the other subtypes, mainly the TFH lymphoma, NOS, is one of exclusion. Additional studies are required to better understand and delineate this category of nodal TFH lymphomas.

Since the diagnosis of nodal TFH lymphomas is very challenging, especially in small core needle biopsies, the diagnostic evaluation of TCLs requires the integration of clinicopathologic features together with a complete panel of TFH markers. We envision that incorporating mutational studies and gene expression profiling in the near future into the routine diagnostic armamentarium will facilitate the diagnosis of TFH lymphomas.

Author contributions

All the authors performed writing, reviewing, and revisions of the manuscript. All authors contributed to the article and approved the submitted version.

Conflict of interest

FV receives research funding from CRISPR Therapeutics, Allogene Therapeutics, and Geron Corporation.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: Lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

2. Campo E, Jaffe ES, Cook JR, Quintanilla-Martinez L, Swerdlow SH, Anderson KC, et al. The international consensus classification of mature lymphoid neoplasms: A report from the clinical advisory committee. *Blood* (2022) 140(11):1229–53. doi: 10.1182/blood.2022015851

3. de Leval L, Rickman DS, Thielen C, Reynies A, Huang YL, Delsol G, et al. The gene expression profile of nodal peripheral T-cell lymphoma demonstrates a molecular link between angioimmunoblastic T-cell lymphoma (AITL) and follicular helper T (TFH) cells. *Blood* (2007) 109(11):4952–63. doi: 10.1182/blood-2006-10-055145

4. Piccaluga PP, Agostinelli C, Califano A, Carbone A, Fantoni L, Ferrari S, et al. Gene expression analysis of angioimmunoblastic lymphoma indicates derivation from T follicular helper cells and vascular endothelial growth factor deregulation. *Cancer Res* (2007) 67(22):10703–10. doi: 10.1158/0008-5472.CAN-07-1708

5. Dobay MP, Lemonnier F, Missiaglia E, Bastard C, Vallois D, Jais JP, et al. Integrative clinicopathological and molecular analyses of angioimmunoblastic T-cell lymphoma and other nodal lymphomas of follicular helper T-cell origin. *Haematologica* (2017) 102(4): e148–51. doi: 10.3324/haematol.2016.158428

6. Grogg KL, Attygalle AD, Macon WR, Remstein ED, Kurtin PJ, Dogan A. Angioimmunoblastic T-cell lymphoma: A neoplasm of germinal-center T-helper cells? *Blood* (2005) 106(4):1501–2. doi: 10.1182/blood-2005-03-1083

7. Crotty S. Follicular helper CD4 T cells (TFH). *Annu Rev Immunol* (2011) 29:621–63. doi: 10.1146/annurev-immunol-031210-101400

8. Ribeiro F, Perucha E, Graca L. T Follicular cells: The regulators of germinal center homeostasis. *Immunol Lett* (2022) 244:1–11. doi: 10.1016/j.imlet.2022.02.008

9. Liu Z, Fan H, Jiang S. CD4(+) T-cell subsets in transplantation. *Immunol Rev* (2013) 252(1):183–91. doi: 10.1111/imr.12038

10. Crotty S. T Follicular helper cell biology: A decade of discovery and diseases. *Immunity* (2019) 50(5):1132–48. doi: 10.1016/j.immuni.2019.04.011

11. Ioannidou K, Ndiaye DR, Noto A, Fenwick C, Fortis SP, Pantaleo G, et al. In situ characterization of follicular helper CD4 T cells using multiplexed imaging. *Front Immunol* (2020) 11:607626. doi: 10.3389/fimmu.2020.607626

12. Grimbacher B, Hutloff A, Schlesier M, Glocker E, Warnatz K, Drager R, et al. Homozygous loss of ICOS is associated with adult-onset common variable immunodeficiency. *Nat Immunol* (2003) 4(3):261–8. doi: 10.1038/ni902

13. Kerfoot SM, Yaari G, Patel JR, Johnson KL, Gonzalez DG, Kleinstein SH, et al. Germinal center b cell and T follicular helper cell development initiates in the interfollicular zone. *Immunity* (2011) 34(6):947–60. doi: 10.1016/j.immuni.2011.03.024

14. Johnston RJ, Poholek AC, DiToro D, Yusuf I, Eto D, Barnett B, et al. Bcl6 and blimp-1 are reciprocal and antagonistic regulators of T follicular helper cell differentiation. *Science* (2009) 325(5943):1006–10. doi: 10.1126/science.1175870

15. Nakayamada S, Kanno Y, Takahashi H, Jankovic D, Lu KT, Johnson TA, et al. Early Th1 cell differentiation is marked by a tfh cell-like transition. *Immunity* (2011) 35 (6):919–31. doi: 10.1016/j.immuni.2011.11.012

16. Hercor M, Anciaux M, Denanglaire S, Debuisson D, Leo O, Andris F. Antigen-presenting cell-derived IL-6 restricts the expression of GATA3 and IL-4 by follicular helper T cells. *J Leukoc Biol* (2017) 101(1):5–14. doi: 10.1189/jlb.1HI1115-511R

17. Nurieva RI, Chung Y, Martinez GJ, Yang XO, Tanaka S, Matskevitch TD, et al. Bcl6 mediates the development of T follicular helper cells. *Science* (2009) 325(5943):1001–5. doi: 10.1126/science.1176676

18. Chen X, Ma W, Zhang T, Wu L, Qi H. Phenotypic tfh development promoted by CXCR5-controlled re-localization and IL-6 from radiation-resistant cells. *Protein Cell* (2015) 6(11):825–32. doi: 10.1007/s13238-015-0210-0

19. Baessler A, Novis CL, Shen Z, Perovanovic J, Wadsworth M, Thiede KA, et al. Tet2 coordinates with Foxo1 and Runx1 to balance T follicular helper cell and T helper 1 cell differentiation. *Sci Adv* (2022) 8(24):eabm4982. doi: 10.1126/sciadv.abm4982

20. Sage PT, Alvarez D, Godec J, von Andrian UH, Sharpe AH. Circulating T follicular regulatory and helper cells have memory-like properties. *J Clin Invest* (2014) 124 (12):5191–204. doi: 10.1172/JCI76861

21. Weber JP, Fuhrmann F, Hutloff A. T-Follicular helper cells survive as long-term memory cells. *Eur J Immunol* (2012) 42(8):1981–8. doi: 10.1002/eji.201242540

22. Herati RS, Muselman A, Vella L, Bengsch B, Parkhouse K, Del Alcazar D, et al. Successive annual influenza vaccination induces a recurrent oligoclonotypic memory response in circulating T follicular helper cells. *Sci Immunol* (2017) 2(8). doi: 10.1126/sciimmunol.aag2152

23. Locci M, Havenar-Daughton C, Landais E, Wu J, Kroenke MA, Arlehamn CL, et al. Human circulating PD-1+CXCR3-CXCR5+ memory tfh cells are highly functional and correlate with broadly neutralizing HIV antibody responses. *Immunity* (2013) 39(4):758–69. doi: 10.1016/j.immuni.2013.08.031

24. Wang J, Shan Y, Jiang Z, Feng J, Li C, Ma L, et al. High frequencies of activated b cells and T follicular helper cells are correlated with disease activity in patients with new-onset rheumatoid arthritis. *Clin Exp Immunol* (2013) 174(2):212–20. doi: 10.1111/cei.12162

25. Linterman MA, Pierson W, Lee SK, Kallies A, Kawamoto S, Rayner TF, et al. Foxp3 + follicular regulatory T cells control the germinal center response. *Nat Med* (2011) 17 (8):975–82. doi: 10.1038/nm.2425

26. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, et al. *WHO classification of tuours of haematopoietic and lymphoid tissues. 4th edition*. Lyon: IARC (2017).

27. de Leval L, Parrens M, Le Bras F, Jais JP, Fataccioli V, Martin A, et al. Angioimmunoblastic T-cell lymphoma is the most common T-cell lymphoma in two distinct French information data sets. *Haematologica* (2015) 100(9):e361–4. doi: 10.3324/haematol.2015.126300

28. Vose J, Armitage J, Weisenburger D, International TCLP. International peripheral T-cell and natural killer/T-cell lymphoma study: Pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26(25):4124–30. doi: 10.1200/JCO.2008.16.4558

29. Attygalle A, Al-Jehani R, Diss TC, Munson P, Liu H, Du MQ, et al. Neoplastic T cells in angioimmunoblastic T-cell lymphoma express CD10. *Blood* (2002) 99(2):627–33. doi: 10.1182/blood.V99.2.627

30. Murakami YI, Yatabe Y, Sakaguchi T, Sasaki E, Yamashita Y, Morito N, et al. C-maf expression in angioimmunoblastic T-cell lymphoma. *Am J Surg Pathol* (2007) 31 (11):1695–702. doi: 10.1097/PAS.0b013e318054dbcf

31. Dorfman DM, Shahsafaei A. CD200 (OX-2 membrane glycoprotein) is expressed by follicular T helper cells and in angioimmunoblastic T-cell lymphoma. *Am J Surg Pathol* (2011) 35(1):76–83. doi: 10.1097/PAS.0b013e31820065c9

32. Roncador G, Garcia Verdes-Montenegro JF, Tedoldi S, Paterson JC, Klapper W, Ballabio E, et al. Expression of two markers of germinal center T cells (SAP and PD-1) in angioimmunoblastic T-cell lymphoma. *Haematologica* (2007) 92(8):1059–66. doi: 10.3324/haematol.10864

33. Lachenal F, Berger F, Ghesquieres H, Biron P, Hot A, Callet-Bauchu E, et al. Angioimmunoblastic T-cell lymphoma: Clinical and laboratory features at diagnosis in 77 patients. *Med (Baltimore)* (2007) 86(5):282–92. doi: 10.1097/MD.0b013e3181573059

34. Khokhar FA, Payne WD, Talwalkar SS, Jorgensen JL, Bueso-Ramos CE, Medeiros LJ, et al. Angioimmunoblastic T-cell lymphoma in bone marrow: A morphologic and immunophenotypic study. *Hum Pathol* (2010) 41(1):79–87. doi: 10.1016/j.humpath.2009.06.016

35. Dunleavy K, Wilson WH, Jaffe ES. Angioimmunoblastic T cell lymphoma: Pathobiological insights and clinical implications. *Curr Opin Hematol* (2007) 14 (4):348–53. doi: 10.1097/MOH.0b013e328186ffbf

36. Mourad N, Mounier N, Briere J, Raffoux E, Delmer A, Feller A, et al. Clinical, biologic, and pathologic features in 157 patients with angioimmunoblastic T-cell lymphoma treated within the groupe d'Etude des lymphomes de l'Adulte (GELA) trials. *Blood* (2008) 111(9):4463–70. doi: 10.1182/blood-2007-08-105759

37. Federico M, Rudiger T, Bellei M, Nathwani BN, Luminari S, Coiffier B, et al. Clinicopathologic characteristics of angioimmunoblastic T-cell lymphoma: Analysis of the international peripheral T-cell lymphoma project. *J Clin Oncol* (2013) 31(2):240–6. doi: 10.1200/JCO.2011.37.3647

38. Tan LH, Tan SY, Tang T, Lim ST, Tan D, Lim LC, et al. Angioimmunoblastic T-cell lymphoma with hyperplastic germinal centres (pattern 1) shows superior survival to patterns 2 and 3: A meta-analysis of 56 cases. *Histopathology* (2012) 60(4):570–85. doi: 10.1111/j.1365-2559.2011.04097.x

39. Wang J, Tang W, Zhang W, Wang J, Chen F, Zhong L, et al. Clinicopathological characterization of follicular helper T-cell-derived peripheral T-cell lymphoma involving waldeyer's ring. *Virchows Arch* (2021) 479(2):355–63. doi: 10.1007/s00428-021-03071-z

40. Steinhilber J, Mederake M, Bonzheim I, Serinsoz-Linke E, Muller I, Fallier-Becker P, et al. The pathological features of angioimmunoblastic T-cell lymphomas with IDH2 (R172) mutations. *Mod Pathol* (2019) 32(8):1123–34. doi: 10.1038/s41379-019-0254-4

41. Huppmann AR, Roullet MR, Raffeld M, Jaffe ES. Angioimmunoblastic T-cell lymphoma partially obscured by an Epstein-Barr virus-negative clonal plasma cell proliferation. *J Clin Oncol* (2013) 31(2):e28–30. doi: 10.1200/JCO.2012.43.3797

42. Attygalle AD, Cabecadas J, Gaulard P, Jaffe ES, de Jong D, Ko YH, et al. Peripheral T-cell and NK-cell lymphomas and their mimics; taking a step forward - report on the lymphoma workshop of the XVIth meeting of the European association for haematopathology and the society for hematopathology. *Histopathology* (2014) 64 (2):171–99. doi: 10.1111/his.12251

43. Hartmann S, Agostinelli C, Klapper W, Korkolopoulou P, Koch K, Marafioti T, et al. Revising the historical collection of epithelioid cell-rich lymphomas of the Kiel lymph node registry: What is lennert's lymphoma nowadays? *Histopathology* (2011) 59 (6):1173–82. doi: 10.1111/j.1365-2559.2011.04069.x

44. Agostinelli C, Hartmann S, Klapper W, Korkolopoulou P, Righi S, Marafioti T, et al. Peripheral T cell lymphomas with follicular T helper phenotype: A new basket or a distinct entity? revising Karl lennert's personal archive. *Histopathology* (2011) 59(4):679–91. doi: 10.1111/j.1365-2559.2011.03981.x

45. Tokunaga T, Shimada K, Yamamoto K, Chihara D, Ichihashi T, Oshima R, et al. Retrospective analysis of prognostic factors for angioimmunoblastic T-cell lymphoma: A multicenter cooperative study in Japan. *Blood* (2012) 119(12):2837–43. doi: 10.1182/blood-2011-08-374371

46. Zettl A, Lee SS, Rudiger T, Starostik P, Marino M, Kirchner T, et al. Epstein-Barr Virus-associated b-cell lymphoproliferative disorders in angloimmunoblastic T-cell lymphoma and peripheral T-cell lymphoma, unspecified. *Am J Clin Pathol* (2002) 117 (3):368–79. doi: 10.1309/6UTX-GVC0-12ND-JJEU

47. Kaffenberger B, Haverkos B, Tyler K, Wong HK, Porcu P, Gru AA. Extranodal marginal zone lymphoma-like presentations of angioimmunoblastic T-cell lymphoma: A T-cell lymphoma masquerading as a b-cell lymphoproliferative disorder. *Am J Dermatopathol* (2015) 37(8):604–13. doi: 10.1097/DAD.0000000000000266

48. Nicolae A, Pittaluga S, Venkataraman G, Vijnovich-Baron A, Xi L, Raffeld M, et al. Peripheral T-cell lymphomas of follicular T-helper cell derivation with Hodgkin/Reed-sternberg cells of b-cell lineage: Both EBV-positive and EBV-negative variants exist. *Am J Surg Pathol* (2013) 37(6):816–26. doi: 10.1097/PAS.0b013e3182785610

49. Chen HC, Wang RC, Tsai HP, Medeiros LJ, Chang KC. Morphologic spectrum of lymphadenopathy in drug reaction with eosinophilia and systemic symptoms syndrome. *Arch Pathol Lab Med* (2022) 146(9):1084–93. doi: 10.5858/arpa.2021-0087-OA

50. Lee SS, Rudiger T, Odenwald T, Roth S, Starostik P, Muller-Hermelink HK. Angioimmunoblastic T cell lymphoma is derived from mature T-helper cells with varying expression and loss of detectable CD4. *Int J Cancer* (2003) 103(1):12–20. doi: 10.1002/ijc.10758

51. Basha BM, Bryant SC, Rech KL, Feldman AL, Vrana JA, Shi M, et al. Application of a 5 marker panel to the routine diagnosis of peripheral T-cell lymphoma with T-follicular helper phenotype. *Am J Surg Pathol* (2019) 43(9):1282–90. doi: 10.1097/PAS.0000000000001315

52. Gaulard P, de Leval L. Follicular helper T cells: Implications in neoplastic hematopathology. *Semin Diagn Pathol* (2011) 28(3):202–13. doi: 10.1053/j.semdp.2011.03.003

53. Onaindia A, Martinez N, Montes-Moreno S, Almaraz C, Rodriguez-Pinilla SM, Cereceda L, et al. CD30 expression by b and T cells: A frequent finding in angioimmunoblastic T-cell lymphoma and peripheral T-cell lymphoma-not otherwise specified. *Am J Surg Pathol* (2016) 40(3):378–85. doi: 10.1097/PAS.0000000000000571

54. Amador C, Bouska A, Wright G, Weisenburger DD, Feldman AL, Greiner TC, et al. Gene expression signatures for the accurate diagnosis of peripheral T-cell lymphoma entities in the routine clinical practice. *J Clin Oncol* (2022) 40(36):JCO2102707. doi: 10.1200/JCO.21.02707

55. Iqbal J, Weisenburger DD, Greiner TC, Vose JM, McKeithan T, Kucuk C, et al. Molecular signatures to improve diagnosis in peripheral T-cell lymphoma and prognostication in angioimmunoblastic T-cell lymphoma. *Blood* (2010) 115(5):1026–36. doi: 10.1182/blood-2009-06-227579

56. Rodríguez M, Alonso-Alonso R, Tomás-Roca L, Rodríguez-Pinilla SM, Manso-Alonso R, Cereceda L, et al. Peripheral T-cell lymphoma: Molecular profiling recognizes subclasses and identifies prognostic markers. *Blood Adv* (2021) 5(24):5588–98. doi: 10.1182/bloodadvances.2021005171

57. Dupuy A, Lemonnier F, Fataccioli V, Martin-Garcia N, Robe C, Pelletier R, et al. Multiple ways to detect IDH2 mutations in angioimmunoblastic T-cell lymphoma from immunohistochemistry to next-generation sequencing. *J Mol Diagn* (2018) 20(5):677–85. doi: 10.1016/j.jmoldx.2018.05.012

58. Singh A, Schabath R, Ratei R, Stroux A, Klemke CD, Nebe T, et al. Peripheral blood sCD3(-) CD4(+) T cells: A useful diagnostic tool in angioimmunoblastic T cell lymphoma. *Hematol Oncol* (2014) 32(1):16–21. doi: 10.1002/hon.2080

59. Yabe M, Gao Q, Ozkaya N, Huet S, Lewis N, Pichardo JD, et al. Bright PD-1 expression by flow cytometry is a powerful tool for diagnosis and monitoring of angioimmunoblastic T-cell lymphoma. *Blood Cancer J* (2020) 10(3):32. doi: 10.1038/s41408-020-0301-x

60. Loghavi S, Wang SA, Medeiros LJ, Jorgensen JL, Li X, Xu-Monette ZY, et al. Immunophenotypic and diagnostic characterization of angioimmunoblastic T-cell lymphoma by advanced flow cytometric technology. *Leuk Lymphoma* (2016) 57 (12):2804–12. doi: 10.3109/10428194.2016.1170827

61. Liao YL, Chang ST, Kuo SY, Lin SH, Chen CK, Chang KM, et al. Angioimmunoblastic T-cell lymphoma of cytotoxic T-cell phenotype containing a large b-cell proliferation with an undersized b-cell clonal product. *Appl Immunohistochem Mol Morphol* (2010) 18(2):185–9. doi: 10.1097/PAI.0b013e3181c0692b

62. Attygalle AD, Dobson R, Chak PK, Vroobel KM, Wren D, Mugalaasi H, et al. Parallel evolution of two distinct lymphoid proliferations in clonal haematopoiesis. *Histopathology* (2022) 80(5):847–58. doi: 10.1111/his.14619

63. Sakata-Yanagimoto M, Enami T, Yoshida K, Shiraishi Y, Ishii R, Miyake Y, et al. Somatic RHOA mutation in angioimmunoblastic T cell lymphoma. *Nat Genet* (2014) 46 (2):171–5. doi: 10.1038/ng.2872

64. Palomero T, Couronne L, Khiabanian H, Kim MY, Ambesi-Impiombato A, Perez-Garcia A, et al. Recurrent mutations in epigenetic regulators, RHOA and FYN kinase in peripheral T cell lymphomas. *Nat Genet* (2014) 46(2):166–70. doi: 10.1038/ng.2873

65. Nguyen TB, Sakata-Yanagimoto M, Asabe Y, Matsubara D, Kano J, Yoshida K, et al. Identification of cell-type-specific mutations in nodal T-cell lymphomas. *Blood Cancer J* (2017) 7(1):e516. doi: 10.1038/bcj.2016.122

66. Fujisawa M, Sakata-Yanagimoto M, Nishizawa S, Komori D, Gershon P, Kiryu M, et al. Activation of RHOA-VAV1 signaling in angioimmunoblastic T-cell lymphoma. *Leukemia* (2018) 32(3):694–702. doi: 10.1038/leu.2017.273

67. Cortes JR, Ambesi-Impiombato A, Couronne L, Quinn SA, Kim CS, da Silva Almeida AC, et al. RHOA G17V induces T follicular helper cell specification and promotes lymphomagenesis. *Cancer Cell* (2018) 33(2):259–273.e7. doi: 10.1016/j.ccell.2018.01.001

68. Li Z, Cai X, Cai CL, Wang J, Zhang W, Petersen BE, et al. Deletion of Tet2 in mice leads to dysregulated hematopoietic stem cells and subsequent development of myeloid malignancies. *Blood* (2011) 118(17):4509–18. doi: 10.1182/blood-2010-12-325241

69. Cortes JR, Palomero T. The curious origins of angioimmunoblastic T-cell lymphoma. *Curr Opin Hematol* (2016) 23(4):434–43. doi: 10.1097/MOH.0000000000000261

70. Wang C, McKeithan TW, Gong Q, Zhang W, Bouska A, Rosenwald A, et al. IDH2R172 mutations define a unique subgroup of patients with angioimmunoblastic T-cell lymphoma. *Blood* (2015) 126(15):1741–52. doi: 10.1182/blood-2015-05-644591

71. Heavican TB, Bouska A, Yu J, Lone W, Amador C, Gong Q, et al. Genetic drivers of oncogenic pathways in molecular subgroups of peripheral T-cell lymphoma. *Blood* (2019) 133(15):1664–76. doi: 10.1182/blood-2018-09-872549

72. Wang P, Wu J, Ma S, Zhang L, Yao J, Hoadley KA, et al. Oncometabolite d-2-Hydroxyglutarate inhibits ALKBH DNA repair enzymes and sensitizes IDH mutant cells to alkylating agents. *Cell Rep* (2015) 13(11):2353–61. doi: 10.1016/j.celrep.2015.11.029

73. Tahiliani M, Koh KP, Shen Y, Pastor WA, Bandukwala H, Brudno Y, et al. Conversion of 5-methylcytosine to 5-hydroxymethylcytosine in mammalian DNA by MLL partner TET1. *Science* (2009) 324(5929):930–5. doi: 10.1126/science.1170116

74. Ito S, D'Alessio AC, Taranova OV, Hong K, Sowers LC, Zhang Y. Role of tet proteins in 5mC to 5hmC conversion, ES-cell self-renewal and inner cell mass specification. *Nature* (2010) 466(7310):1129–33. doi: 10.1038/nature09303

75. Lemonnier F, Couronne L, Parrens M, Jais JP, Travert M, Lamant L, et al. Recurrent TET2 mutations in peripheral T-cell lymphomas correlate with TFH-like features and adverse clinical parameters. *Blood* (2012) 120(7):1466–9. doi: 10.1182/blood-2012-02-408542

76. Vallois D, Dobay MP, Morin RD, Lemonnier F, Missiaglia E, Juilland M, et al. Activating mutations in genes related to TCR signaling in angioimmunoblastic and other follicular helper T-cell-derived lymphomas. *Blood* (2016) 128(11):1490–502. doi: 10.1182/blood-2016-02-698977

77. Rohr J, Guo S, Huo J, Bouska A, Lachel C, Li Y, et al. Recurrent activating mutations of CD28 in peripheral T-cell lymphomas. *Leukemia* (2016) 30(5):1062–70. doi: 10.1038/leu.2015.357

78. Tiacci E, Venanzi A, Ascani S, Marra A, Cardinali V, Martino G, et al. High-risk clonal hematopoiesis as the origin of AITL and NPM1-mutated AML. *N Engl J Med* (2018) 379(10):981–4. doi: 10.1056/NEJMc1806413

79. Lewis NE, Petrova-Drus K, Huet S, Epstein-Peterson ZD, Gao Q, Sigler AE, et al. Clonal hematopoiesis in angioimmunoblastic T-cell lymphoma with divergent evolution to myeloid neoplasms. *Blood Adv* (2020) 4(10):2261–71. doi: 10.1182/bloodadvances.2020001636

80. Cheng S, Zhang W, Inghirami G, Tam W. Mutation analysis links angioimmunoblastic T-cell lymphoma to clonal hematopoiesis and smoking. *Elife* (2021) 10. doi: 10.7554/eLife.66395

81. Quivoron C, Couronne L, Della Valle V, Lopez CK, Plo I, Wagner-Ballon O, et al. TET2 inactivation results in pleiotropic hematopoietic abnormalities in mouse and is a recurrent event during human lymphomagenesis. *Cancer Cell* (2011) 20(1):25–38. doi: 10.1016/j.ccr.2011.06.003

82. Yao WQ, Wu F, Zhang W, Chuang SS, Thompson JS, Chen Z, et al. Angioimmunoblastic T-cell lymphoma contains multiple clonal T-cell populations derived from a common TET2 mutant progenitor cell. *J Pathol* (2020) 250(3):346–57. doi: 10.1002/path.5376

83. Iqbal J, Wright G, Wang C, Rosenwald A, Gascoyne RD, Weisenburger DD, et al. Gene expression signatures delineate biological and prognostic subgroups in peripheral T-cell lymphoma. *Blood* (2014) 123(19):2915–23. doi: 10.1182/blood-2013-11-536359

84. Iqbal J, Wilcox R, Naushad H, Rohr J, Heavican TB, Wang C, et al. Genomic signatures in T-cell lymphoma: How can these improve precision in diagnosis and inform prognosis? *Blood Rev* (2016) 30(2):89–100. doi: 10.1016/j.blre.2015.08.003

85. Huang Y, Moreau A, Dupuis J, Streubel B, Petit B, Le Gouill S, et al. Peripheral T-cell lymphomas with a follicular growth pattern are derived from follicular helper T cells (TFH) and may show overlapping features with angioimmunoblastic T-cell lymphomas. *Am J Surg Pathol* (2009) 33(5):682–90. doi: 10.1097/PAS.0b013e3181971591

86. Miyoshi H, Sato K, Niino D, Arakawa F, Kimura Y, Kiyasu J, et al. Clinicopathologic analysis of peripheral T-cell lymphoma, follicular variant, and comparison with angioimmunoblastic T-cell lymphoma: Bcl-6 expression might affect progression between these disorders. *Am J Clin Pathol* (2012) 137(6):879–89. doi: 10.1309/AJCPBPNV86VZENGV

87. Streubel B, Vinatzer U, Willheim M, Raderer M, Chott A. Novel t(5;9)(q33;q22) fuses ITK to SYK in unspecified peripheral T-cell lymphoma. *Leukemia* (2006) 20(2):313–8. doi: 10.1038/sj.leu.2404045

88. Debackere K, van der Krogt JA, Tousseyn T, Ferreiro JAF, Van Roosbroeck K, Marcelis L, et al. FER and FES tyrosine kinase fusions in follicular T-cell lymphoma. *Blood* (2020) 135(8):584–8. doi: 10.1182/blood.2019002401

89. Kurita D, Miyoshi H, Yoshida N, Sasaki Y, Kato S, Niino D, et al. A clinicopathologic study of lennert lymphoma and possible prognostic factors: The importance of follicular helper T-cell markers and the association with angioimmunoblastic T-cell lymphoma. *Am J Surg Pathol* (2016) 40(9):1249–60. doi: 10.1097/PAS.0000000000000694

90. Watatani Y, Sato Y, Miyoshi H, Sakamoto K, Nishida K, Gion Y, et al. Molecular heterogeneity in peripheral T-cell lymphoma, not otherwise specified revealed by comprehensive genetic profiling. *Leukemia* (2019) 33(12):2867–83. doi: 10.1038/s41375-019-0473-1

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 09 February 2023
DOI 10.3389/fonc.2023.1101441

OPEN ACCESS

EDITED BY
Jean El Cheikh,
American University of Beirut Medical Center, Lebanon

REVIEWED BY
Reuben Matthew Tooze,
University of Leeds, United Kingdom
Anjali Mishra,
Sidney Kimmel Cancer Center, United States
Brad Haverkos,
University of Colorado Hospital, United States

*CORRESPONDENCE
Ryan A. Wilcox
rywilcox@med.umich.edu

SPECIALTY SECTION
This article was submitted to Hematologic Malignancies, a section of the journal Frontiers in Oncology

RECEIVED 17 November 2022
ACCEPTED 27 January 2023
PUBLISHED 09 February 2023

CITATION
Weiss J, Reneau J and Wilcox RA (2023) PTCL, NOS: An update on classification, risk-stratification, and treatment. *Front. Oncol.* 13:1101441. doi: 10.3389/fonc.2023.1101441

PTCL, NOS: An update on classification, risk-stratification, and treatment

Jonathan Weiss[1], John Reneau[2] and Ryan A. Wilcox[1*]

[1]Department of Internal Medicine, Division of Hematology and Oncology, University of Michigan, Ann Arbor, MI, United States, [2]Department of Medicine, Division of Hematology, The Ohio State University Comprehensive Cancer Center, Columbus, OH, United States

The peripheral T-cell lymphomas (PTCL) are relatively rare, heterogeneous, and therapeutically challenging. While significant therapeutic gains and improved understanding of disease pathogenesis have been realized for selected PTCL subtypes, the most common PTCL in North America remains "not otherwise specified (NOS)" and is an unmet need. However, improved understanding of the genetic landscape and ontogeny for the PTCL subtypes currently classified as PTCL, NOS have been realized, and have significant therapeutic implications, which will be reviewed here.

KEYWORDS

PTCL-NOS, PTCL, GATA-3, risk stratification, classification, treatment

Introduction

Historically, treatment paradigms utilized successfully in the management of many aggressive B-cell lymphomas – namely, multiagent, anthracycline-based regimens, including CHOP – have been empirically applied, perhaps unjustifiably (1), to the T-cell lymphomas. With one notable exception (2), attempts to improve outcomes by "adding to" or "replacing" anthracycline-based regimens have failed, and most T-cell lymphoma patients will succumb to their lymphoma, or complications related to its treatment, within a few years of diagnosis. Indeed, most of the T-cell lymphomas have been, and remain, a challenging and unmet medical need (3).

The sheer geographic, clinical, histopathologic, molecular, and genetic heterogeneity of the more than 25 peripheral T-cell lymphoma (PTCL) subtypes recognized by the WHO have frustrated efforts to improve outcomes for patients afflicted with these mature T-cell derived non-Hodgkin lymphomas (NHL) (4). Not surprisingly then, PTCL diagnosis and classification are challenging, as demonstrated by the relatively high-rate (≈33%) of reclassification following "expert" hematopathology review (5), after which ≈25-40% will remain "unspecified" (6, 7). At the dawn of the 21st century, the PTCL were viewed as the "next, and largely unexplored, frontier in lymphoma management" (8). Twenty years later, significant gains, driven by collaborative and multidisciplinary efforts, have improved our understanding of the PTCL generally, and PTCL, not otherwise specified (PTCL, NOS)

specifically. The new frontier in lymphoma management spawned by these efforts is dominated by novel therapeutic approaches, as evidenced by the more than 100 ongoing clinical trials investigating novel agents. Despite these significant advances, and evidence to support a cautiously optimistic outlook (9), the PTCL, NOS remain a diagnostic and therapeutic challenge, and are the subject of this review.

Epidemiology

In North America and Europe, with the exception of American Indians, PTCL, NOS is the most common PTCL subtype, accounting for ≈30% of PTCL, and is twice as prevalent as either angioimmunoblastic T-cell lymphomas (AITL) or anaplastic large cell lymphomas (ALCL) (6, 7, 10). The median age at diagnosis is 60 years, and males are more commonly affected, with a male-to-female ratio of 1.9:1 (6). A history of celiac disease, psoriasis, and a family history of any hematologic malignancy in a first-degree relative are risk factors for PTCL, NOS, with adjusted odds ratios ranging from ≈2-9, whereas a history of allergies and moderate alcohol consumption are associated with a decreased risk (11). While long-term (>40 years) cigarette smoking is a significant risk factor in PTCL overall, this association did not reach statistical significance for PTCL, NOS (11). Immune suppression may also confer an increased risk of PTCL, NOS, possibly including those derived from cytotoxic T cells (12, 13), and a history of prior immunosuppressive therapies has been reported in 3.8% of PTCL, NOS patients (14). Consistent with the importance of immune surveillance, PTCL, NOS is observed following solid organ transplant, and accounts for approximately one-third of T-cell derived post-transplant lymphoproliferative disorders (15). Deleterious mutations or copy number alterations/structural variants are recurrently observed in genes required for immune surveillance (or evasion), including MHC class I, β2M, and PD-L1 (16, 17). When observed, PTLD (PTCL, NOS) often occur "late" (>5 years post-transplant), frequently involve extranodal sites, and are often, but not always, EBV-associated, regardless of the time of onset following transplant (15), and are seemingly associated with a genetic landscape reminiscent of that observed in PTCL, NOS generally (18). Not surprisingly, T-cell derivation is an adverse prognostic factor on multivariate analysis in the setting of PTLD (19). With the possible exception of PTLD, EBV more commonly affects B-cells within the tumor microenvironment, and rarely infects malignant T cells, at least in the West (14). However, EBV associated PTCL, NOS is more common in Asia, where EBV infection is associated with inferior outcomes (20). While previously classified as a subtype of PTCL, NOS, in the current WHO classification these lymphomas are now classified as nodal EBV-positive T and NK-cell lymphomas (4).

Natural history and risk-stratification

The majority of patients diagnosed with PTCL, NOS present with advanced-stage (III/IV) disease, usually with nodal or nodal/extranodal involvement (14). Involvement of multiple extranodal sites or exclusively extranodal involvement are less common, being observed in 29% and 13% of patients, respectively (14). Common extranodal sites of involvement, observed in at least 10% of patients, include the spleen, bone marrow, liver, and skin (14). Associated complications, including hypercalcemia, hypogammaglobulinemia, and hemophagocytic lymphohistiocytosis (HLH) are observed in fewer than 10% of patients (14).

Approximately 30% of patients remain alive and disease free 24 months following initial diagnosis and treatment (14, 21), and achieving this landmark is a surrogate for overall survival (22, 23). Among PTCL, NOS patients who experienced disease progression or recurrence within 24 months of diagnosis, the 3-year overall survival from the time of progression was 19.4%. In stark contrast, event-free survival at 24 months (EFS24) was associated with a 3-year overall survival, from the time EFS24 was achieved, of 84.6% (22). As with most aggressive NHL, advanced age and poor performance status are adverse prognostic factors (6, 24–26), as are an elevated LDH (14, 24, 25), bone marrow involvement (14, 24), bulky (≥10 cm) disease (14), thrombocytopenia (14), lymphopenia (27), neutrophilia (26), hypoalbuminemia (26), and a high proliferative index (Ki67 ≥80%) (25). These prognostic variables have been variously combined to form prognostic indices, stratifying patients into low- and high-risk groups, with EFS24 ranging from ≈20% to ≈50%, respectively. While still utilized, one suspects that these indices may be supplanted by, or at least incorporate, ontologically and/or genetically based approaches for risk-stratification in the near future.

Disease ontology and classification

PTCL, NOS are morphologically heterogeneous, ranging from sheets of intermediate- to large-sized cells that are relatively devoid of an inflammatory environment (i.e. immunologically "cold") to those that are polymorphous and enriched for a range of inflammatory cells (i.e. immunologically "hot") (28, 29), including epithelioid histiocytes [in "Lennert's lymphomas" (30)]. T-cell specific antigens (e.g. CD2, CD3, CD4) are commonly expressed, while CD4 and/or CD8 expression is variable, and not always associated with the presumed cell of origin (31). The majority (>90%) of PTCL, NOS are derived from mature αβ T-cells. T-cell lymphomas derived from γδ T cells classically involve specific extranodal sites, but when classified as PTCL, NOS, transcriptionally resemble extranodal NK/T cell lymphomas (32).

Early transcriptional profiling efforts highlighted PTCL, NOS heterogeneity, with subsets transcriptionally resembling either ALK-ALCL or AITL, while others were transcriptionally disparate (33, 34). Subsequent transcriptional profiling efforts demonstrated that ≈15% of PTCL, NOS cases transcriptionally resemble follicular helper T-cell (T_{FH})-derived PTCL (35). Consequently, the current WHO classification separates TFH-derived PTCL, previously classified as PTCL, NOS, based on the expression of at least two T_{FH}-associated antigens (i.e. CD10, CD279/PD-1, Bcl-6, CXCL13, ICOS, SAP, or CXCR5) (4, 28).

Consistent with an ontologically informed classification schema, two contemporaneous studies demonstrated that PTCL, NOS may be further sub-classified into those that highly express one of two transcription factors that regulate normal T-cell differentiation. One subset expresses the zinc-finger transcription GATA-3, classically

associated with T-helper type 2 (Th2) differentiation, and are enriched for a number of its canonical transcripts (i.e. target genes). A second, more heterogeneous subtype(s) is characterized by the expression of the transcription factor T-bet (gene name: *TBX21*), which is classically associated with T-helper type 1 (Th1) and cytotoxic T-cell (CTL) differentiation and function. Iqbal et al. adopted an unbiased approach, profiling 121 PTCL, NOS cases, and observed two dominant subtypes upon unsupervised hierarchical clustering, one enriched for GATA-3, and the other enriched for T-bet transcripts (35). Expression of GATA-3 and T-bet transcripts and protein were inversely correlated (35). A gene expression classifier was able to confidently assign 33% of cases to the GATA-3 group and 49% to the T-bet group, and 18% of cases remained unclassifiable. More recently, PTCL,NOS cases included in this study were utilized as a training cohort to identify an abbreviated molecular classifier (36). This abbreviated molecular classifier, which included 153 transcripts (including 21 viral or housekeeping genes), accurately and reproducibly discriminated "unspecified" from "specified" PTCL subtypes, but also discriminated GATA-3 and T-bet PTCL and was generally concordant (concordance 80%) with an immunohistochemistry-based algorithm previously developed by the same group (29). Consistent with prior observations, T-bet PTCL was enriched in cytotoxic T-cell (CTL) related transcripts (35, 37). As the transcripts utilized in the molecular classifiers were not disclosed, interpreting this work within a broader context is challenging. In contrast, Wang et al. adopted a biased approach after observing evidence for cytokine-driven alternative macrophage polarization in PTCL. Transcriptional profiling of cytokines, including those regulated by T-bet (e.g. IFN-γ) and GATA-3 (e.g. IL-4/IL-13), identified two distinct clusters (37). GATA-3 expression, demonstrated by immunohistochemistry, identified a distinct subset in a multicenter cohort of PTCL, NOS patients, and was associated with inferior progression-free and overall survival. In fact, not a single long-term, disease-free survivor was observed in the GATA-3 group (37), as primary refractory disease is frequently observed in these patients (38). Strikingly, a multicenter study failed to observe a significant difference in survival when comparing patients within the GATA-3 group that had been treated with an anthracycline-based regimen (most commonly CHOP or CHOEP) with those that had received supportive care alone (most commonly hospice) [Figure 1 (39)].

The contrasting genetic landscape observed in "GATA-3 PTCL" and "T-bet PTCL" provides further evidence that these are truly distinct PTCL subtypes, and should be classified as such, although the current WHO and ICC classifications view the current evidence as insufficient for such a subclassification (4, 40, 41). For example, copy number gains or amplifications involving c-Myc, STAT3, EZH2, and Rel, among others, were recurrent in (and specific for) the GATA-3 subgroup, whereas losses involving the tumor suppressors p53, PTEN, and CDKN2A/B were observed (42). T-cell receptor signaling plays an important role in upregulating GATA-3 protein expression in both conventional and malignant T cells in a PI3K/AKT dependent manner (38, 43), suggesting that PTEN loss may promote translation of GATA-3 transcripts in these lymphomas. Signaling pathways influenced by the aberrant loci observed included PI3K/mTOR and T-cell receptor dependent signaling, consistent with prior studies (35, 38). The distinct, and "high-risk", genetic landscape associated with GATA-3 PTCL has led some to suggest that GATA-3 does not have an independent oncogenic role, but is simply a surrogate biomarker for a genetically high-risk PTCL subset. However, a comprehensive and systematic analysis of GATA-3 target genes in malignant T cells demonstrated that GATA-3 targets (e.g. including c-myc) are oncogenic, and further demonstrated *via* loss-of-function and gain-of-function studies that GATA-3 is a bona fide proto-oncogene in these lymphomas (39). While less aberrant, distinct and recurrent copy number alterations were observed in T-bet PTCL, including gains involving BCL11B (42), which impairs GATA-3 (Th2) dependent cytokine gene expression upon binding GATA-3 (44). Compared to GATA-3 PTCL, recurrent copy number losses were infrequently observed in T-bet PTCL,

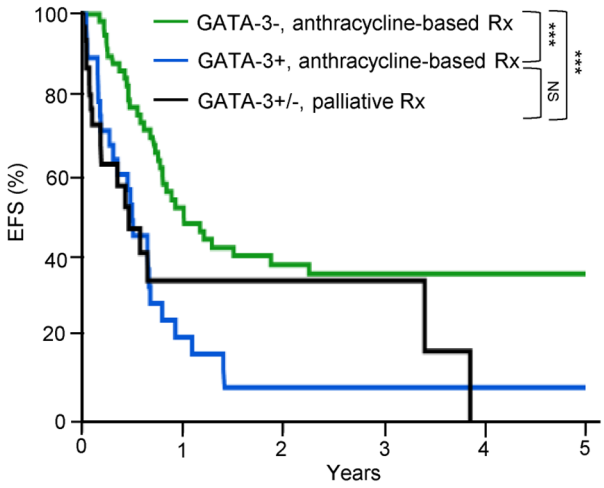


FIGURE 1
GATA-3 expression is associated with dismal outcomes in PTCL, NOS. GATA-3 expression was determined by immunohistochemistry and patients were stratified by treatment. Among GATA-3 positive patients (n=28) that received first-line anthracycline-based chemotherapy, 86% received CHOP or CHOEP. Among GATA-3 negative patients (n=61), 87% were treated with CHOP/CHOEP. Consistent with prior studies, GATA-3 expression was associated with inferior event-free survival (EFS). Importantly, no significant improvement in EFS was observed for GATA-3+ patients treated with CHOP/CHOEP in comparison to PTCL, NOS patients (n=33) who received palliative or best supportive care alone (72% hospice care with or without corticosteroids), highlighting the futility of current therapies in this subset. [Data reprinted with permission from (39)].

although relevant focal losses affecting relevant genes, including TNFAIP3, for example, were occasionally observed. Collectively, the transcriptional and genomic differences observed between GATA-3 and T-bet PTCL demonstrate that these are genetically distinct PTCL subtypes. Furthermore, the high-risk genetic landscape observed in GATA-3 PTCL, including 17p/TP53 deletions and mutations, collectively observed in ≈50% of GATA-3 PTCL, may explain, at least in part, the chemotherapy resistance and dismal outcomes observed in this subgroup.

In contrast to GATA-3 PTCL, the T-bet subgroup is more heterogeneous, and includes minor subsets resembling, and likely derived from, γδ T cells and conventional cytotoxic T cells (CTL) (31–33), including a subset with mutations preferentially affecting epigenetic modifiers, including the DNMT3A methyltransferase domain, leading to hypomethylation of the *EOMES* locus and increased eomesodermin expression, culminating in the induction of a cytotoxic T-cell transcriptional program (31). DNMT3A mutated cases were associated with dismal outcomes, but the inferior survival observed was restricted to T-bet PTCL. While DNMT3A mutations were also observed in GATA-3 PTCL, those mutations did not involve the methyltransferase domain, and were of no prognostic significance. TET2 mutations are also recurrent in T-bet PTCL, and rarely (prevalence <10%) observed in GATA-3 PTCL (36, 45). A rare subset of CTL-derived PTCL, NOS harboring a recurrent IRF4 translocation have also been described (46). In addition, EBV-associated, nodal PTCL, are rarely observed, and usually express CD8 and cytotoxicity-related proteins (45, 47). These findings further highlight the genetic disparities between GATA-3 and T-bet PTCL, but also highlight the heterogeneity within the T-bet subgroup, which is likely comprised of, at the very least, both Th1- and CTL-related lymphomas. Efforts to discriminate these PTCL, NOS subtypes using immunohistochemistry-based algorithms, analogous to those utilized in the classification of diffuse large B-cell lymphoma subtypes, are ongoing (29). The Amador algorithm, for example, includes stains for GATA-3, CCR4, T-bet, and CXCR3, and accurately classified 85% of PTCL, NOS cases when compared with a transcriptionally defined classifier (29). Of course, this ongoing work has significant therapeutic implications, as PTCL, NOS subsets likely have different dependencies, and thus varying degrees of vulnerability to both conventional and novel agents. In fact, the dismal outcomes observed in GATA-3 PTCL, as defined by immunohistochemical staining for GATA-3 alone, following multiagent, anthracycline-based chemotherapy may suggest that this approach is futile in these patients (37, 39, 48, 49). While acknowledging the limitations of making cross-cohort comparisons, it is notable that the outcomes observed in these studies is comparable to those observed in a genetically high-risk subset of PTCL, NOS, defined by TP53 and/or CDKN2A deletions/mutations (>50% of which were biallelic/homozygous) (16). While not reported, the constellation of copy number alterations observed and the prevalent GATA-3 expression reported in this TP53/CDKN2A-altered group (or "group 2", as defined by Watatani et. al.) would suggest that this group, if transcriptionally profiled, would likely be classified as falling within the GATA-3 subtype (16). Given the anticipated prevalence of TP53/CDKN2A alterations in GATA-3 PTCL (≈50%), these findings may further suggest that GATA-3 expression itself is an adverse prognostic factor, irrespective of the underlying genetic landscape (or at least TP53/CDKN2A status). This contention is supported by the recent observation that GATA-3 itself functions as a proto-oncogene (39), and is consistent with its role in the differentiation, homeostatic survival, and proliferation of conventional (non-malignant) T-cell subsets (50–58). These recent findings also have therapeutic implications, as GATA-3 target genes, including ITK, are targetable (39).

PTCL, NOS pathogenesis

The antigen-, costimulatory-, and cytokine-dependent signals that regulate the differentiation, proliferation, survival, and function of conventional (non-malignant) T cells are co-opted by malignant T cells during T-cell lymphomagenesis (Figure 2). Ligands and cytokines, provided by constituents of the tumor microenvironment (TME), instigate signaling cascades that metabolically and transcriptionally regulate malignant T cells. The antigen-, costimulatory-, and cytokine-receptor dependent signaling inputs provided by the TME regulate and activate transcription factors that regulate target genes with either a cell-autonomous and/or non-cell-autonomous role in T-cell lymphomagenesis. Of course, the repertoire of target genes regulated in this manner is not easily extrapolated from our understanding of conventional (non-malignant) T cells, as transcriptional reprogramming is highly context dependent, being determined by the enhancer and epigenetic landscapes, both of which, in comparison to their conventional counterparts, are altered in malignant T cells (16, 42, 59, 60). Target genes with a cell-autonomous role may cooperate with recurrently altered oncogenes (e.g. c-myc, STAT3) and tumor suppressors (e.g. p53, CDKN2A), promoting the growth and survival of malignant T cells. Conversely, target genes with a non-cell-autonomous role (e.g. cytokines/chemokines) regulate the recruitment, homeostatic survival, expansion, and functional polarization of constituents of the TME. These distinctions can be arbitrary, and are by no means mutually exclusive. For example, CSF-1, a critically important homeostatic cytokine for tissue-resident macrophages, may also activate malignant T cells in an autocrine manner upon binding its receptor (i.e. CSF-1R), which is aberrantly expressed by a subset of PTCL, NOS, culminating in PI3K/AKT activation (61). This model of T-cell lymphomagenesis, described as a "three signal" model [Figure 2 (62)], has been recently reviewed (63, 64), and is consistent with the genetic landscape associated with PTCL, NOS, as many of the recurrent copy number variants and mutations observed in these lymphomas regulate signaling cascades normally associated with antigen-, costimulation-, or cytokine-receptor dependent signaling. While many gain-of-function mutations may render cells independent from exogenous, ligand-dependent signaling, this is not universally true, and thus a three-signal model also highlights the TME's supportive role in disease pathogenesis. Conversely, receptors (e.g. Notch) that are rarely subject to mutations or copy number alterations in mature T-cell lymphomas (in contrast to T-ALL) (42, 65), remain dependent upon ligand- and TME-dependent signaling (66). Finally, the signaling cascades triggered by exogenous and TME-dependent factors, and those activated by gain-of-function genetic alterations, confer sensitivity (or resistance) to novel, targeted agents. Consequently,

10.3389/fonc.2023.1101441

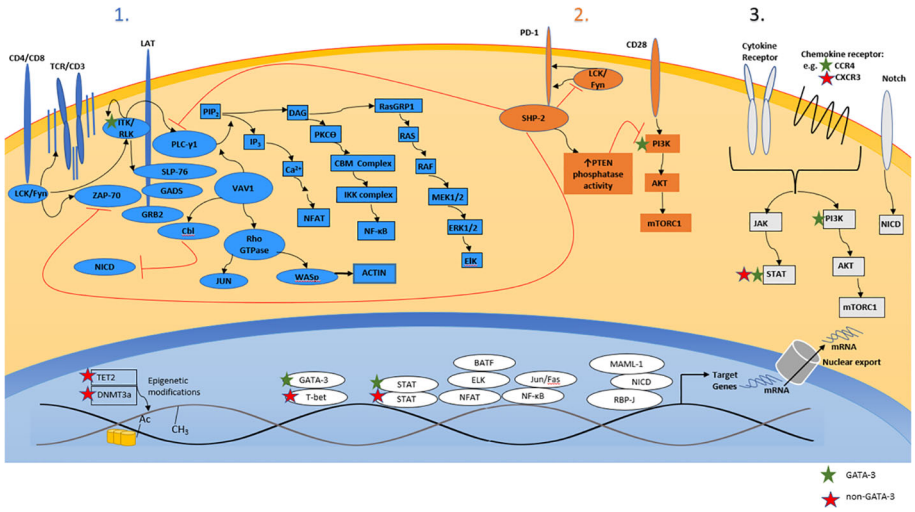


FIGURE 2
A three-signal model of T-cell lymphomagenesis. Ligands and cytokines/chemokines provided by constituents of the tumor microenvironment engage three broad classes of cell-surface receptors expressed by malignant T cells, including the T-cell receptor ("signal 1"), costimulatory/coinhibitory receptors ("signal 2"), and cytokine/chemokine receptors ("signal 3"). Receptor engagement instigates a signaling barrage culminating in significant transcriptional changes that regulate the proliferation, survival, cytoskeletal remodeling, and metabolism of malignant T cells. Relevant primary/secondary messengers and transcription factors are shown, and those that are preferentially expressed (or activated) in GATA-3 or non-GATA-3 PTCL are indicated.

our current, three-signal understanding of PTCL, NOS pathogenesis has significant therapeutic implications.

Analogous to the pathogenic role of "chronic active" and "tonic" B-cell receptor (BCR) signaling in many B-cell lymphomas (67), malignant T cells seemingly exploit T-cell receptor (TCR) dependent signals (38). Just as the genetic landscape observed in B-cell lymphomas supports the BCR's pathogenic role, activating translocations and mutations involving signaling intermediates downstream of the TCR are recurrently observed in PTCL, NOS. For example, gain-of-function mutations in Src family kinases (SFK), including Fyn, are observed in PTCL, NOS (prevalence <5%). These mutations predominantly disrupt inhibitory interactions between the Fyn SH2 domain and its c-terminal domain by rendering Fyn resistant to Csk-dependent phosphorylation (68). More recently, rare translocations involving the SFK genes for Fyn and Lck have been described (69). Among these, a FYN-TRAF3IP2 fusion, which juxtaposes the Fyn membrane localization (and SH3) domains (but not the kinase domain) with almost the complete open reading frame of TRAF3IP2 (including its TRAF6 binding domain), was most prevalent. TRAF3IP2, which normally activates NF-kB and MAPK pathways downstream from IL-17 receptor signaling, activates these pathways in an IL-17 independent manner. When ectopically expressed in a T-ALL cell line, this fusion led to constitutive NF-kB (but not MAPK) activation. Activation of proximal kinases required for TCR signaling (e.g. Zap-70) was not observed, and NF-kB activation occurred independently from formation of the CARD11/BCL10/MALT1 (CBM) complex, yet TCR activation further increased NF-kB activation in these cells. Therefore, while this novel fusion "hijacks" TCR-dependent NF-kB signaling, further amplification of signaling following TCR activation was observed in these cells. In contrast, a novel (and rare) KHDRBS1-LCK fusion, which includes the Lck kinase domain, increased proximal (and SFK-dependent) TCR signaling, resembling "chronic active" antigen-receptor signaling.

Vav1, mutated in ≈10% of PTCL, NOS (16, 70, 71), is a multidomain and multifunctional protein, with both guanine nucleotide exchange factor (GEF)-dependent and GEF-independent functions, that amplifies and diversifies TCR signaling. In addition to preferentially activating the Rho family GTPase Rac1 and remodeling the actin cytoskeleton, Vav1 also activates NFAT and promotes the ubiquitin-dependent degradation of the Notch intracellular domain independently from its GEF activity. In addition, optimal NFAT activation, even in cells that express constitutively active Vav1, requires TCR activation (72). Systematic interrogation of the broad spectrum of Vav1 mutations (and translocations) observed in many cancer types suggest that approximately 50% of Vav1 mutations are non-functional passenger mutations (73). Among the remaining mutations (and translocations) that are functional, a subset (or so-called "trivalent" mutations) activates both Rac1 and NFAT, but also eliminates Notch inhibitory functions associated with wildtype Vav1. As a class, these trivalent mutations are translocations or truncating mutants that involve the c-terminal SH3 domain, and are commonly observed in PTCL, NOS, but are quite distinct from a class of monovalent mutations that, despite eliminating Notch inhibitor capacity, preserve Rac1 and NFAT activating capacity, and are more commonly observed in alternative T-cell lymphomas (including CTCL, AITL, and ATLL). A less common bivalent class of gain-of-function (GOF) missense mutations observed in multiple Vav1 domains culminate in Rac1 and NFAT activation, but do not affect Notch inhibition, and are also observed in PTCL, NOS (and other T-cell lymphomas). Selected Vav1 mutants and fusions lacking the c-terminal SH3 domain, when transgenically expressed in mouse T cells, lead to the development of PTCL resembling nodal T_{FH}-derived lymphomas (73). However, the expression of similarly mutated Vav1 in a p53-deficient context did not lead to the development of PTCL resembling nodal T_{FH}-derived lymphomas (74). Instead, the PTCL that emerged in transgenic mice highly expressed GATA-3 and transcriptionally resembled GATA-3

associated PTCL, NOS, and resembling their human counterparts (42), marked c-Myc pathway activation was also observed in these lymphomas. Collectively, these observations highlight the cooperativity between specific mutations (and presumably the pathways they activate) and the broader genetic landscape across the spectrum of PTCL. To date, very few PTCL, NOS cases, stratified into GATA-3 and non-GATA-3 subtypes, have been sequenced. Therefore, the mutational spectrum, at least within the context of a "three signal" model, and its potential relationship to both the putative cell-of-origin and the broader genetic landscape within these ontologically defined PTCL, NOS subsets, remain a significant gap in knowledge.

Consistent with the three-signal model previously proposed, copy number alterations and GOF mutations are recurrently observed in other TCR (signal 1)-related enzymes and adaptors (e.g. PLCγ, CARD11), and either costimulation (signal 2)-related (e.g. ICOS-CD28 fusions, CD28 mutations) or cytokine (signal 3)-related (e.g. JAK2, JAK3, STAT3, SOCS1) genes (16, 42, 75, 76). As noted, the relationship between this mutational spectrum, PTCL, NOS ontology and classification, and the broader genetic landscape, are not yet fully defined. Furthermore, improved understanding of poorly understood or previously uncharacterized drivers may lead to further additions to, or refinements of, our current understanding of T-cell lymphomagenesis and unveil novel therapeutic targets. For example, ligand-dependent Notch signaling was recently discovered as an important driver of proliferation in PTCL, NOS (66).

Constituents of the TME play a direct role in PTCL pathogenesis, by providing ligands/cytokines that engage antigen, costimulatory, and cytokine receptors expressed by malignant T cells, but also play an indirect role, by suppressing host anti-tumor immunity (62). Macrophages, for example, are constituents of the TME in PTCL, NOS (37, 77), and when abundant are associated with poor outcomes (48). In contrast, abundant B-cells and/or dendritic cells within the TME may be associated with more favorable outcomes (77). PTCL-derived and subtype-specific cytokines promote the recruitment, expansion and functional polarization of TME constituents. Macrophages, for example, are characterized by considerable plasticity, and under the influence of GATA-3 dependent cytokines highly express PD-L1 (17, 37), and other immune checkpoints (77), and foster immune evasion and disease progression (78). Given the importance of the TME in PTCL pathogenesis, novel therapeutic strategies to functionally attenuate, exploit, or even deplete constituents of the TME are being actively investigated. The extent to which the microenvironmental ecosystem diverges across, or is defined by, PTCL, NOS subtypes, and its relative contribution to disease pathogenesis, or utility as a therapeutic target, are poorly understood, but important questions.

Frontline Treatment

As previously noted, the use of anthracycline-based regimens (usually CHOP) in the frontline setting is not only an extrapolation from the treatment paradigm utilized in the management of aggressive B-cell lymphomas, but is not curative for most PTCL, NOS patients. In fact, a recently reported retrospective study demonstrated that survival was dismal for patients with GATA-3 PTCL (as defined by immunohistochemistry) whether they received CHOP/CHOEP or supportive care alone (39). And, more to the point, there was no significant difference between the two groups. Attempts to improve a suboptimal backbone by adding additional agents (79), even a relatively effective one [e.g. brentuximab vedotin (2)], have failed to improve outcomes in PTCL, NOS. For example, the ECHELON-2 trial randomized patients to receive either brentuximab vedotin (BV)-CHP or CHOP. Notably, ≈75% of those enrolled were ALCL patients (2), for whom the overall response rate with single-agent brentuximab vedotin exceeds 80% (80). While a significant survival benefit was observed in the overall study population treated with BV-CHP, this benefit was likely restricted to patients with ALCL. While underpowered to address the potential benefit of BV-CHP in PTCL, NOS, no significant difference in progression-free [hazard ratio: 0.79, 95% CI 0.43-1.43] or overall survival [hazard ratio 0.75, 95% CI 0.37-1.48] was observed among the 72 PTCL, NOS patients randomized (81). Consistent with prior studies (82), CD30 expression among these patients was highly variable (ranging from 10-100%, with a median of 25%) and no correlation with response was observed when using a median cut-point for CD30 expression (81), consistent with prior observations (83).

Consolidation with high-dose therapy followed by autologous stem cell transplantation (HDT-ASCT) in first remission following frontline therapy is associated with ≈40-50% event-free survival at 24 months in single-arm and registry studies (10, 84–86). In contrast, a retrospective study (LYSA) failed to demonstrate a significant benefit associated with HDT-ASCT in an intention-to-treat analysis, but it is notable that 16% within the transplantation arm never proceeded to transplant, often due to primary refractory disease (87). Consequently, anthracycline-based regimens (commonly CHOP or CHOEP), despite uncertainty and a paucity of high-level evidence, and consolidation with high-dose therapy and autologous stem cell transplantation in first remission, remain the cornerstone of treatment for many patients, and is an approach consistent with current treatment guidelines. While this approach may be a "standard of care", it is not curative for most patients, and primary refractory disease remains a significant challenge (38, 39). Therefore, studies investigating combinatorial and rationally designed strategies using novel and immunomodulatory agents with significant single-agent and/or synergistic activity are most certainly needed (1). The transcriptional and genetic heterogeneity increasingly appreciated across the PTCL, NOS spectrum suggests that the identification of relevant biomarkers in future studies ought to facilitate a more personalized approach, and one that will hopefully improve our ability to pair the "right" – and rationally designed – treatment with the "right" patient.

Treatment of relapsed and refractory PTCL, NOS

Outcomes for patients with relapsed/refractory PTCL, NOS are dismal, as a median event-free and overall survival <6 months is reasonably anticipated (88, 89), and salvage therapies, short of HDT-ASCT or allogeneic stem-cell transplantation, are largely palliative. Analogous to the approach adopted in the frontline setting,

traditional salvage regimens used in the setting of relapsed aggressive B-cell lymphomas (e.g. ICE, DHAP) were often utilized in the setting of relapsed/refractory PTCL, NOS prior to the advent of the novel agents (belinostat, brentuximab vedotin, pralatrexate) currently approved in the United States. The British Columbia Cancer Agency reported their experience in relapsed/refractory PTCL, 52% of which were PTCL, NOS, who did not undergo transplant at relapse (82). Among these patients, 58% (n=89) received either combination regimens or single-agent chemotherapy, including GDP (n=19), non-gemcitabine containing multiagent regimens (e.g. ICE, n=22), and various single agents (e.g. etoposide, gemcitabine, n=48). The median overall survival in the entire cohort was 3.7 months, and was only marginally improved for those receiving chemotherapy, at 6.5 months. As this retrospective study included patients treated prior to the widespread availability of novel agents, including pralatrexate and romidepsin, the median progression-free survival (PFS) following salvage chemotherapy was compared to that observed in phase II studies with pralatrexate or romidepsin (90, 91), and median PFS of 3.7, 3.5, and 4 months were observed, respectively (88). Furthermore, exceptional, durable responses may be achieved with novel agents, particularly HDAC inhibitors. Among 130 PTCL patients treated with romidepsin, 10 patients (5 with PTCL, NOS) achieved a durable (>12 months) and complete response, 6 of whom (3 with PTCL, NOS) remained on treatment >2 years (92). Given the favorable toxicity profile associated with novel agents, many patients are exposed to these agents in a sequential manner. The exceptional responses observed in selected patients, and the ability to provide treatment in a sequential manner for many others, may explain the improved overall survival observed with single, novel agents when compared with more toxic, multiagent regimens in retrospective studies (9, 89, 93). For transplant eligible patients who did not undergo consolidation with HDT-ASCT in first remission, HDT-ASCT among those responding to salvage therapies is potentially curative, with 3-year OS ≈50% (94, 95). Allogeneic stem cell transplantation, while associated with significant transplant related-mortality, is also a potentially curative approach for eligible patients (94, 95). Autologous and "off-the-shelf" allogeneic cellular therapies, including CAR-T products specific for T-cell specific antigens, are an active area of ongoing investigation [reviewed in (96)].

Given the advances achieved over the past decade, the PTCL are anything but an "unexplored frontier". In fact, the dramatic expansion in the development of targeted agents, and the sheer number of possible doublet (and triplet) combinations incorporating them, has seemingly outstripped the capacity for their methodical interrogation in a rare disease. This significant challenge is only exacerbated by recognition of the divergent genetic landscape and disease ontogeny between GATA-3 and the more heterogeneous non-GATA-3 subsets, and should be accounted for in future clinical trials using novel agents. While a systematic review of those agents currently under investigation is beyond the scope of the present review, a summary is provided in Table 1. However, it is notable that approximately one-third of the trials

TABLE 1 Novel agents utilized in PTCL, NOS.

Drug	MOA	ORR (PTCL, NOS)	ORR (non-PTCL, NOS)
Bendamustine (97)	Alkylating agent	30/60 (50%)**	AITL 22/32 (69%)
Gemcitabine (98)	Nucleoside analog	11/20 (55%)	MF 9/19 (48%)
5-azacytidine (99)	Nucleoside analog	N/A (not eligible)	AITL 9/12 (75%)
Guadecitabine (100)	Hypomethylating agent	0/2 (0%) 8/20 (40%)***	TFH 7/16 (44%)
Pralatrexate (90)	Dihydrofolate reductase inhibitor	19/59 (32%)	AITL 1/13 (8%) ALCL 6/17(35%)
Fenretinide (101)	Synthetic retinoid	0/1 (0%)	AITL 2/3 (66%) CTCL 2/6 (33%) Gamma-Delta TCLs 0/1 (0%)
Belinostat (102)	Histone deacetylase (HDAC) inhibitor	18/77 (23%)	AITL 10/22 (46%) ALK- ALCL 2/13 (15%) ALK+ ALCL 0/2 (0%)
Romidepsin (91)	Histone deacetylase (HDAC) inhibitor	20/69 (29%)	AITL 8/27 (30%) ALK- ALCL 5/21 (24%)
Tucidinostat (103)	Histone deacetylase (HDAC) inhibitor	12/34 (35%)	AITL 7/8 (87%) ALK- ALCL 1/3 (33%) EATL 1/1 (100%)

(Continued)

TABLE 1 Continued

Drug	MOA	ORR (PTCL, NOS)	ORR (non-PTCL, NOS)
Chidamide (104)	Histone deacetylase (HDAC) inhibitor	6/27 (22%)	ENKL 3/16 (18%) ALK- ALCL 5/11 (45%) ALK+ ALCL 2/6 (33%) AITL 5/10 (50%)
Duvelisib (105)	PI3K inhibitor	8/16 (50%)***	CTCL 6/19 (31.6%)
Linperlisib (106)	PI3K inhibitor	6/12 (50%)	AITL 8/10 (80%)
Tenalisib (107)	PI3K inhibitor	7/15 (46%) ***	CTCL 9/20 (45%)
Copanlisib (108)	PI3K inhibitor	5/10 (50%)***	
Enzastaurin (109)	Serine/threonine kinase inhibitor	0/13 (0%)***	CTCL 1/11 (9%)
Alisertib (110)	Aurora A kinase inhibitor	34/102 (33%)**	
Everolimus (111)	mTORC1 inhibitor	3/4 (75%)	CTCL 3/7 (43%)
Ruxolitinib (112)	Janus Kinase (JAK) inhibitor	2/11 (18%)	TPLL 3/8 (37%) TFH 3/9 (33%) T-LGL 2/5 (40%) ALCL 1/4 (25%) MF 1/7 (14%) Gamma-Delta TCLs 1/4 (25%)
Golidocitinib (113)	Janus Kinase (JAK) inhibitor	5/19 (26%)	AITL 13/20 (65%) ALK- ALCL 2/4 (50%) NKTCL 1/4 (25%)
Cerdulatinib (114)	SYK/JAK inhibitor	9/38 (35%) *	AITL 12/22 (55%)
Cpi-818 (115)	Interleukin-2-Inducible T-cell Kinase (ITK) inhibitor	1/4 (25%)	CTCL 1/7 (14%)
Bortezomib (116)	Proteasome inhibitor	1/2 (50%)	MF 7/10 (70%)
Ixazomib (117)	Proteasome inhibitor	1/2 (50%)	CTCL 0/5 (0%) ALK- ALCL 0/2 (0%) TFH 0/3 (0%)
Tolinapant (118)	Antagonist of the cellular and X-linked inhibitor of apoptosis proteins (cIAP1/2 and XIAP)	21/98 (21%) ***	CTCL 13/50 (26%)
Forodesine (119)	Purine nucleoside phosphorylase inhibitor	5/22 (23%)	AITL 6/18 (33%)
Ibrutinib (120)	Bruton Tyrosine Kinase (BTK) inhibitor	0/3 (0%)	MF 1/6 (16%)
Valemetostat (121)	EZH1/EZH2 inhibitor	24/44 (55%) ***	ATLL 8/14 (57%)
Imatinib mesylate (122)	Platelet derived growth factor inhibitor	0/12 (0%)	
Tipifarnib (123)	Farnesyltransferase inhibitor	4/10 (40%) Note: PTCL CXCL12 chemokine receptor +	AITL 18/32 (56%)
Alemtuzumab (124)	CD52 monoclonal antibody	3/6 (50%)	CTCL 3/4 (75%)
Brentuximab vedotin for CD30+ (83)	CD30 monoclonal antibody-drug conjugate	7/21 (33%) Note: CD30+ PTCL NOS	CD30+ AITL 7/13 (54%)
Camidanlumab tesirine (125)	CD25 monoclonal antibody-drug conjugate	2/6 (33.3%)	ATLL 3/7 (43%)
Mogamulizumab (126)	CCR4 monoclonal antibody	3/16 (19%)	AITL 6/12 (50%), ALK- ALCL 1/1 (100%) CTCL 3/8 (38%)
Pembrolizumab (127)	PD-1 monoclonal antibody	1/5 (20%)	ALCL 1/1(100%) MF 1/3 (33%) FTCL 2/4 (50%) HSTCL 0/1(0%) MEITL 0/1 (0%)

(Continued)

TABLE 1 Continued

Drug	MOA	ORR (PTCL, NOS)	ORR (non-PTCL, NOS)
Nivolumab (128)	PD-1 monoclonal antibody	2/5 (40%) ***	MF 2/13 (15%)
TTI-621 (SIRPα-IgG1 Fc) (129)	Binds CD47	2/11 (18%)***	CTCL 6/29 (21%)
Lenalidomide (130)	Immunomodulation	4/20 (20%)	AITL 8/26 (31%)
Doublets:			
Romidepsin + Pralatrexate (131)		10/14 (71%) **	
Romidepsin + 5-Azacytidine (132)		14/23 (61%) ***	TFH 12/15 (80%)
Romidepsin + Duvelisib (133)		12/22 (55%) ***	CTCL 6/13 (46%)
Romidepsin +Tenalisib (134)		4/5 (80%)	CTCL 8/15 (53%) AITL 4/5 (80%)
Romidepsin + Lenalidomide (135)		5/10 (50%)	CTCL 5/9 (56%)
Duvelisib + Bortezomib (133)		5/14 (36%) ***	CTCL 4/14 (29%)

*Response among all non-AITL subtypes.
**Response among all T-cell lymphomas (PTCL/CTCL).
***Response among all PTCL subtypes.
AITL, Angioimmunoblastic T-cell lymphoma; ALCL, Anaplastic large cell lymphoma; ALK, Anaplastic Lymphoma Kinase; ATLL, Adult T-cell leukemia/lymphoma; CTCL, Cutaneous T-cell lymphoma; EATL, Enteropathy-associated T-cell Lymphoma; ENKL, Extranodal Natural Killer/T-cell lymphoma, nasal type; FTCL, Follicular T-cell lymphoma; Gamma-Delta TCL, Gamma-Delta T-cell lymphoma; HSTCL, Hepatosplenic T-cell lymphoma; MOA, Mechanism of Action; MEITL, Monomorphic epitheliotropic intestinal T-cell lymphoma; MF, Mycosis Fungoides; NKTCL, Natural Killer/T-cell Lymphoma; ORR, Overall response rates; PTCL, NOS, Peripheral T-cell lymphoma not otherwise specified; TFH, Nodal peripheral T-Cell lymphoma, T-follicular helper phenotype; TLGL, T-cell large granular lymphocytic leukemia; T-PLL, T-cell prolymphocytic leukemia.

reviewed failed to discriminate between PTCL, NOS and other PTCL subtypes. Similarly, among the almost 500 PTCL, NOS patients reported in the remaining clinical trials, the distinction between GATA-3 and non-GATA-3 (T-bet) PTCL is unknown. Given the stark – and therapeutically significant – differences between these PTCL subtypes, this obvious gap in knowledge must be addressed in future studies. In contrast to the road previously travelled, the path forward will increasingly rely on an improved understanding of PTCL, NOS pathogenesis, and clinical trial participation – perhaps like never before – will remain a critically important consideration in the management of these patients, as we believe targeted agents and novel combinations targeting both malignant T cells and constituents of their TME are particularly promising.

Author contributions

JW, JR, and RW reviewed the literature, organized and wrote the manuscript. All authors contributed to the article and approved the submitted version.

Funding

This was supported by R37CA233476, R01CA236722, and R01CA265929 (RW)

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Ma H, Marchi E, O'Connor OA. The peripheral T-cell lymphomas: An unusual path to cure. *Lancet Haematology* (2020) 7:e765–71. doi: 10.1016/S2352-3026(20)30207-6

2. Horwitz S, O'Connor OA, Pro B, Illidge T, Fanale M, Advani R, et al. Brentuximab vedotin with chemotherapy for CD30-positive peripheral T-cell lymphoma (ECHELON-2): a global, double-blind, randomised, phase 3 trial. *Lancet* (2019) 393:229–40. doi: 10.1016/S0140-6736(18)32984-2

3. Wilcox RA. Optimising initial treatment for peripheral T-cell lymphoma: A tough nut to CHOP. *Lancet Haematology* (2018) 5:e182–3. doi: 10.1016/S2352-3026(18)30047-4

4. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: Lymphoid neoplasms. *Leukemia* (2022) 36:1720–48. doi: 10.1038/s41375-022-01620-2

5. Laurent C, Baron M, Amara N, Haioun C, Dandoit M, Maynadie M, et al. Impact of expert pathologic review of lymphoma diagnosis: Study of patients from the French lymphopath network. *J Clin Oncol* (2017) 35:2008–17. doi: 10.1200/JCO.2016.71.2083

6. Vose J, Armitage J, Weisenburger D. International peripheral T-cell and natural killer/T-cell lymphoma study: Pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26:4124–30. doi: 10.1200/JCO.2008.16.4558

7. Adams SV, Newcomb PA, Shustov AR. Racial patterns of peripheral T-cell lymphoma incidence and survival in the united states. *J Clin Oncol* (2016) 34:963–71. doi: 10.1200/JCO.2015.63.5540

8. Armitage JO, Vose JM, Weisenburger DD. Towards understanding the peripheral T-cell lymphomas. *Ann Oncol* (2004) 15:1447–9. doi: 10.1093/annonc/mdh409

9. Ma H, Cheng B, Falchi L, Marchi E, Sawas A, Bhagat G, et al. Survival benefit in patients with peripheral T-cell lymphomas after treatments with novel therapies and clinical trials. *Hematol Oncol* (2020) 38:51–8. doi: 10.1002/hon.2705

10. Ellin F, Landstrom J, Jerkeman M, Relander T. Real-world data on prognostic factors and treatment in peripheral T-cell lymphomas: A study from the Swedish lymphoma registry. *Blood* (2014) 124:1570–7. doi: 10.1182/blood-2014-04-573089

11. Wang SS, Flowers CR, Kadin ME, Chang ET, Hughes AM, Ansell SM, et al. Medical history, lifestyle, family history, and occupational risk factors for peripheral T-cell lymphomas: The InterLymph non-Hodgkin lymphoma subtypes project. *J Natl Cancer Inst Monogr* (2014) 2014:66–75. doi: 10.1093/jncimonographs/lgu012

12. Martinez A, Pittaluga S, Villamor N, Colomer D, Rozman M, Raffeld M, et al. Clonal T-cell populations and increased risk for cytotoxic T-cell lymphomas in b-CLL patients: clinicopathologic observations and molecular analysis. *Am J Surg Pathol* (2004) 28:849–58. doi: 10.1097/00000478-200407000-00002

13. Nijland ML, Koens L, Pals ST, Berge I, Bemelman FJ, Kersten MJ. Clinicopathological characteristics of T-cell non-Hodgkin lymphoma arising in patients with immunodeficiencies: A single-center case series of 25 patients and a review of the literature. *Haematologica* (2018) 103:486–96. doi: 10.3324/haematol.2017.169987

14. Weisenburger DD, Savage KJ, Harris NL, Gascoyne RD, Jaffe ES, MacLennan KA, et al. Peripheral T-cell lymphoma, not otherwise specified: A report of 340 cases from the international peripheral T-cell lymphoma project. *Blood* (2011) 117:3402–8. doi: 10.1182/blood-2010-09-310342

15. Swerdlow SH. T-Cell and NK-cell posttransplantation lymphoproliferative disorders. *Am J Clin Pathol* (2007) 127:887–95. doi: 10.1309/LYXN3RGF7D7KPYG0

16. Watatani Y, Sato Y, Miyoshi H, Sakamoto K, Nishida K, Gion Y, et al. Molecular heterogeneity in peripheral T-cell lymphoma, not otherwise specified revealed by comprehensive genetic profiling. *Leukemia* (2019) 33:2867–83. doi: 10.1038/s41375-019-0473-1

17. Wilcox RA, Feldman AL, Wada DA, Yang ZZ, Comfere NI, Dong H, et al. B7-H1 (PD-L1, CD274) suppresses host immunity in T-cell lymphoproliferative disorders. *Blood* (2009) 114:2149–58. doi: 10.1182/blood-2009-04-216671

18. Margolskee E, Jobanputra V, Jain P, Chen J, Ganapathi K, Nahum O, et al. Genetic landscape of T- and NK-cell post-transplant lymphoproliferative disorders. *Oncotarget* (2016) 7:37636–48. doi: 10.18632/oncotarget.9400

19. Caillard S, Porcher R, Provot F, Dantal J, Choquet S, Durrbach A, et al. Post-transplantation lymphoproliferative disorder after kidney transplantation: Report of a nationwide French registry and the development of a new prognostic score. *J Clin Oncol* (2013) 31:1302–9. doi: 10.1200/JCO.2012.43.2344

20. Kim TY, Min GJ, Jeon YW, Park SS, Park S , Shin SH, et al. Impact of Epstein-Barr virus on peripheral T-cell lymphoma not otherwise specified and angioimmunoblastic T-cell lymphoma. *Front Oncol* (2021) 11:797028. doi: 10.3389/fonc.2021.797028

21. Briski R, Feldman AL, Bailey NG, Lim MS, Ristow K, Habermann TM, et al. The role of front-line anthracycline-containing chemotherapy regimens in peripheral T-cell lymphomas. *Blood Cancer J* (2014) 4:e214. doi: 10.1038/bcj.2014.34

22. Maurer MJ, Ellin F, Srour L, Jerkeman M, Bennani NN, Connors JM, et al. International assessment of event-free survival at 24 months and subsequent survival in peripheral T-cell lymphoma. *J Clin Oncol* (2017) 35:4019–26. doi: 10.1200/JCO.2017.73.8195

23. Cederleuf H, Hjort Jakobsen L, Ellin F, de Nully Brown P, Stauffer Larsen T, Bogsted M, et al. Outcome of peripheral T-cell lymphoma in first complete remission: A Danish-Swedish population-based study. *Leuk Lymphoma* (2017) 58:2815–23. doi: 10.1080/10428194.2017.1300888

24. Gallamini A, Stelitano C, Calvi R, Bellei M, Mattei D, Vitolo U, et al. Peripheral T-cell lymphoma unspecified (PTCL-u): A new prognostic model from a retrospective multicentric clinical study. *Blood* (2004) 103:2474–9. doi: 10.1182/blood-2003-09-3080

25. Went P, Agostinelli C, Gallamini A, Piccaluga PP, Ascani S, Sabattini E, et al. Marker expression in peripheral T-cell lymphoma: A proposed clinical-pathologic prognostic score. *J Clin Oncol* (2006) 24:2472–9. doi: 10.1200/JCO.2005.03.6327

26. Federico M, Bellei M, Marcheselli L, Schwartz M, Manni M, Tarantino V, et al. Peripheral T cell lymphoma, not otherwise specified (PTCL-NOS). a new prognostic model developed by the international T cell project network. *Br J Haematol* (2018) 181:760–9. doi: 10.1111/bjh.15258

27. Mitrovic Z, Perry AM, Suzumiya J, Armitage JO, Au WY, Coiffier B, et al. The prognostic significance of lymphopenia in peripheral T-cell and natural killer/T-cell lymphomas: A study of 826 cases from the international peripheral T-cell lymphoma project. *Am J Hematol* (2012) 87:790–4. doi: 10.1002/ajh.23205

28. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, et al. The 2016 revision of the world health organization classification of lymphoid neoplasms. *Blood* (2016) 127:2375–90. doi: 10.1182/blood-2016-01-643569

29. Amador C, Greiner TC, Heavican TB, Smith LM, Galvis KT, Lone W, et al. Reproducing the molecular subclassification of peripheral T-cell lymphoma-NOS by immunohistochemistry. *Blood* (2019) 134:2159–70. doi: 10.1182/blood.2019000779

30. Lennert K, Feller AC, Godde-Salz E. Morphologie, immunhistochemie und genetik peripherer T-Zellen-Lymphome. *Onkologie* (1986) 9:97–107.

31. Herek TA, Bouska A, Lone WG, Sharma S, Amador C, Heavican-Foral TB, et al. DNMT3A mutations define a unique biological and prognostic subgroup associated with cytotoxic T-cells in PTCL-NOS. *Blood* (2022) 140:1278–90. doi: 10.1182/blood.2021015019

32. Iqbal J, Weisenburger DD, Chowdhury A, Tsai MY, Srivastava G, Greiner TC, et al. Natural killer cell lymphoma shares strikingly similar molecular features with a group of non-hepatosplenic gammadelta T-cell lymphoma and is highly sensitive to a novel aurora kinase a inhibitor in vitro. *Leukemia* (2011) 25:348–58. doi: 10.1038/leu.2010.255

33. Iqbal J, Weisenburger DD, Greiner TC, Vose JM, McKeithan T, Kucuk C, et al. Molecular signatures to improve diagnosis in peripheral T-cell lymphoma and prognostication in angioimmunoblastic T-cell lymphoma. *Blood* (2010) 115:1026–36. doi: 10.1182/blood-2009-06-227579

34. Piccaluga PP, Agostinelli C, Califano A, Rossi M, Basso K, Zupo S, et al. Gene expression analysis of peripheral T cell lymphoma, unspecified, reveals distinct profiles and new potential therapeutic targets. *J Clin Invest* (2007) 117:823–34. doi: 10.1172/JCI26833

35. Iqbal J, Wright G, Wang C, Rosenwald A, Gascoyne RD, Weisenburger DD, et al. Gene expression signatures delineate biological and prognostic subgroups in peripheral T-cell lymphoma. *Blood* (2014) 123:2915–23. doi: 10.1182/blood-2013-11-536359

36. Amador C, Bouska A, Wright G, Weisenburger DD, Feldman AL, Greiner TC, et al. Gene expression signatures for the accurate diagnosis of peripheral T-cell lymphoma entities in the routine clinical practice. *J Clin Oncol* (2022) 40:4261–75. doi: 10.1200/JCO.21.02707

37. Wang T, Feldman AL, Wada DA, Lu Y, Polk A, Briski R, et al. GATA-3 expression identifies a high-risk subset of PTCL, NOS with distinct molecular and clinical features. *Blood* (2014) 123:3007–15. doi: 10.1182/blood-2013-12-544809

38. Wang T, Lu Y, Polk A, Chowdhury P, Zamalloa CM, Fujiwara H, et al. T-Cell receptor signaling activates an ITK/NF-kappaB/GATA-3 axis in T-cell lymphomas facilitating resistance to chemotherapy. *Clin Cancer Res* (2017) 23:2506–15. doi: 10.1158/1078-0432.CCR-16-1996

39. Geng X, Wang C, Gao X, Chowdhury P, Weiss JM, Villegas J, et al. GATA-3 is a proto-oncogene in T-cell lymphoproliferative neoplasms. *Blood Cancer Journal* (2022) 12. doi: 10.1038/s41408-022-00745-y

40. Feldman AL, Laurent C, Narbaitz M, Nakamura S, Chan WC, de Leval L, et al. Classification and diagnostic evaluation of nodal T- and NK-cell lymphomas. *Virchows Arch* (2022) 482:265–79. doi: 10.1007/s00428-022-03412-6

41. Campo E, Jaffe ES, Cook JR, Quintanilla-Martinez L, Swerdlow SH, Anderson KC, et al. The international consensus classification of mature lymphoid neoplasms: A report from the clinical advisory committee. *Blood* (2022) 140:1229–53. doi: 10.1182/blood.2022015851

42. Heavican TB, Bouska A, Yu J, Lone W, Amador C, Gong Q, et al. Genetic drivers of oncogenic pathways in molecular subgroups of peripheral T-cell lymphoma. *Blood* (2019) 133:1664–76. doi: 10.1182/blood-2018-09-872549

43. Cook KD, Miller J. TCR-dependent translational control of GATA-3 enhances Th2 differentiation. *J Immunol* (2010) 185:3209–16. doi: 10.4049/jimmunol.0902544

44. Fang D, Gurram Krishna R, Cui K, Zhong C, Hu G, Oler A, et al. Bcl11b, a novel GATA3-interacting protein, regulates T-helper-2-cell differentiation and function. *J Immunol* (2017) 198:223.13. doi: 10.4049/jimmunol.198.Supp.223.13

45. Nicolae A, Bouilly J, Lara D, Fataccioli V, Lemonnier F, Drieux F, et al. Nodal cytotoxic peripheral T-cell lymphoma occurs frequently in the clinical setting of immunodysregulation and is associated with recurrent epigenetic alterations. *Mod Pathol* (2022) 35:1126–36. doi: 10.1038/s41379-022-01022-w

46. Feldman AL, Law M, Remstein ED, Macon WR, Erickson LA, Grogg KL, et al. Recurrent translocations involving the IRF4 oncogene locus in peripheral T-cell lymphomas. *Leukemia* (2009) 23:574–80. doi: 10.1038/leu.2008.320

47. Ng SB, Chung TH, Kato S, Nakamura S, Takahashi E, Ko YH, et al. Epstein-Barr Virus-associated primary nodal T/NK-cell lymphoma shows a distinct molecular signature and copy number changes. *Haematologica* (2018) 103:278–87. doi: 10.3324/haematol.2017.180430

48. Zhang W, Wang Z, Luo Y, Zhong D, Luo Y, Zhou D. GATA3 expression correlates with poor prognosis and tumor-associated macrophage infiltration in peripheral T cell lymphoma. *Oncotarget* (2016) 7:65284–94. doi: 10.18632/oncotarget.11673

49. Manso R, Bellas C, Martin-Acosta P, Mollejo M, Menarguez J, Rojo F, et al. C-MYC is related to GATA3 expression and associated with poor prognosis in nodal peripheral T-cell lymphomas. *Haematologica* (2016) 101:e336–8. doi: 10.3324/haematol.2016.143768

50. Hosoya T, Kuroha T, Moriguchi T, Cummings D, Maillard I, Lim KC, et al. GATA-3 is required for early T lineage progenitor development. *J Exp Med* (2009) 206:2987–3000. doi: 10.1084/jem.20090934

51. Hendriks RW, Nawijn MC, Engel JD, van Doorninck H, Grosveld F, Karis A. Expression of the transcription factor GATA-3 is required for the development of the earliest T cell progenitors and correlates with stages of cellular proliferation in the thymus. *Eur J Immunol* (1999) 29:1912–8. doi: 10.1002/(SICI)1521-4141(199906)29:06<1912::AID-IMMU1912>3.0.CO;2-D

52. Ting CN, Olson MC, Barton KP, Leiden JM. Transcription factor GATA-3 is required for development of the T-cell lineage. *Nature* (1996) 384:474–8. doi: 10.1038/384474a0

53. Wang L, Wildt KF, Zhu J, Zhang X, Feigenbaum L, Tessarollo L, et al. Distinct functions for the transcription factors GATA-3 and ThPOK during intrathymic differentiation of CD4(+) T cells. *Nat Immunol* (2008) 9:1122–30. doi: 10.1038/ni.1647

54. Zhu J, Min B, Hu-Li J, Watson CJ, Grinberg A, Wang Q, et al. Conditional deletion of Gata3 shows its essential function in T(H)1-T(H)2 responses. *Nat Immunol* (2004) 5:1157–65. doi: 10.1038/ni1128

55. Zhu J, Yamane H, Cote-Sierra J, Guo L, Paul WE. GATA-3 promotes Th2 responses through three different mechanisms: Induction of Th2 cytokine production, selective growth of Th2 cells and inhibition of Th1 cell-specific factors. *Cell Res* (2006) 16:3–10. doi: 10.1038/sj.cr.7310002

56. Wang Y, Misumi I, Gu AD, Curtis TA, Su L, Whitmire JK, et al. GATA-3 controls the maintenance and proliferation of T cells downstream of TCR and cytokine signaling. *Nat Immunol* (2013) 14:714–22. doi: 10.1038/ni.2623

57. Wang Y, Su MA, Wan YY. An essential role of the transcription factor GATA-3 for the function of regulatory T cells. *Immunity* (2011) 35:337–48. doi: 10.1016/j.immuni.2011.08.012

58. Yu F, Sharma S, Edwards J, Feigenbaum L, Zhu J. Dynamic expression of transcription factors T-bet and GATA-3 by regulatory T cells maintains immunotolerance. *Nat Immunol* (2015) 16:197–206. doi: 10.1038/ni.3053

59. Vahedi G, Takahashi H, Nakayamada S, Sun HW, Sartorelli V, Kanno Y, et al. STATs shape the active enhancer landscape of T cell populations. *Cell* (2012) 151:981–93. doi: 10.1016/j.cell.2012.09.044

60. Hosoya T, D'Oliveira Albanus R, Hensley J, Myers G, Kyono Y, Kitzman J, et al. Global dynamics of stage-specific transcription factor binding during thymocyte development. *Sci Rep* (2018) 8:5605. doi: 10.1038/s41598-018-23774-9

61. Murga-Zamalloa C, Rolland DCM, Polk A, Wolfe A, Dewar H, Chowdhury P, et al. Colony-stimulating factor 1 receptor (CSF1R) activates AKT/mTOR signaling and promotes T-cell lymphoma viability. *Clin Cancer Res* (2019) 26:690–703. doi: 10.1158/1078-0432.CCR-19-1486

62. Wilcox RA. A three-signal model of T-cell lymphoma pathogenesis. *Am J Hematol* (2016) 91:113–22. doi: 10.1002/ajh.24203

63. Fiore D, Cappelli LV, Broccoli A, Zinzani PL, Chan WC, Inghirami G. Peripheral T cell lymphomas: From the bench to the clinic. *Nat Rev Cancer* (2020) 20:323–42. doi: 10.1038/s41568-020-0247-0

64. Pizzi M, Margolskee E, Inghirami G. Pathogenesis of peripheral T cell lymphoma. *Annu Rev Pathol* (2018) 13:293–320. doi: 10.1146/annurev-pathol-020117-043821

65. Brandstadter JD, Maillard I. Notch signalling in T cell homeostasis and differentiation. *Open Biol* (2019) 9:190187. doi: 10.1098/rsob.190187

66. Gao X, Wang C, Abdelrahman S, Kady N, Murga-Zamalloa C, Gann P, et al. Notch signaling promotes mature T-cell lymphomagenesis. *Cancer Res* (2022) 82:3763–73. doi: 10.1158/0008-5472.CAN-22-1215

67. Young RM, Phelan JD, Wilson WH, Staudt LM. Pathogenic b-cell receptor signaling in lymphoid malignancies: New insights to improve treatment. *Immunol Rev* (2019) 291:190–213. doi: 10.1111/imr.12792

68. Palomero T, Couronne L, Khiabanian H, Kim MY, Ambesi-Impiombato A, Perez-Garcia A, et al. Recurrent mutations in epigenetic regulators, RHOA and FYN kinase in peripheral T cell lymphomas. *Nat Genet* (2014) 46:166–70. doi: 10.1038/ng.2873

69. Debackere K, Marcelis L, Demeyer S, Vanden Bempt M, Mentens N, Gielen O, et al. Fusion transcripts FYN-TRAF3IP2 and KHDRBS1-LCK hijack T cell receptor signaling in peripheral T-cell lymphoma, not otherwise specified. *Nat Commun* (2021) 12:3705. doi: 10.1038/s41467-021-24037-4

70. Boddicker RL, Razidlo GL, Dasari S, Zeng Y, Hu G, Knudson RA, et al. Integrated mate-pair and RNA sequencing identifies novel, targetable gene fusions in peripheral T-cell lymphoma. *Blood* (2016) 128:1234–45. doi: 10.1182/blood-2016-03-707141

71. Abate F, da Silva-Almeida AC, Zairis S, Robles-Valero J, Couronne L, Khiabanian H, et al. Activating mutations and translocations in the guanine exchange factor VAV1 in peripheral T-cell lymphomas. *Proc Natl Acad Sci U.S.A.* (2017) 114:764–9. doi: 10.1073/pnas.1608839114

72. Barreira M, Fabbiano S, Couceiro JR, Torreira E, Martinez-Torrecuadrada JL, Montoya G, et al. The c-terminal SH3 domain contributes to the intramolecular inhibition of vav family proteins. *Sci Signal* (2014) 7:ra35. doi: 10.1126/scisignal.2004993

73. Robles-Valero J, Fernandez-Nevado L, Lorenzo-Martin LF, Cuadrado M, Fernandez-Pisonero I, Rodriguez-Fdez S, et al. Cancer-associated mutations in VAV1 trigger variegated signaling outputs and T-cell lymphomagenesis. *EMBO J* (2021) 40:e108125. doi: 10.15252/embj.2021108125

74. Fukumoto K, Sakata-Yanagimoto M, Fujisawa M, Sakamoto T, Miyoshi H, Suehara Y, et al. VAV1 mutations contribute to development of T-cell neoplasms in mice. *Blood* (2020) 136:3018–32. doi: 10.1182/blood.2020006513

75. Rohr J, Guo S, Huo J, Bouska A, Lachel C, Li Y, et al. Recurrent activating mutations of CD28 in peripheral T-cell lymphomas. *Leukemia* (2016) 30:1062–70. doi: 10.1038/leu.2015.357

76. Vallois D, Dobay MP, Morin RD, Lemonnier F, Missiaglia E, Juilland M, et al. Activating mutations in genes related to TCR signaling in angioimmunoblastic and other follicular helper T-cell-derived lymphomas. *Blood* (2016) 128:1490–502. doi: 10.1182/blood-2016-02-698977

77. Sugio T, Miyawaki K, Kato K, Sasaki K, Yamada K, Iqbal J, et al. Microenvironmental immune cell signatures dictate clinical outcomes for PTCL-NOS. *Blood Adv* (2018) 2:2242–52. doi: 10.1182/bloodadvances.2018018754

78. Cortes JR, Filip I, Albero R, Patino-Galindo JA, Quinn SA, Lin WW, et al. Oncogenic Vav1-Myo1f induces therapeutically targetable macrophage-rich tumor microenvironment in peripheral T cell lymphoma. *Cell Rep* (2022) 39:110695. doi: 10.1016/j.celrep.2022.110695

79. Schmitz N, Trumper L, Ziepert M, Nickelsen M, Ho AD, Metzner B, et al. Treatment and prognosis of mature T-cell and NK-cell lymphoma: An analysis of patients with T-cell lymphoma treated in studies of the German high-grade non-Hodgkin lymphoma study group. *Blood* (2010) 116:3418–25. doi: 10.1182/blood-2010-02-270785

80. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, et al. Brentuximab vedotin (SGN-35) in patients with relapsed or refractory systemic anaplastic large-cell lymphoma: Results of a phase II study. *J Clin Oncol* (2012) 30:2190–6. doi: 10.1200/JCO.2011.38.0402

81. Horwitz S, O'Connor OA, Pro B, Trumper L, Iyer S, Advani R, et al. The ECHELON-2 trial: 5-year results of a randomized, phase III study of brentuximab vedotin with chemotherapy for CD30-positive peripheral T-cell lymphoma. *Ann Oncol* (2022) 33:288–98. doi: 10.1016/j.annonc.2021.12.002

82. Karube K, Kakimoto Y, Tonozuka Y, Ohshima K. The expression of CD30 and its clinico-pathologic significance in peripheral T-cell lymphomas. *Expert Rev Hematol* (2021) 14:777–87. doi: 10.1080/17474086.2021.1955344

83. Horwitz SM, Advani RH, Bartlett NL, Jacobsen ED, Sharman JP, O'Connor OA, et al. Objective responses in relapsed T-cell lymphomas with single-agent brentuximab vedotin. *Blood* (2014) 123:3095–100. doi: 10.1182/blood-2013-12-542142

84. d'Amore F, Relander T, Lauritzsen GF, Jantunen E, Hagberg H, Anderson H, et al. Up-front autologous stem-cell transplantation in peripheral T-cell lymphoma: NLG-T-01. *J Clin Oncol* (2012) 30:3093–9. doi: 10.1200/JCO.2011.40.2719

85. Park SI, Horwitz SM, Foss FM, Pinter-Brown LC, Carson KR, Rosen ST, et al. The role of autologous stem cell transplantation in patients with nodal peripheral T-cell lymphomas in first complete remission: Report from COMPLETE, a prospective, multicenter cohort study. *Cancer* (2019) 125:1507–17. doi: 10.1002/cncr.31861

86. Schmitz N, Truemper L, Bouabdallah K, Ziepert M, Leclerc M, Cartron G, et al. A randomized phase 3 trial of autologous vs allogeneic transplantation as part of first-line therapy in poor-risk peripheral T-NHL. *Blood* (2021) 137:2646–56. doi: 10.1182/blood.2020008825

87. Fossard G, Broussais F, Coelho I, Bailly S, Nicolas-Virelizier E, Toussaint E, et al. Role of up-front autologous stem-cell transplantation in peripheral T-cell lymphoma for patients in response after induction: An analysis of patients from LYSA centers. *Ann Oncol* (2018) 29:715–23. doi: 10.1093/annonc/mdx787

88. Mak V, Hamm J, Chhanabhai M, Shenkier T, Klasa R, Sehn LH, et al. Survival of patients with peripheral T-cell lymphoma after first relapse or progression: spectrum of disease and rare long-term survivors. *J Clin Oncol* (2013) 31:1970–6. doi: 10.1200/JCO.2012.44.7524

89. Zhang JY, Briski R, Devata S, Kaminski MS, Phillips TJ, Mayer TL, et al. Survival following salvage therapy for primary refractory peripheral T-cell lymphomas (PTCL). *Am J Hematol* (2018) 93:394–400. doi: 10.1002/ajh.24992

90. O'Connor OA, Pro B, Pinter-Brown L, Bartlett N, Popplewell L, Coiffier B, et al. Pralatrexate in patients with relapsed or refractory peripheral T-cell lymphoma: Results from the pivotal PROPEL study. *J Clin Oncol* (2011) 29:1182–9. doi: 10.1200/JCO.2010.29.9024

91. Coiffier B, Pro B, Prince HM, Foss F, Sokol L, Greenwood M, et al. Results from a pivotal, open-label, phase II study of romidepsin in relapsed or refractory peripheral T-cell lymphoma after prior systemic therapy. *J Clin Oncol* (2012) 30:631–6. doi: 10.1200/JCO.2011.37.4223

92. Coiffier B, Pro B, Prince HM, Foss F, Sokol L, Greenwood M, et al. Romidepsin for the treatment of relapsed/refractory peripheral T-cell lymphoma: Pivotal study update demonstrates durable responses. *J Hematol Oncol* (2014) 7:11. doi: 10.1186/1756-8722-7-11

93. Stuver RN, Khan N, Schwartz M, Acosta M, Federico M, Gisselbrecht C, et al. Single agents vs combination chemotherapy in relapsed and refractory peripheral T-cell lymphoma: Results from the comprehensive oncology measures for peripheral T-cell lymphoma treatment (COMPLETE) registry. *Am J Hematol* (2019) 94:641–9. doi: 10.1002/ajh.25463

94. Smith SM, Burns LJ, van Besien K, Lerademacher J, He W, Fenske TS, et al. Hematopoietic cell transplantation for systemic mature T-cell non-Hodgkin lymphoma. *J Clin Oncol* (2013) 31:3100–9. doi: 10.1200/JCO.2012.46.0188

95. Kameda K, Kako S, Kim SW, Usui Y, Kato K, Fukuda T, et al. Autologous or allogeneic hematopoietic cell transplantation for relapsed or refractory PTCL-NOS or AITL. *Leukemia* (2022) 36:1361–70. doi: 10.1038/s41375-022-01545-w

96. Safarzadeh Kozani P, Safarzadeh Kozani P, Rahbarizadeh F. CAR-T cell therapy in T-cell malignancies: Is success a low-hanging fruit? *Stem Cell Res Ther* (2021) 12:527. doi: 10.3389/fimmu.2021.765097

97. Damaj G, Gressin R, Bouabdallah K, Cartron G, Choufi B, Gyan E, et al. Results from a prospective, open-label, phase II trial of bendamustine in refractory or relapsed T-

cell lymphomas: The BENTLY trial. *J Clin Oncol* (2013) 31:104–10. doi: 10.1200/JCO.2012.43.7285

98. Zinzani PL, Magagnoli M, Bendandi M, Orcioni GF, Gherlinzoni F, Albertini P, et al. Therapy with gemcitabine in pretreated peripheral T-cell lymphoma patients. *Ann Oncol* (1998) 9:1351–3. doi: 10.1023/A:1008409601731

99. Lemonnier F, Dupuis J, Sujobert P, Tournillhac O, Cheminant M, Sarkozy C, et al. Treatment with 5-azacytidine induces a sustained response in patients with angioimmunoblastic T-cell lymphoma. *Blood* (2018) 132:2305–9. doi: 10.1182/blood-2018-04-840538

100. Wong J, Gruber E, Maher B, Waltham M, Sabouri-Thompson Z, Jong I, et al. Integrated clinical and genomic evaluation of guadecitabine (SGI-110) in peripheral T-cell lymphoma. *Leukemia* (2022) 36:1654–65. doi: 10.1038/s41375-022-01571-8

101. Mohrbacher AM, Yang AS, Groshen S, Kummar S, Gutierrez ME, Kang MH, et al. Phase I study of fenretinide delivered intravenously in patients with relapsed or refractory hematologic malignancies: A California cancer consortium trial. *Clin Cancer Res* (2017) 23:4550–5. doi: 10.1158/1078-0432.CCR-17-0234

102. O'Connor OA, Horwitz S, Masszi T, Van Hoof A, Brown P, Doorduijn J, et al. Belinostat in patients with relapsed or refractory peripheral T-cell lymphoma: Results of the pivotal phase II BELIEF (CLN-19) study. *J Clin Oncol* (2015) 33:2492–9. doi: 10.1200/JCO.2014.59.2782

103. Kim WS, Rai S, Ando K, Choi I, Izutsu K, Tsukamoto N, et al. A phase 2b open-label single arm study to evaluate the efficacy and safety of hbi-8000 (tucidinostat) in patients with relapsed or refractory peripheral t-cell lymphoma (ptcl). *Hematological Oncol 39* (2021). doi: 10.1002/hon.121_2880

104. Shi Y, Dong M, Hong X, Zhang W, Feng J, Zhu J, et al. Results from a multicenter, open-label, pivotal phase II study of chidamide in relapsed or refractory peripheral T-cell lymphoma. *Ann Oncol* (2015) 26:1766–71. doi: 10.1093/annonc/mdv237

105. Horwitz SM, Koch R, Porcu P, Oki Y, Moskowitz A, Perez M, et al. Activity of the PI3K-delta,gamma inhibitor duvelisib in a phase 1 trial and preclinical models of T-cell lymphoma. *Blood* (2018) 131:888–98. doi: 10.1182/blood-2017-08-802470

106. Jin J, Cen H, Zhou K, Xu X, Li F, Wu T, et al. A phase ib study of a PI3Kδ inhibitor linperlisib in patients with relapsed or refractory peripheral T-cell lymphoma. *J Clin Oncol* (2021) 39:7531–1. doi: 10.1200/JCO.2021.39.15_suppl.7531

107. Huen A, Haverkos BM, Zain J, Radhakrishnan R, Lechowicz MJ, Devata S, et al. Phase I/Ib study of tenalisib (RP6530), a dual PI3K delta/gamma inhibitor in patients with Relapsed/Refractory T-cell lymphoma. *Cancers (Basel)* (2020) 12. doi: 10.3390/cancers12082293

108. Dreyling M, Morschhauser F, Bouabdallah K, Bron D, Cunningham D, Assouline SE, et al. Phase II study of copanlisib, a PI3K inhibitor, in relapsed or refractory, indolent or aggressive lymphoma. *Ann Oncol* (2017) 28:2169–78. doi: 10.1093/annonc/mdx289

109. Forsyth CJ, Gomez-Almaguer D, Camargo JF, Eliadis PE, Crespo-Solis E, Pereira J, et al. A multicenter, open-label, noncomparative screening study of enzastaurin in adult patients with non-Hodgkin lymphomas. *Clin Lymphoma Myeloma Leuk* (2013) 13:398–403. doi: 10.1016/j.clml.2013.03.005

110. O'Connor OA, Ozcan M, Jacobsen ED, Roncero JM, Trotman J, Demeter J, et al. Randomized phase III study of alisertib or investigator's choice (Selected single agent) in patients with relapsed or refractory peripheral T-cell lymphoma. *J Clin Oncol* (2019) 37:613–23. doi: 10.1200/JCO.18.00899

111. Witzig TE, Reeder C, Han JJ, LaPlant B, Stenson M, Tun HW, et al. The mTORC1 inhibitor everolimus has antitumor activity *in vitro* and produces tumor responses in patients with relapsed T-cell lymphoma. *Blood* (2015) 126:328–35. doi: 10.1182/blood-2015-02-629543

112. Moskowitz AJ, Ghione P, Jacobsen E, Ruan J, Schatz JH, Noor S, et al. A phase 2 biomarker-driven study of ruxolitinib demonstrates effectiveness of JAK/STAT targeting in T-cell lymphomas. *Blood* (2021) 138:2828–37. doi: 10.1182/blood.2021013379

113. Kim WS, Yoon DH, Song Y, Yang H, Cao J, Ji D, et al. A phase I/II study of golidocitinib, a selective JAK1 inhibitor, in refractory or relapsed peripheral T-cell lymphoma. *J Clin Oncol* (2022) 40:7563–3. doi: 10.1200/JCO.2022.40.16_suppl.7563

114. Horwitz SM, Feldman T, Hess B, Khodadoust M, Youn K, Munoz J, et al. (2019). A phase 2 study of the dual SYK/JAK inhibitor cerdulatinib demonstrates good tolerability and clinical response in Relapsed/Refractory peripheral T-cell lymphoma and cutaneous T-cell lymphoma. Blood 134(Supplement 1): 466. doi: 10.1182/blood-2019-123986

115. Khodadoust MS, Feldman TA, Yoon DH, Yannakou CK, Radeski D, Kim YH, et al. Cpi-818, an oral interleukin-2-Inducible T-cell kinase inhibitor, is well-tolerated and active in patients with T-cell lymphoma. *Blood* (2020) 136:19–20. doi: 10.1182/blood-2020-137782

116. Zinzani PL, Musuraca G, Tani M, Stefoni V, Marchi E, Fina M, et al. Phase II trial of proteasome inhibitor bortezomib in patients with relapsed or refractory cutaneous T-cell lymphoma. *J Clin Oncol* (2007) 25:4293–7. doi: 10.1200/JCO.2007.11.4207

117. Boonstra PS, Polk A, Brown N, Hristov AC, Bailey NG, Kaminski MS, et al. A single center phase II study of ixazomib in patients with relapsed or refractory cutaneous or peripheral T-cell lymphomas. *Am J Hematol* (2017) 92:1287–94. doi: 10.1002/ajh.24895

118. Michot JM, Mehta A, Samaniego F, Bachy E, Zinzani PL, Prica A, et al. (2022). Preliminary analysis of the phase ii study using tolinapant (astx660) monotherapy in 98 peripheral t-cell lymphoma and 51 cutaneous t-cell lymphoma subjects with relapsed refractory disease, in: *Presented at the 27th Congress of the European Hematology Association*, (Congress of the European Hematology Association).

119. Makita S, Maeshima AM, Maruyama D, Izutsu K, Tobinai K. Forodesine in the treatment of relapsed/refractory peripheral T-cell lymphoma: An evidence-based review. *OncoTargets Ther* (2018) 11:2287–93. doi: 10.2147/OTT.S140756

120. Kumar A, Vardhana S, Moskowitz AJ, Porcu P, Dogan A, Dubovsky JA, et al. Pilot trial of ibrutinib in patients with relapsed or refractory T-cell lymphoma. *Blood Adv* (2018) 2:871–6. doi: 10.1182/bloodadvances.2017011916

121. Kusumoto S, Izutsu K, Maruyuma D, Makita S, E. jacobsen HSM, K. Ishitsuka P, et al. (2021). First-in-human study of the ezh1 and ezh2 dual inhibitor valemetostat tosylate (ds-3201b) in patients with relapsed or refractory non-hodgkin lymphomas, in: *Presented at the Congress of the European Hematology Association*, (Congress of the European Hematology Association).

122. Jacobsen E, Pozdnyakova O, Redd R, Fisher DC, Dorfman DM, Dal Cin P, et al. Imatinib mesylate lacks efficacy in relapsed/refractory peripheral T cell lymphoma. *Leuk Lymphoma* (2015) 56:993–8. doi: 10.3109/10428194.2014.941835

123. Witzig TE, Sokol L, Kim WS, de la Cruz Vicente F, Caballero D, Advani R, et al. Final results from a phase 2 study of tipifarnib in subjects with relapsed or refractory peripheral T-cell lymphoma. *Blood* (2021) 138:621. doi: 10.1182/blood-2021-147279

124. Zinzani PL, Alinari L, Tani M, Fina M, Pileri S, Baccarani M. Preliminary observations of a phase II study of reduced-dose alemtuzumab treatment in patients with pretreated T-cell lymphoma. *Haematologica* (2005) 90:702–3.

125. Hamadani M, Collins GP, Caimi PF, Samaniego F, Spira A, Davies A, et al. Camidanlumab tesirine in patients with relapsed or refractory lymphoma: A phase 1, open-label, multicentre, dose-escalation, dose-expansion study. *Lancet Haematol* (2021) 8:e433–45. doi: 10.1016/S2352-3026(21)00103-4

126. Ogura M, Ishida T, Hatake K, Taniwaki M, Ando K, Tobinai K, et al. Multicenter phase II study of mogamulizumab (KW-0761), a defucosylated anti-cc chemokine receptor 4 antibody, in patients with relapsed peripheral T-cell lymphoma and cutaneous T-cell lymphoma. *J Clin Oncol* (2014) 32:1157–63. doi: 10.1200/JCO.2013.52.0924

127. Barta SK, Zain J, A.W.t. MacFarlane SM, Ruan J, Fung HC, Tan CR, et al. Phase II study of the PD-1 inhibitor pembrolizumab for the treatment of relapsed or refractory mature T-cell lymphoma. *Clin lymphoma myeloma leukemia* (2019) 19:356–64. doi: 10.1016/j.clml.2019.03.022

128. Lesokhin AM, Ansell SM, Armand P, Scott EC, Halwani A, Gutierrez M, et al. Nivolumab in patients with relapsed or refractory hematologic malignancy: Preliminary results of a phase ib study. *J Clin Oncol* (2016) 372:311–9. doi: 10.1200/JCO.2015.65.9789

129. Ansell SM, Maris MB, Lesokhin AM, Chen RW, Flinn IW, Sawas A, et al. Phase I study of the CD47 blocker TTI-621 in patients with relapsed or refractory hematologic malignancies. *Clin Cancer Res* (2021) 27:2190–9. doi: 10.1158/1078-0432.CCR-20-3706

130. Morschhauser F, Fitoussi O, Haioun C, Thieblemont C, Quach H, Delarue R, et al. A phase 2, multicentre, single-arm, open-label study to evaluate the safety and efficacy of single-agent lenalidomide (Revlimid) in subjects with relapsed or refractory peripheral T-cell non-Hodgkin lymphoma: The EXPECT trial. *Eur J Cancer* (2013) 49:2869–76. doi: 10.1016/j.ejca.2013.04.029

131. Amengual JE, Lichtenstein R, Lue J, Sawas A, Deng C, Lichtenstein E, et al. A phase 1 study of romidepsin and pralatrexate reveals marked activity in relapsed and refractory T-cell lymphoma. *Blood* (2018) 131:397–407. doi: 10.1182/blood-2017-09-806737

132. Falchi L, Ma H, Klein S, Lue JK, Montanari F, Marchi E, et al. Combined oral 5-azacytidine and romidepsin are highly effective in patients with PTCL: a multicenter phase 2 study. *Blood* (2021) 137:2161–70. doi: 10.1182/blood.2020009004

133. Horwitz SM, Moskowitz AJ, Jacobsen ED, Mehta-Shah N, Khodadoust MS, Fisher DC, et al. The combination of duvelisib, a PI3K-δ,γ inhibitor, and romidepsin is highly active in Relapsed/Refractory peripheral T-cell lymphoma with low rates of transaminitis: Results of parallel multicenter, phase 1 combination studies with expansion cohorts. *Blood* (2018) 132:683–3. doi: 10.1182/blood-2018-99-115241

134. Iyer SP, Huen A, Ai WZ, Jagadeesh D, Lechowicz MJ, Okada C, et al. Safety and efficacy of tenalisib given in combination with romidepsin in patients with Relapsed/Refractory T-cell lymphoma: Final results from a phase I/II open label multi-center study. *Blood* (2021) 138:1365. doi: 10.1182/blood-2021-145851

135. Mehta-Shah N, Lunning MA, Boruchov AM, Ruan J, Nair S, Lynch P, et al. A phase I/II trial of the combination of romidepsin and lenalidomide in patients with relapsed/refractory lymphoma and myeloma: Activity in T-cell lymphoma. *J Clin Oncol* (2015) 33:8521–1. doi: 10.1200/jco.2015.33.15_suppl.8521

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 14 April 2023
DOI 10.3389/fonc.2023.1141108

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Shih-Sung Chuang,
Chi Mei Medical Center, Taiwan
Alexandra Hristov,
Michigan Medicine, University of Michigan, United States

*CORRESPONDENCE
Denis Miyashiro
denisrmiyashiro@gmail.com

SPECIALTY SECTION
This article was submitted to Hematologic Malignancies, a section of the journal Frontiers in Oncology

RECEIVED 10 January 2023
ACCEPTED 27 March 2023
PUBLISHED 14 April 2023

CITATION
Miyashiro D and Sanches JA (2023) Mycosis fungoides and Sézary syndrome: clinical presentation, diagnosis, staging, and therapeutic management.
Front. Oncol. 13:1141108.
doi: 10.3389/fonc.2023.1141108

Mycosis fungoides and Sézary syndrome: clinical presentation, diagnosis, staging, and therapeutic management

Denis Miyashiro* and José Antonio Sanches

Division of Clinical Dermatology, Hospital das Clínicas, University of São Paulo Medical School, São Paulo, Brazil

Mycosis fungoides (MF) and Sézary syndrome (SS) are cutaneous T-cell lymphomas. MF is the most common cutaneous lymphoma, and it is classified into classic Alibert-Bazin MF, folliculotropic MF, pagetoid reticulosis, and granulomatous slack skin, each with characteristic clinical presentation, histopathological findings, and distinct clinical behaviors. SS is an aggressive leukemic variant of cutaneous lymphoma, and it is characterized by erythroderma, lymphadenopathy, and peripheral blood involvement by malignant cells. There is a wide range of dermatological manifestations of MF/SS, and prompt recognition is essential for early diagnosis. Skin biopsy for histopathology and immunohistochemical analysis is imperative to confirm the diagnosis of MF/SS. Histopathology may also provide information that may influence prognosis and treatment. Staging follows the TNMB system. Besides advanced stage, other factors associated with poorer prognosis are advanced age, male gender, folliculotropism in histopathology of patients with infiltrated plaques and tumors in the head and neck region, large cell transformation, and elevated lactate dehydrogenase. Treatment is divided into skin-directed therapies (topical treatments, phototherapy, radiotherapy), and systemic therapies (biological response modifiers, targeted therapies, chemotherapy). Allogeneic bone marrow transplantation and extracorporeal photopheresis are other treatment modalities used in selected cases. This review discusses the main clinical characteristics, the histopathological/immunohistochemical findings, the staging system, and the therapeutic management of MF/SS.

KEYWORDS

mycosis fundgoides, Sézary syndrome, cutaneous T cell lymphoma, histopathology, prognosis, treatment

1 Introduction

Extranodal lymphomas account for approximately 30% of non-Hodgkin lymphomas (1). The skin is the second most involved organ, after the gastrointestinal tract (2). Primary cutaneous lymphomas are classified into cutaneous NK/T-cell lymphomas (CTCL) and cutaneous B-cell lymphomas (CBCL), by the World Health Organization (WHO) and the

European Organization for Research and Treatment of Cancer (EORTC). The CTCLs represent approximately 70% to 82% of all primary cutaneous lymphomas. Mycosis fungoides (MF) is the most common CTCL, representing almost 50% of all primary cutaneous lymphomas, and Sézary syndrome (SS) is a rare and aggressive leukemic disease (2–7).

MF was described in 1806 by Jean-Louis-Marc Alibert and, later, Pierre-Antoine-Ernest Bazin described the progression from patches (non-infiltrated lesions with erythema, scaling, and atrophy), to infiltrated plaques and tumors. Histologically, it is characterized by the proliferation of small and medium-sized epidermotropic CD4+ T-lymphocytes with convoluted nuclei (4).

SS was described by Albert Sézary and Yves Bouvrain in 1938 (8, 9). It is a leukemic variant of CTCL and occurs almost exclusively in adults. It is classically characterized by erythroderma, lymphadenopathy, and neoplastic cells in peripheral blood (3, 10).

In this review, we aim to describe the clinical presentation, diagnosis, staging system, and treatment currently available for MF and SS.

2 Epidemiology

It is estimated that primary cutaneous lymphomas have an annual incidence between 0.3 and 10.7 new cases per 1,000,000 individuals (1, 2, 5, 6). Approximately 70 to 82% of primary cutaneous lymphomas are CTCL, with annual incidences ranging between 3.4 and 7.7 per 1,000,000 individuals (2, 5, 6) and absolute predominance of MF and its variants and subtypes, which accounts for approximately 50% of the cases of cutaneous lymphomas (3, 4). Annual incidences of MF and SS range from 2.0 to 4.1 cases per 1,000,000 and 0.1 to 0.3 cases per 1,000,000, respectively (2, 5, 6). The incidence of cutaneous lymphomas is increasing, currently with an incidence three times higher than in the early 1980s (2).

MF typically affects adults with 50 to 60 years at diagnosis, and the disease is rarely described in childhood and young adults, except in the hypopigmented subtype of MF, with median age at diagnosis of 32 years (2, 11–15). Sézary syndrome (SS) is a rare and aggressive variant of CTCL that typically affect the elderly with a median age at diagnosis between 60 and 65 years (3, 4, 10, 13).

A systematic review and meta-analysis study (16) that analyzed North American (11, 17–22), European (23–26), Asian (12), and Australian (27) studies, included 6,279 patients with MF and SS and showed a predominance of males, accounting for 53% to 73% of the patients.

The analysis of the distribution of cutaneous lymphomas among different ethnicities is difficult due to the heterogeneity of this characteristic in different countries. An international and prospective multicenter study (Prospective Cutaneous Lymphoma International Prognostic Index - PROCLIPI study), which encompasses 19 countries from 6 continents, reported an absolute predominance of white patients (82.8%), reflecting a large number of North American and European centers (28). Data on the impact of ethnicity on disease behavior are conflicting due to the difficulty in the analysis of this parameter, however, there is evidence that black MF/SS patients are younger, have a female predominance, and are at higher risk of disease progression (28–30).

3 Pathogenesis

The pathogenesis of cutaneous lymphomas is not completely understood. It is believed that the chronic activation of T-cells by antigen-presenting cells leads to the gradual accumulation of mutations that culminate in the development of neoplastic cells. However, the triggering antigen for this chronic stimulation is unknown, and it could vary between patients (31). There is also the hypothesis that large plaque parapsoriasis and pityriasis lichenoides chronica would represent lymphocytic dyscrasias that could correspond to the link between polyclonal processes and MF (32, 33).

Bacterial, viral, fungal, and mycobacterial infections (34, 35), medications, and low vitamin D levels (36–39) have already been studied as the triggering factors in the origin of cutaneous lymphoproliferative processes. Analysis of regional grouping of patients with cutaneous lymphomas suggests exposure to unknown environmental factors and even occupational exposure (benzene and trichloroethylene) (40–43). However, none of these factors was consistently associated with the genesis of cutaneous lymphomas, and most of these studies are restricted to small series and case reports.

Studies analyzing the data of exome sequences of MF patients detected ultraviolet (UV) signatures in 10.8 to 57.6% of the mutational burden, suggesting that UV has a significant impact on the pathogenesis of MF (44). On the other hand, phototherapy with UVA or UVB is frequently used as a skin-directed therapy for MF/SS. The assessment of the interaction between UV and neoplastic cells and tumor microenvironment will add to the understanding of the real role of UV in MF/SS genesis and treatment.

MF and SS were considered the same disease for many years. However, the neoplastic cells of MF and SS have distinct immunophenotypic profiles. In MF, cells strongly express CCR4 and CLA, skin-homing receptors, and lack CCR7 and L-selectin, lymph nodes-homing receptors. This immunophenotype is typically observed in skin-resident memory T-cells. On the other hand, SS cells express CCR7 and L-selectin molecules, as well as CD27, a central memory T-cell marker, and they also strongly express CCR4 and other skin-homing receptors (CCR6, CCR10, CLA) (45). These findings suggest that MF and SS are distinct diseases, originating in different subtypes of T lymphocytes. However, the description of patches and plaques in MF patients that progress to a typical clinical picture of SS favors the hypothesis that there is a spectrum of manifestations encompassing the two entities. In addition to these observations, genetic and epigenetic studies demonstrate a great diversity in mutations and activated/inactivated signaling pathways in MF and SS (46, 47), and the same patient may have individually heterogeneous neoplastic cells (48). The genomic heterogeneity hinders the definition of whether MF and SS are part of the same disease, or if they are different diseases.

4 Clinical presentations

4.1 Mycosis fungoides

The WHO and EORTC classification of cutaneous lymphomas recognize the classic Alibert-Bazin type of MF and three variants: folliculotropic MF, pagetoid reticulosis, and granulomatous slack skin (GSS) (4). Besides the classic MF and the three variants, other clinicopathological subtypes of MF have been described: hypopigmented, poikilodermatous, erythrodermic, granulomatous, hyperpigmented, ichthyosiform, syringotropic, papular, purpuric, interstitial, pustular, bullous, verrucous, and psoriasiform MF. These other subtypes have clinical and pathological peculiarities but are included in the classic Alibert-Bazin MF group due to their similar prognostic features (14, 49–51).

Classic MF is the most frequent type, corresponding to 88.6%, followed by folliculotropic MF which corresponds to 11.4% of MF cases. Pagetoid reticulosis and granulomatous slack skin are extremely rare and represent less than 1% (4). Among the other MF subtypes included in the classic MF group, poikilodermatous MF is the most frequent (10% to 11%) followed by hypopigmented MF (3% to 10%) (13, 24).

Pruritus is the most frequent symptom, present in 80% of MF patients, and all patients with SS (13). Pruritus may be intense, especially in SS (10), and it is associated with significant worsening in quality of life (52).

Systemic symptoms are rarely seen in MF/SS. In a literature review study, Morris et al. evaluated 63 articles, including 505 patients with unusual findings in SS. Of these patients, just 8 patients (1.6%) had B symptoms, including fever, night sweats, and weight loss (53).

The time between symptom onset and diagnosis of MF varies between 2 years and 4.2 years (11, 54, 55). The PROCLIPI study observed that there is a diagnostic delay in early-stage MF, with median time between the first symptoms and diagnosis of 36 months (54). Cutaneous lymphomas are rare and are often misdiagnosed as eczematous processes, especially in the early stages of the disease. In addition, there is no single gold standard test that makes the diagnosis of MF or SS, but a set of clinical, histopathological, and molecular findings are needed, which contributes to the delay in the diagnosis.

4.1.1 Classic Alibert-Bazin MF

The classic type of MF, called the Alibert-Bazin type, is a progressive disease, with an indolent course. It is characterized by the development of patches, plaques, and tumors (Figures 1A–C) (3). Initially, there are non-infiltrated patches with erythema, scaling, and atrophy. These lesions may progress to erythematous infiltrated plaques, with well-defined borders and often with bizarre contours, with a foveolar, semi-annular, and serpiginous appearance, and eventually, tumors appear over the preexisting plaques or areas of healthy skin. It is common to observe a combination of lesions with patches, plaques, and tumors concomitantly. Not all MF patients progress from patches to plaques and tumors, but patches are always present (if a patient presents tumors without patches, other cutaneous lymphomas must

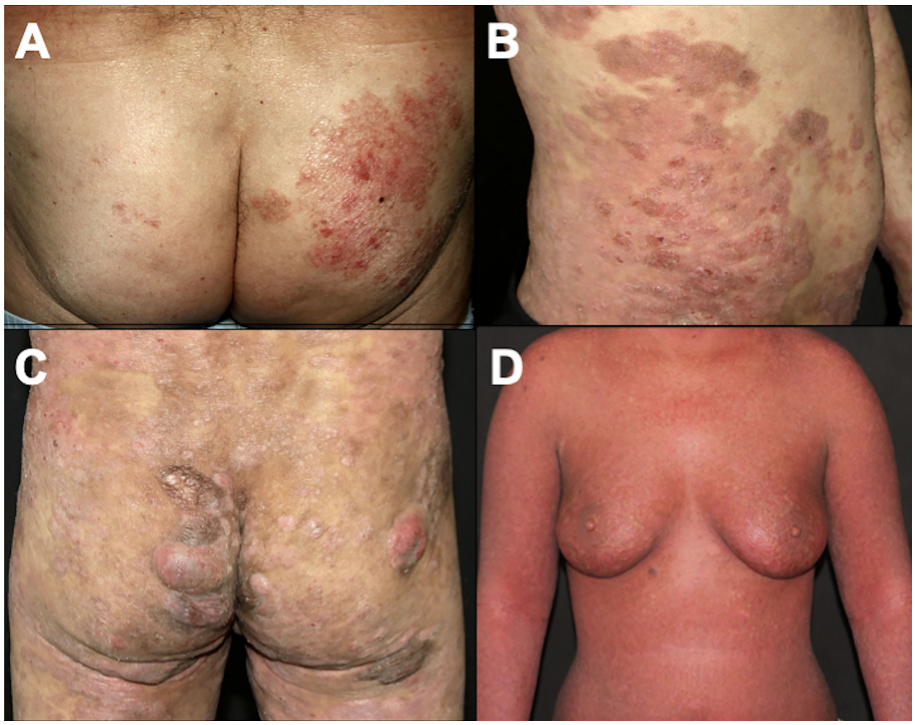


FIGURE 1
Clinical presentation of classic MF. Patches on the buttocks **(A)**; plaques on the abdomen **(B)**; patches, plaques, and tumors on the buttocks **(C)**; erythroderma **(D)**.

be considered in the differential diagnoses). Thick infiltrated plaques and tumors may ulcerate, and erythroderma (involvement of more than 80% of the body surface area) may develop as the disease progresses (Figure 1D) (3, 56). A study with 1,502 patients with MF/SS showed that 71.4% had patches, 36.3% had plaques and 13.5% had tumors. Erythroderma was observed in 16.6% of cases (24).

The lesions initially appear on sun-protected areas, especially on the buttocks and breasts. The lower trunk, inguinal regions, axillae, and proximal areas of the upper and lower limbs are frequently involved. Lesions appear in variable numbers, and they spread gradually (3, 14).

Extracutaneous dissemination to lymph nodes, blood, or viscera is rare but has a significant negative impact on disease prognosis (57).

4.1.2 Folliculotropic MF

Folliculotropic MF is the most common variant, and it was considered a variant with a poorer prognosis, due to the presence of neoplastic infiltrate deeper in the dermis. However, recent studies divide the cases into advanced folliculotropic MF, with infiltrated plaques and tumors in the head and neck region with intense pruritus, cicatricial alopecia, and poorer prognosis (Figure 2A); and early folliculotropic MF, with patches and thin plaques with follicular accentuation, comedones, and milia in the trunk, milder pruritus, and a favorable prognosis (Figure 2B) (58, 59).

Not all patients with clinical features of folliculotropic MF have the description of folliculotropism on histopathology. This discrepancy occurs because sometimes the follicles are not represented in skin biopsy samples. On the other hand, there is the description of follicular involvement even in lesions that clinically do not suggest follicular involvement. The diagnosis of folliculotropic MF is based on the clinical picture in association with histopathology (60). Even in cases of childhood MF, including hypopigmented MF, folliculotropism can be observed, with no impact on the course of the disease (61). It is known that the hair follicle is a region of immune privilege, and the disruption of this barrier is observed in folliculotropic MF (62). It is still uncertain whether this breach of the immune privilege barrier also occurs in other MF variants in which folliculotropism can be observed on histopathology, and whether there is any impact on the prognosis. Furthermore, follicular accentuation may not always demonstrate infiltration of the follicular epithelium by neoplastic cells, but only follicular mucinosis, which can make confirmation of folliculotropic MF diagnosis difficult (63).

4.1.3 Pagetoid reticulosis

Pagetoid reticulosis is a rare and indolent MF variant. It is characterized by psoriasiform or hyperkeratotic lesions affecting the extremities (Figure 3). On histopathology, there is epidermal hyperplasia with a pagetoid proliferation of atypical T-lymphocytes with CD4+, CD8+, or CD4-CD8- phenotype (3, 64).

4.1.4 Granulomatous slack skin

Granulomatous slack skin is a rare and indolent MF variant, with peculiar clinical and histopathological characteristics. Initially, infiltrated papules and plaques appear on the skin folds, which may evolve with skin laxity (Figure 4). Histopathology shows granulomas, elastophagocytosis, and atypical lymphocytes infiltrating the skin. Patients with granulomatous slack skin have an increased risk of a second hematologic malignancy, especially anaplastic large T-cell lymphoma and Hodgkin's lymphoma (65).

4.1.5 Other clinicopathological subtypes

4.1.5.1 Hypopigmented MF

The hypopigmented subtype of MF affects most frequently young patients with a median age at diagnosis of 32.2 years and with skin types IV-V (14, 15). An American study with 1,502 patients reported a diagnosis of hypopigmented MF in 3.4% of patients with MF (24). On the other hand, a Brazilian study

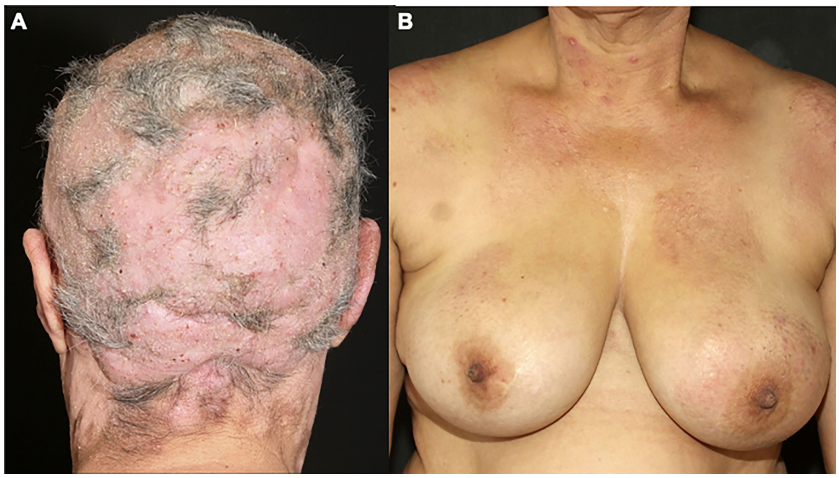


FIGURE 2
Folliculotropic MF. Advanced folliculotropic MF with infiltrated plaques on the scalp causing alopecia and milia **(A)**; early folliculotropic MF with patches and plaques with comedones and milia on the breasts, thorax, and neck **(B)**.

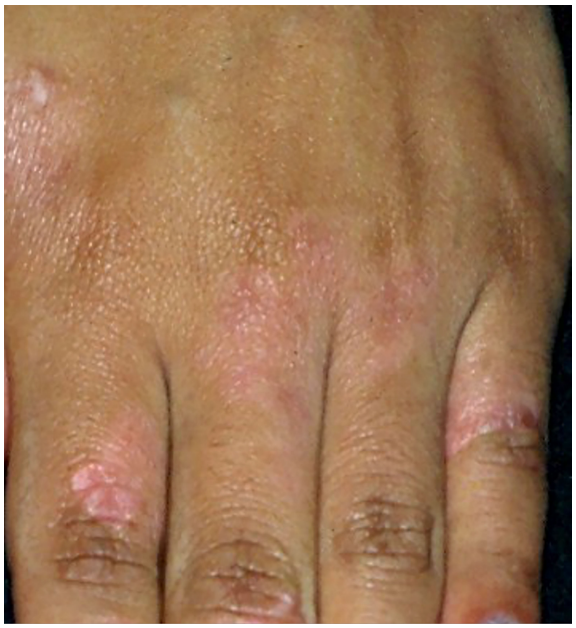


FIGURE 3
Pagetoid reticulosis. Psoriasiform plaques on the dorsum of the hand.

reported a frequency of 19.6%, and studies with the pediatric population report hypopigmented MF in up to 50% of the total number of patients analyzed. Thus, geographical variations in the frequency of hypopigmented MF occur according to the ethnic characteristics of the populations (13, 15, 61, 66, 67). Hypopigmented MF is characterized by hypopigmented patches on the trunk, thighs, buttocks, and extremities (Figure 5) (15).

4.1.5.2 Poikilodermatous MF

Poikilodermatous MF is characterized by hypopigmented, hyperpigmented, atrophic, and telangiectatic lesions, typically affecting flexural areas and the trunk. This type of MF accounts for 10% of cases and is more frequent in young patients (median age at diagnosis of 40 to 50 years) (51, 68). It can be subdivided into poikilodermatous MF with localized lesions and with generalized lesions (Figure 6). Patients with generalized poikilodermatous MF present erythroderma (more than 80% of the body surface area affected) and, despite the extensive skin area affected, the prognosis is excellent (51).

4.1.5.3 Erythrodermic MF

The diagnosis and clinical management of erythrodermic MF are difficult. The patient presents with erythema and scaling on more than 80% of the body surface area (Figure 7), and the differential diagnoses include all causes of erythroderma, such as psoriasis, eczema, pityriasis rubra pilaris, drug eruption, and Sézary syndrome (69, 70). It is considered an aggressive MF variant, and peripheral blood assessment is critical to differentiate erythrodermic MF from SS (14, 71). It is discussed whether idiopathic cases of erythroderma may correspond to pre-malignant phases or even a type of MF in which it was not possible to identify neoplastic cells infiltrating the skin, due to the fewer epidermotropism of atypical lymphocytes observed in these erythrodermic cases (72).

4.1.5.4 Other

Other rare clinicopathological subtypes of MF include granulomatous, hyperpigmented, ichthyosiform, syringotropic, papular, purpuric, interstitial, pustular, bullous, verrucous, and psoriasiform MF. These other subtypes are classified within the group of classic MF, and patients frequently present a mixed clinical presentation, together with the presence of patches, plaques, and tumors described by Alibert and Bazin (14, 49–51).

4.2 Sézary syndrome

SS is a leukemic variant of CTCL and occurs almost exclusively in adults. It presents with erythroderma with a diffusely infiltrated

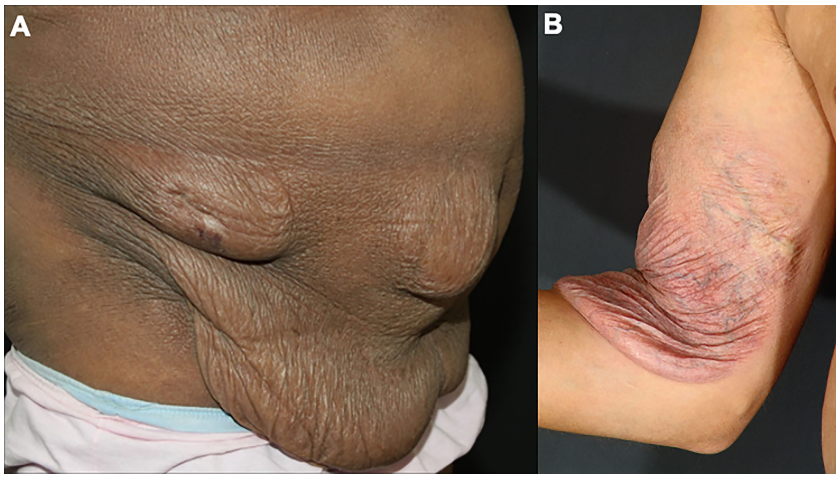


FIGURE 4
Granulomatous slack skin. Skin laxity on the abdomen **(A)** and the arm **(B)**.

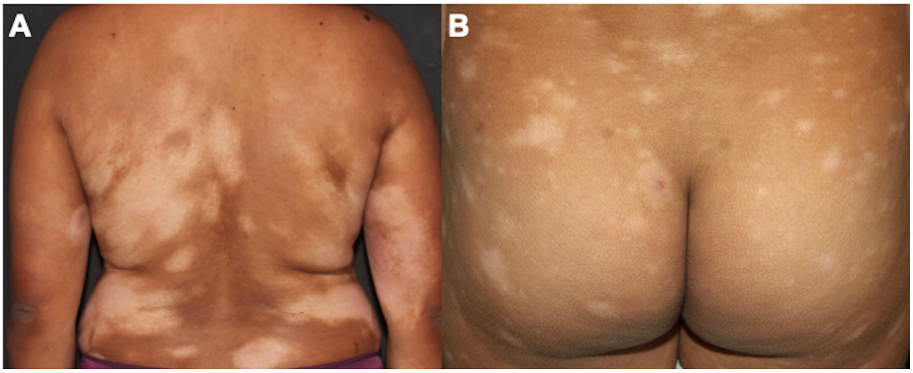


FIGURE 5
Hypopigmented MF. Multiple hypochromic patches on the trunk and arms **(A)**, and on the buttocks **(B)**.

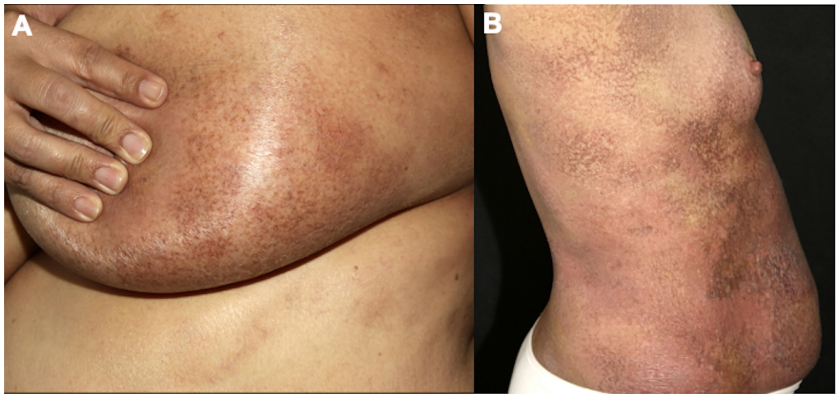


FIGURE 6
Poikilodermatous MF with localized **(A)** and generalized lesions **(B)**.

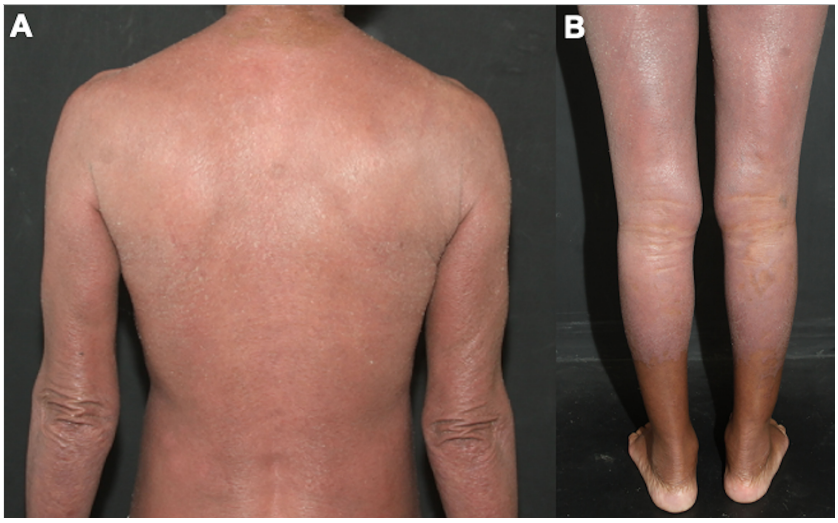


FIGURE 7
Erythrodermic MF. Difuse erythema and scaling affecting on the back and arms **(A)**, and on the lower limbs **(B)**.

aspect of the skin, often associated with non-scarring alopecia, palmoplantar keratoderma, nail dystrophies, generalized lymph nodes enlargement, and intense itching (Figures 8, 9). Circulating neoplastic T-cells are detected by searching for Sézary cells (large lymphocytes with a cerebriform nucleus) in the blood smear and by immunophenotyping of lymphocytes by flow cytometry. Search for T-cell receptor (TCR) gene rearrangement detects monoclonal populations in peripheral blood, skin, and lymph nodes. For the diagnosis of SS, the International Society for Cutaneous Lymphomas (ISCL) propose that the presence of a monoclonal population in the skin and blood (same clone) and one of the phenotypic alterations (CD4/CD8 ≥ 10 and/or CD4+CD7- ≥ 40% and/or CD4+CD26- ≥ 30%) or detection of more than 1,000 Sézary cells/μL are mandatory (Table 1) (3, 10, 73). According to the most recent World Health Organization (WHO) classification, SS diagnosis also requires erythroderma, generalized lymphadenopathy, and clonally related malignant T-cells in the skin, peripheral blood, and lymph nodes (74).

5 Skin biopsy

The confirmation of MF/SS diagnosis is difficult. There are no "gold standard" tests, and diagnosis is based on clinical, histopathological, and molecular findings. On histopathology, the diagnosis of the neoplasm is usually suggested by experienced pathologists by the cytomorphological characteristics and the disposition of the architectural arrangement of the infiltrate. Currently, for the classification of lymphomas, it is essential to carry out an immunohistochemical study, whose panel of antibodies is rationalized according to the histological findings (75). It is recommended to biopsy different lesions simultaneously to increase the accuracy of histopathology (14). In MF, the cytological aspects and the architectural pattern of the cellular infiltrate are correlated with the clinical stage of the disease. Epidermotropism of atypical lymphocytes is the most striking feature. Atypical lymphocytes have a large and hyperchromatic nucleus, sometimes with cerebriform contours (76). In the early patch stages and in the erythrodermic phase, the number of atypical lymphocytes infiltrating the epidermis is scarce, and it increases in plaques and tumors (77). However, sometimes epidermotropism may be absent even in plaques and tumors, making the diagnosis if MF more difficult. Other typical pathological findings are Pautrier's microabscesses (grouping of at least 4 lymphocytes around a Langerhans cell in the epidermis) (Figure 10) (78, 79). The alignment of lymphocytes in the basal layer may also be observed in MF (80). The absence of spongiosis favors the diagnosis of MF (78); however, it has low specificity (60%) (81).

Syringotropic, granulomatous, and interstitial MF are histological MF variants rarely described in the literature. Syringotropic MF is more commonly seen in palmoplantar lesions (82). Granulomatous MF should be distinguished from granulomatous slack skin based on the clinical picture (in granulomatous MF there are no areas of lax skin) and histopathology (elastolysis and elastophagocytosis are seen in granulomatous slack skin) (65).

Large cell transformation (LCT) is a histopathological finding characterized by the presence of large cells (cells four times larger than normal lymphocytes) in 25% or more of the dermal infiltrate, or when these large cells are grouped in nodules (the definition of nodules of large cells is vague, most pathologists do not use it, and these nodules can be focal and hard to identify in skin or lymph node biopsies) (83). It can occur in tumor lesions and, less frequently, in plaque and erythrodermic MF lesions. These cells may or may not express CD30, and it is essential to differentiate MF with LCT from CD30-positive anaplastic large T-cell lymphoma (73). A higher frequency of CD56 expression is reported in MF with LCT compared to non-transformed MF, however, the clinical significance of this finding is not clear (84). The detection of LCT confers a poorer prognosis (57).

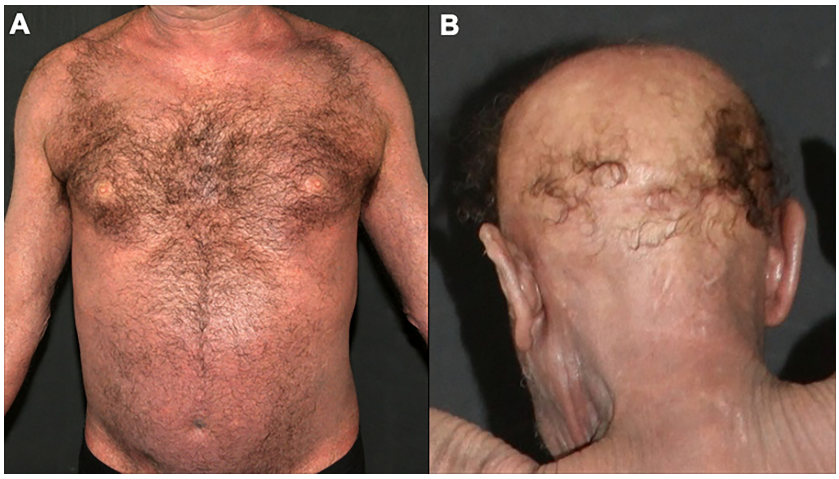


FIGURE 8
Sézary syndrome. Diffuse erythema and scaling **(A)** and non-scarring alopecia **(B)**.

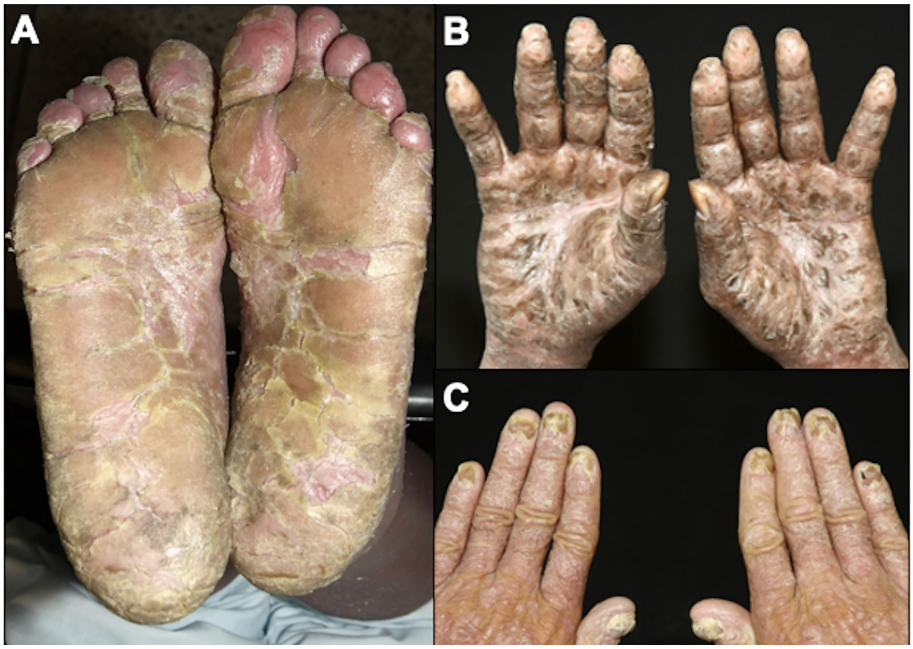


FIGURE 9
Sézary syndrome. Plantar **(A)** and palmar **(B)** keratoderma, onychodystrophy **(C)**.

TABLE 1 Sézary syndrome diagnostic criteria.

Sézary cells in peripheral blood smear	≥ 1,000 Sézary cells/μL
Immunophenotyping of lymphocytes in peripheral blood	CD4/CD8 ≥ 10
	CD4+CD7- ≥ 40%
	CD4+CD26- ≥ 30%
T-cell receptor gene rearrangement	Monoclonal population on the skin and peripheral blood (same clone)

*For the diagnosis of SS, detection of a monoclonal population of T-cells on the skin and the blood is mandatory together with at least one morphologic or immunophenotypic alteration.

5.1 Immunohistochemistry

Malignant cells are CD4-positive T-cells with a memory phenotype (CD3+CD4+CD45RO+CD8–) and negativity for CD7 antigen expression in 70% of cases (Figure 11). Rarely, neoplastic cells express a CD3+CD4-CD8+ phenotype, especially in hypopigmented MF, with the same clinical behavior and prognosis as cases with CD4+CD8- cells. Losses of CD2 and CD5 are observed less frequently (56, 85, 86).

CD30 is a molecule of the tumor necrosis factor family and is expressed on the cell surface of activated B and T-lymphocytes. Search for CD30 expression in MF is important, especially in advanced-stage cases. The brentuximab-vedotin, a drug-antibody conjugate in which the monomethyl auristatin E (MMAE) molecule, an antimicrotubule agent, is linked to an anti-CD30 monoclonal antibody, was approved for the treatment of CD30+ MF refractory to prior systemic treatment. This drug has shown good responses in cases of MF/SS that express CD30 in at least 10% of neoplastic cells in a phase 3 clinical trial (87). Studies show CD30 expression in most patients (88), but its expression is low, with an average expression in 4% of neoplastic cells (89).

Despite its relationship with cell proliferation, the Ki-67 expression was not consistently associated with poorer prognosis (57), but a higher expression is observed in cases of large cell transformation (89).

There are reports of phenotypic variability of neoplastic cells in the same patient. These variations have uncertain clinical significance, but it is important to emphasize the need to perform biopsies in different topographies and repeatedly in cases where the clinical suspicion of MF/SS is not supported by the histopathological examination (90).

In SS, histopathology is similar to MF. However, in erythrodermic forms of cutaneous lymphomas (erythrodermic MF and SS) epidermotropism is less evident, and in up to a third of SS cases the histopathology may be nonspecific (10).

5.2 T-cell clonality in the skin

The search for TCR gene rearrangement, demonstrating monoclonal T lymphocyte proliferation in the skin, lymph nodes, and/or peripheral blood, may contribute to the diagnosis of T-cell lymphomas. It is done by polymerase chain reaction (PCR), Southern blot, or more recently by next-generation sequencing (NGS) (91–93).

T-cell monoclonality is observed in 50% of patch lesions, 73% of plaque lesions, 83 to 100% of tumors, and 77.5% of patients with SS, and the detection of the same clone in different skin lesions favors the diagnosis of CTCL. On the other hand, 25 to 65% of inflammatory dermatoses may exhibit oligoclones in the skin. Therefore, the presence of a monoclonal population should be

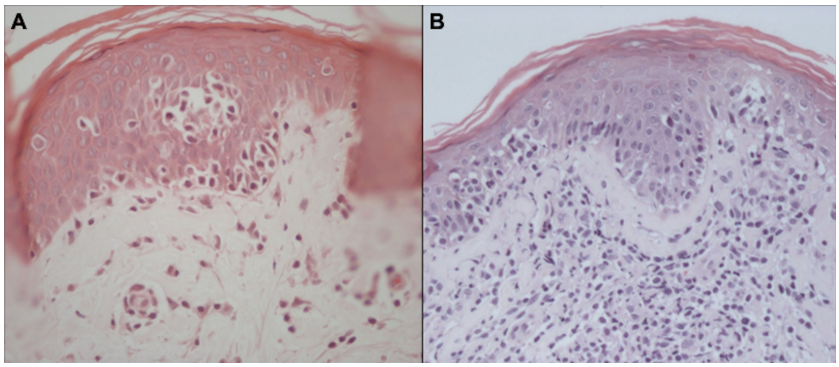


FIGURE 10
Histopathology of MF. Epidermotropism of atypical lymphocytes with Pautrier microabscess and scant dermal lymphocytic infiltrate **(A)**. Epidermotropism, Pautrier microabscesses, and a more prominent dermal lymphocytic infiltrate **(B)**.

analyzed with caution, especially in cases where histopathology does not confirm the diagnosis of MF/SS (10, 78, 94).

6 Staging

Early-stage diseases (stages IA to IIA) account for 78% to 93% of MF/SS cases; advanced-stage (≥IIB) corresponds to 7% to 22% (21, 95, 96).

6.1 Skin (T)

For MF and SS, the modified TNM system (tumor, lymph nodes, and metastasis) of Bunn and Lamberg (1979) was adopted, considering T as cutaneous lesions, N as lymph nodes, M as visceral lesions, and adding B as peripheral blood (Tables 2, 3) (73, 97, 98).

The prognosis of patients with MF is directly associated with the clinical stage. Progressively poorer survival curves are observed depending on the extent of cutaneous involvement, presence of tumors and erythroderma, lymph node, visceral, and peripheral blood involvement (73). A study with 489 MF patients showed that T1 patients had survival similar to the general population. However, T2, T3, and T4 patients had significantly lower survival rates. T2 patients with plaque stage (T2b) had poorer survival than T2 patients with patches only (T2a) (99). The presence of a tumor greater than or equal to 1 cm in diameter is sufficient to define the T3 stage. In the presence of more than one T category, the highest one must be included, and in cases of erythroderma (T4) associated with tumor lesions (T3), both forms must be recorded [T4 (3)] (73).

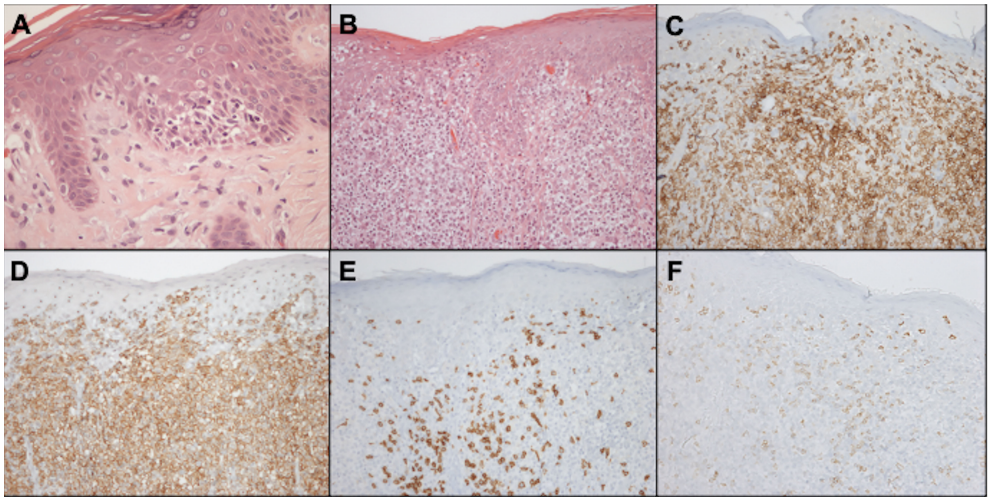


FIGURE 11
Histopathology and immunohistochemistry of the skin. Epidermotropism of atypical lymphocytes, hematoxylin-eosin staining at 400x magnification **(A)**. Exuberant atypical infiltrate affecting the epidermis and dermis, hematoxylin-eosin at 200x magnification **(B)**. CD3 positivity, demonstrating that epidermal and dermal cells correspond to T-lymphocytes, 200x magnification **(C)**. CD4 positivity, 200x magnification **(D)**. Expression of CD8 on small reactive cells, 200x magnification **(E)**. Loss of CD7 expression in CD4-positive neoplastic T cells, 200x magnification **(F)**.

TABLE 2 Revised TMNB classification for MF and SS.

Skin (T)	
T1	Limited patches/plaques (covering <10% of the total skin surface)
T1a	Patches only
T1b	Plaques/papules ± patches
T2	Generalized patches/plaques (covering ≥10% of the total skin surface)
T2a	Patches only
T2b	Plaques/papules ± patches
T3	Tumor(s) ≥ 1cm diameter
T4	Confluence of erythema covering ≥80% of the body surface area
Lymph node (N)	
N0	No clinically abnormal peripheral lymph nodes
N1	Clinically abnormal peripheral lymph nodes; dermatopathic lymphadenopathy or histopathological involvement by isolated atypical lymphocytes, without alteration of the lymph node architecture
N1a	Clone negative or equivocal
N1b	Clone positive and identical to skin
N2	Clinically abnormal peripheral lymph nodes; histopathological involvement by aggregates of atypical lymphocytes, without alteration of the lymph node architecture
N2a	Clone negative or equivocal
N2b	Clone positive and identical to skin
N3	Clinically abnormal peripheral lymph nodes; frank histopathological involvement and partial/complete effacement of the lymph node architecture
N3a	Clone negative or equivocal
N3b	Clone positive and identical to skin
Nx	Clinically abnormal peripheral lymph nodes; without histopathological confirmation
Viscera (M)	
M0	Without visceral involvement
M1	With visceral involvement
M1a	Bone marrow involvement
M1b	Non-bone marrow visceral involvement
Blood (B)	
B0	Absence of significant blood involvement (< 250/μL of CD4+CD26- or CD4+CD7- cells)
B0a	Clone negative or equivocal
B0b	Clone positive and identical to skin
B1	Low tumor burden in the blood, but do not fulfill B2 criteria
B1a	Clone negative or equivocal
B1b	Clone positive and identical to skin
B2	High tumor load in the blood (CD4/CD8 ≥ 10; CD4+CD7- ≥ 40%; CD4+CD26- ≥ 30%; ≥ 1000/μL of CD4+CD26- or CD4+CD7- cells or other aberrant population of lymphocytes identified by flow cytometry)
B2a	Clone negative or equivocal
B2b	Clone positive and identical to skin
Bx	Unable to quantify blood involvement

TABLE 3 Clinical staging system for MF and SS.

Clinical staging	TNMB classification			
IA	T1	N0	M0	B0 or B1
IB	T2	N0	M0	B0 or B1
IIA	T1 or T2	N1 or N2	M0	B0 or B1
IIB	T3	N0 to N2	M0	B0 or B1
IIIA	T4	N0 to N2	M0	B0
IIIB	T4	N0 to N2	M0	B1
IVA1	T1 to T4	N0 to N2	M0	B2
IVA2	T1 to T4	N3	M0	B0 to B2
IVB	T1 to T4	N0 to N3	M1	B0 to B2

6.2 Lymph nodes (N)

Around 80% of patients have no clinical suspicion of lymph node involvement at the diagnosis (N0) (24, 95).

A biopsy of lymph nodes greater than or equal to 1.5 cm in diameter or of any palpable lymph node, regardless of size, that is hardened, irregular, fixed, or forming a conglomerate of lymph nodes is recommended. Lymph node enlargement can be confirmed by ultrasound, computed tomography (CT), positron emission tomography (PET), or magnetic resonance imaging, before the biopsy is performed. Excisional biopsy is preferred, except in cases with a high risk of infection, especially erythrodermic patients, when core needle biopsy may be indicated. In the presence of multiple altered lymph nodes, the order of preference of biopsy is cervical, axillary, and inguinal lymph nodes, as cervical lymph nodes are more likely to demonstrate lymphomatous involvement (73, 100, 101).

Erythrodermic patients, such as patients with SS, regardless of the erythroderma etiology, frequently have enlarged lymph nodes (69, 70). This occurs due to intense skin inflammation in erythrodermic patients. Even in SS cases, almost 20% have no lymph node involvement by the disease confirmed by histopathology, and dermatopathic lymphadenitis is detected (10).

6.2.1 Histopathological examination of the lymph node

Dermatopathic lymphadenopathy (N1) is characterized by paracortical layer hyperplasia, usually secondary to chronic cutaneous inflammation (102). The N2 stage corresponds to partial lymph node involvement; and N3, to complete involvement with an architectural alteration (Figure 12). In the literature, nodal involvement (N2 or N3) is observed in 30 to 50% of biopsied cases (24, 103).

6.2.2 Search for T clonality in the lymph node

Studies show lymph node T-cell monoclonality in 77.8% to 81.5% of patients with SS (10, 104). Even in patients with MF and biopsy with dermatopathic lymphadenopathy (N1), the presence of a monoclonal population can be detected (stage N1b), and studies suggest that the presence of this clone confers a poorer prognosis (104). However, more studies are needed to assess whether systemic treatments change the course of these patients with early MF and N1b stage, who would usually be treated with skin-directed therapies.

6.3 Viscera (M)

Regarding the M stage, splenomegaly detected on physical examination or imaging studies is considered a visceral disease regardless of histological confirmation. The suspicion of involvement of other organs, through clinical, laboratory, or imaging evaluation, must be confirmed by a biopsy (73).

Less than 1% of MF/SS cases have visceral involvement at the diagnosis, and its presence is associated with a poorer prognosis, with a median overall survival of 33.3 months (57, 95).

It is recommended that imaging tests be performed for the staging of all patients with MF/SS. For early-stage disease, chest radiography and abdominal and lymph nodes ultrasonography may be performed. In advanced cases, CT of the chest, abdomen, and pelvis, PET scan, or MRI are indicated depending on the organ to be evaluated (73). Studies show that the PET scan is more accurate in diagnosing nodal involvement in patients with MF/SS, but this test is not available in all medical centers, and its high cost makes it difficult to perform routinely (103).

6.3.1 Visceral biopsies

An autopsy study with 45 MF patients observed lymph node and visceral involvement in 24 cases (53.3%), particularly in patients with advanced disease (stage IIB-IVB) (105, 106). Another study with autopsies of MF/SS patients observed that the most affected extracutaneous sites are: lymph nodes (60%), spleen (50%), lungs (43%), liver (41%), bones (27%), kidneys (27%), tongue and mucous membranes (19%), heart (17%), pancreas (17%), thyroid (14%) (107). Currently, autopsy studies are scarce, but these older studies show that visceral involvement may be more

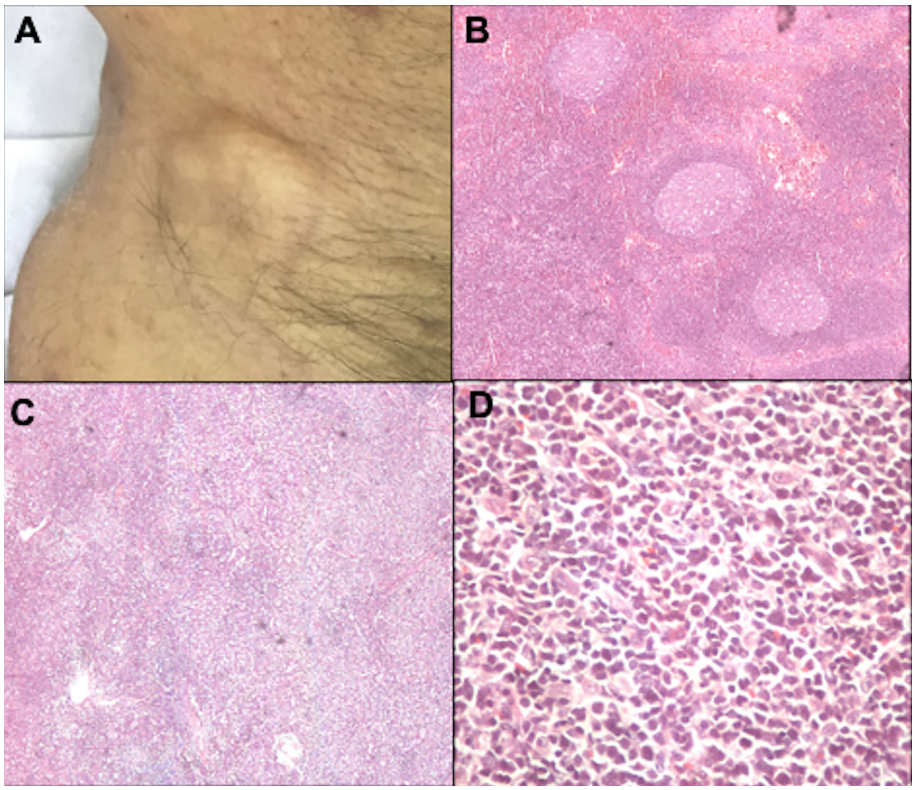


FIGURE 12
Lymph node evaluation. Inguinal lymph node enlargement **(A)**. Dermatopathic lymphadenopathy with the presence of germinal centers, hematoxylin-eosin at 20x magnification **(B)**. Lymph node involvement by lymphoma, with architectural alteration, hematoxylin-eosin at 20x magnification **(C)**. Pleomorphic lymphoid cells, hematoxylin-eosin at 200x magnification **(D)**.

frequent than what is detected. However, the impact of visceral infiltration not detected clinically or by imaging methods (only at autopsy) on outcome and prognosis is unknown.

6.3.2 Myelogram and bone marrow biopsy

Bone marrow biopsy is indicated in patients with significant peripheral blood involvement (stage B2) or when there are hematological abnormalities that cannot be explained by other causes. Bone marrow involvement in MF/SS is rare and is considered visceral involvement (M1) (24, 73). A myelogram is rarely indicated (24). The role of bone marrow biopsy in MF/SS is not well established, and studies on the impact of bone marrow involvement on disease prognosis are conflicting (108, 109). A study with SS patients showed that 31.6% of cases had bone marrow involvement, but there was no negative impact on survival in cases with bone marrow infiltration (10). A study on autopsies of patients with MF/SS showed bone marrow involvement in 25% of patients. On the other hand, a study with 50 patients with different stages of cutaneous T-cell lymphoma did not observe bone marrow involvement in any case (105). Benign lymphoid nodules can be seen in the bone marrow of healthy people, and the definition of tumor infiltration depends on the criteria used (10, 105). Thus, it is necessary to standardize the histopathological analysis to assess the presence of malignant T-cells in the bone marrow. Studies evaluating clonality in the bone marrow are scarce, but one study demonstrated T-lymphocyte clones in 53.6% (15/28) of patients with SS (10).

6.4 Blood (B)

Hematologic involvement is assessed by peripheral blood smear (search for Sézary cells), immunophenotyping of lymphocytes by flow cytometry, and search for T-cell clonality. Hematologic involvement is stratified into B0 (no blood involvement, ≤ 5% Sézary cells or < 250/μL of CD4+CD26- or CD4+CD7- cells), B1 (low blood tumor burden, with > 5% Sézary cells or ≥ 250/μL of CD4+CD26- or CD4+CD7- cells, but not meeting the criteria for B2), and B2 (high tumor burden in the blood, with ≥ 1,000 Sézary cells/μL, an increase in CD4+ cells with a CD4/CD8 ratio ≥ 10, CD4+/CD7- ≥ 40%, CD4+/CD26- ≥ 30%, ≥ 1000/μL of CD4+CD26- or CD4+CD7- cells, or other aberrant population of lymphocytes identified by flow cytometry). The B stages may be further subdivided into B0a, B1a, B2a (negative blood clonality) or B0b, B1b, B2b (positive blood clonality) (73, 98, 100, 110).

6.4.1 Search for Sézary cells in peripheral blood smear

The morphological evaluation of lymphocytes on a peripheral blood smear by experienced hematopathologists is an easy and

convenient tool to differentiate patients with erythrodermic cutaneous lymphomas and erythroderma caused by inflammatory skin conditions. However, with the advancement of flow cytometry techniques, the evaluation of lymphocyte morphology by peripheral blood smear is less frequently performed. This stems from technical difficulties, such as high time consumption and imprecision of manual search for Sézary cells, especially when these cells are small. In addition, phenotypic alterations occur earlier than morphological alterations (111).

6.4.2 Immunophenotyping of lymphocytes by flow cytometry

Although flow cytometry has become the standard tool for the evaluation of peripheral blood, there is no consensus and standardization regarding the markers used and the way to report this test. Ideally, it would be necessary to include CD3 and CD19 in the studied panel, to differentiate T and B-lymphocytes, respectively. Subsequently, T-lymphocytes should be better characterized, with the evaluation of CD4 and CD8, as well as CD7 and CD26, which are lost in neoplastic cells in MF/SS. Other T-lymphocyte markers, such as CD3, CD2, and CD5, are rarely lost, however, a diminished intensity or partial loss may be observed in some cases (112). CD52, CD25, CD30, and CCR4 are other markers that can aid in the therapeutic decision, as these molecules are targets of specific monoclonal antibodies (alemtuzumab, denileukin diftitox, brentuximab-vedotin, mogamulizumab, respectively) (111, 113).

Preliminary data from the PROCLIPI study show changes in the peripheral blood of patients with early MF with patches and plaques, fulfilling criteria for B1, in up to 5.9% of patients (114). The clinical significance of these changes in early MF is uncertain, and it is unknown whether they would be cases of true hematological involvement of MF. In erythrodermic patients with peripheral blood involvement compatible with stage B1, there is a greater risk of disease progression (115).

In SS, in addition to the increased CD4-positive T-cells, CD8 cytotoxic T-cells are reduced. Also, functional alterations of CD8 and NK cells are described, leading to a state of immunosuppression by compromising the innate immune response (116, 117).

6.4.3 T-cell clonality in blood

Preliminary data from the PROCLIPI study show that the frequency ofmonoclonal T-cell population in the blood matching the skin clone of patients with MF increases with higher stages (114). In patients with SS, clonality in the blood is positive in 86.7% to 100% of patients (10, 118). However, the presence of T-cell clones in the blood is not always a sign of malignancy, since clones may be observed in healthy elderly individuals and patients with benign dermatoses, and the significance of these circulating clones is unknown (119, 120).

6.5 Other laboratory tests

Lactate dehydrogenase (LDH) is an enzyme that catalyzes the conversion of lactate to pyruvate and vice versa in the process of glycolysis. Its activity is high in tumor cells of several malignancies (121). When increased at the diagnosis, a poorer prognosis is observed in MF/SS (57). An increase in LDH is observed in 25% of MF patients (10, 13, 122, 123).

Beta-2 microglobulin is part of the major histocompatibility complex class 1 and is present in all nucleated cells. Increased levels are associated with poorer prognosis in several hematologic malignancies, but the mechanism of association between beta-2 microglobulin and prognosis is unknown. Studies suggest that this molecule is involved in the survival and proliferation of malignant cells, as well as their ability to metastasize (124). In MF, studies demonstrate the association between its levels and disease progression, and higher levels of beta-2 microglobulin are detected in patients with late-stage MF compared to early MF, and in SS compared to MF.

Low albumin levels are associated with poorer prognosis in nodal lymphomas, and it is used as a predictor of response to first-line chemotherapy for peripheral T-cell lymphomas (125). In erythrodermic patients, the intense protein loss due to the universal skin alteration may cause hypoalbuminemia (126, 127). Hypoalbuminemia is more frequent in patients with advanced MF (20.9%) compared to early MF (5.4%), and in patients with SS (30.8%) compared to patients with MF (11.7%) (13). However, in multivariate analyses hypoalbuminemia was not associated with poorer prognosis in cutaneous lymphomas (13, 24, 57).

7 Prognosis

Lymphomas that present primarily in the skin, without evidence of extracutaneous disease at the diagnosis, often have a more indolent clinical behavior and a better prognosis compared with systemic lymphomas of a similar histological subtype (3).

In MF, patients with skin-limited disease have a good response to topical treatments and the overall survival is similar to the healthy population. Only 2% of the patients with localized lesions (involvement of less than 10% of the body surface area) die after 32 years of follow-up and only 9% show progression to plaques and tumors with lymph node, blood, and visceral dissemination (11). The 5-year overall survival is 91 to 97% for patients with non-infiltrating patch lesions or localized plaques (<10% of the skin surface), 81 to 85% for patients with generalized lesions (≥10% of the skin surface), 44% for patients with tumors, and 20 to 30% for those with lymph node disease. Sepsis, especially caused by *Staphylococcus aureus*, represents one of the most frequent causes of death in advanced cases (24). Transformed MF has a poorer prognosis, with a 5-year overall survival of 38.5% (57). SS is an aggressive lymphoma, with five-year survival rates ranging from 40 to 50% (3, 10, 57, 128, 129).

The CLIC study proposed a prognostic stratification index based on age (≥ 60 years), increase in LDH, large cell transformation, and stage IV at diagnosis (57). However, even this prognostic index proposed by this international multicentricstudy was not applicable in all series (130). Other factors reported to be associated with poorer prognosis are

advanced age (20, 24, 131), male gender (24), folliculotropism in histopathology in patients with infiltrated plaques and tumors in the head and neck region (24, 58, 132–134). However, the results of studies on prognostic factors in MF/SS are conflicting. Such prognostic studies are mostly retrospective observational studies. The PROCLIPI study is an international multicenter prospective study that was derived from the CLIC study, which was retrospective. It aims to develop a prognostic index to achieve better stratification and management of patients (135).

8 Treatment

Watch and wait may be indicated in cases of stage IA MF that are at low risk of progression and with survival similar to the general population for their gender and age (136).

For early-stage patients with no extracutaneous involvement, skin-directed therapies are used with good response rates. For advanced-stage disease with multiple tumors, erythroderma, or extracutaneous disease, or early-stage with multiple infiltrated plaques refractory to topical therapy, systemic treatments are indicated (137).

Skin-directed therapies include topical steroids, nitrogen mustard, bexarotene, phototherapy with narrow-band UVB and PUVA, and radiation therapy (localized and total skin electron beam irradiation) (113, 138, 139). Topical tacrolimus has been used in a few young patients with hypopigmented MF. It is effective in early-stage MF, but its effectiveness is based on a few case series (140). Radiotherapy is an effective treatment, especially for tumoral lesions, but relapses frequently occur after a few months, and maintenance therapy is mandatory (141).

Systemic treatments are used mainly in advanced-stage disease, and the most used therapeutic modalities are steroids and biological response modifiers, which include interferon-alpha, retinoids (bexarotene, acitretin, isotretinoin), histone deacetylase inhibitors (romidepsin, vorinostat), brentuximab vedotin (anti-CD30), mogamulizumab (anti-CCR4), alemtuzumab (anti-CD52). Few patients receive monochemotherapy or polychemotherapy. Chemotherapy drugs have high response rates, however, the duration of response is short, with rapid recurrence. There are no standardized chemotherapy protocols, but most studies have evaluated the efficacy of gemcitabine, chlorambucil, methotrexate, pralatrexate, liposomal doxorubicin, CHOP, and CHOP-like regimens (137, 140, 142).

Allogeneic bone marrow transplantation is an effective and potentially curative treatment in advanced and refractory cases (143). Extracorporeal photopheresis is primarily indicated for erythrodermic forms (erythrodermic MF and SS) (144). Surgical excision is rarely used and is reserved for the treatment of localized nodular or tumor lesions (140).

A study comparing the therapeutic modalities used around the world has shown important differences, especially when comparing North American practices with those of other countries (European, Latin American, Asian, and Australian). Such differences occur due to the availability of different treatments and institutional experiences. Despite this variability, there are no significant differences in survival (113, 137, 141).

9 Conclusion

Cutaneous lymphomas are rare extranodal non-Hodgkin lymphomas. Mycosis fungoides is the most frequent cutaneous T-cell lymphoma. It is an indolent disease, with different clinical manifestations. Sézary syndrome is a rare aggressive leukemic T-cell lymphoma that presents with erythroderma, lymphadenopathy, and peripheral blood involvement. Despite the relatively good prognosis, with high survival rates in most cases, MF patients suffer from skin lesions that significantly decrease the quality of life due to concerns that the disease may progress, due to the pruritus that may be intense, and due to the social stigma caused by skin diseases. Thus, clinical recognition and correct management are essential for early diagnosis and appropriate care of these patients.

Author contributions

DM: elaboration and final revision of the manuscript. JS: conception, elaboration, and final revision of the manuscript. All authors contributed to the article and approved the submitted version.

Funding

This manuscript received no funding.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Newton R, Ferlay J, Beral V, Devesa S. The epidemiology of non-hodgkin's lymphoma: comparison of nodal and extra-nodal sites. *Int J Cancer* (1997) 72(6):923–30. doi: 10.1002/(SICI)1097-0215(19970917)72:6<923::AID-IJC1>3.0.CO;2-R

2. Bradford PT, Devesa SS, Anderson WF, Toro JR. Cutaneous lymphoma incidence patterns in the united states: a population-based study of 3884 cases. *Blood* (2009) 113 (21):5064–73. doi: 10.1182/blood-2008-10-184168

3. Willemze R, Jaffe ES, Burg G, Cerroni L, Berti E, Swerdlow SH, et al. WHO-EORTC classification for cutaneous lymphomas. *Blood* (2005) 105(10):3768–85. doi: 10.1182/blood-2004-09-3502

4. Willemze R, Cerroni L, Kempf W, Berti E, Facchetti F, Swerdlow SH, et al. The 2018 update of the WHO-EORTC classification for primary cutaneous lymphomas. *Blood* (2019) 133(16):1703–14. doi: 10.1182/blood-2018-11-881268

5. Criscione VD, Weinstock MA. Incidence of cutaneous T-cell lymphoma in the united states, 1973-2002. *Arch Dermatol* (2007) 143(7):854–9. doi: 10.1001/archderm.143.7.854

6. Riou-Gotta MO, Fournier E, Mermet I, Pelletier F, Humbert P, Danzon A, et al. Primary cutaneous lymphomas: a population-based descriptive study of 71 consecutive cases diagnosed between 1980 and 2003. *Leuk Lymphoma* (2008) 49(8):1537–44. doi: 10.1080/10428190802136368

7. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: Lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

8. Sézary A, Bouvrain Y. Érythrodermie avec présence de cellules monstrueuses dans le derme et le sang circulant. *Bull la Société francaise dermatol syphiligraphie Paris.* (1938) 45:254–60.

9. Steffen C. The man behind the eponym dermatology in historical perspective: Albert sézary and the sézary syndrome. *Am J Dermatopathol.* (2006) 28(4):357–67. doi: 10.1097/00000372-200608000-00014

10. Kubica AW, Davis MD, Weaver AL, Killian JM, Pittelkow MR. Sezary syndrome: a study of 176 patients at Mayo clinic. *J Am Acad Dermatol* (2012) 67 (6):1189–99. doi: 10.1016/j.jaad.2012.04.043

11. Kim YH, Liu HL, Mraz-Gernhard S, Varghese A, Hoppe RT. Long-term outcome of 525 patients with mycosis fungoides and sezary syndrome: clinical prognostic factors and risk for disease progression. *Arch Dermatol* (2003) 139 (7):857–66. doi: 10.1001/archderm.139.7.857

12. Suzuki SY, Ito K, Ito M, Kawai K. Prognosis of 100 Japanese patients with mycosis fungoides and sezary syndrome. *J Dermatol Sci* (2010) 57(1):37–43. doi: 10.1016/j.jdermsci.2009.10.010

13. Miyashiro D, Sanches JA. Characteristics and outcomes of 727 patients with mycosis fungoides and sezary syndrome from a Brazilian cohort. *Int J Dermatol* (2022) 61(4):442–54. doi: 10.1111/ijd.15865

14. Hodak E, Amitay-Laish I. Mycosis fungoides: A great imitator. *Clin Dermatol* (2019) 37(3):255–67. doi: 10.1016/j.clindermatol.2019.01.004

15. Rodney IJ, Kindred C, Angra K, Qutub ON, Villanueva AR, Halder RM. Hypopigmented mycosis fungoides: a retrospective clinicohistopathologic study. *J Eur Acad Dermatol Venereol* (2017) 31(5):808–14. doi: 10.1111/jdv.13843

16. Mourad A, Gniadecki R. Overall survival in mycosis fungoides: A systematic review and meta-analysis. *J Invest Dermatol* (2020) 140(2):495–7 e5. doi: 10.1016/j.jid.2019.07.712

17. Sausville EA, Eddy JL, Makuch RW, Fischmann AB, Schechter GP, Matthews M, et al. Histopathologic staging at initial diagnosis of mycosis fungoides and the sezary syndrome. definition of three distinctive prognostic groups. *Ann Intern Med* (1988) 109 (5):372–82. doi: 10.7326/0003-4819-109-5-372

18. Diamandidou E, Colome M, Fayad L, Duvic M, Kurzrock R. Prognostic factor analysis in mycosis fungoides/Sezary syndrome. *J Am Acad Dermatol* (1999) 40(6 Pt 1):914–24. doi: 10.1016/S0190-9622(99)70079-4

19. Kashani-Sabet M, McMillan A, Zackheim HS. A modified staging classification for cutaneous T-cell lymphoma. *J Am Acad Dermatol* (2001) 45(5):700–6. doi: 10.1067/mjd.2001.117722

20. Talpur R, Singh L, Daulat S, Liu P, Seyfer S, Trynosky T, et al. Long-term outcomes of 1,263 patients with mycosis fungoides and sezary syndrome from 1982 to 2009. *Clin Cancer Res* (2012) 18(18):5051–60. doi: 10.1158/1078-0432.CCR-12-0604

21. Desai M, Liu S, Parker S. Clinical characteristics, prognostic factors, and survival of 393 patients with mycosis fungoides and sezary syndrome in the southeastern united states: a single-institution cohort. *J Am Acad Dermatol* (2015) 72(2):276–85. doi: 10.1016/j.jaad.2014.10.019

22. Lebowitz E, Geller S, Flores E, Pulitzer M, Horwitz S, Moskowitz A, et al. Survival, disease progression and prognostic factors in elderly patients with mycosis fungoides and sezary syndrome: a retrospective analysis of 174 patients. *J Eur Acad Dermatol Venereol.* (2019) 33(1):108–14. doi: 10.1111/jdv.15236

23. Abeni D, Frontani M, Sampogna F, Sera F, Bolli S, Corona R, et al. Circulating CD8+ lymphocytes, white blood cells, and survival in patients with mycosis fungoides. *Br J Dermatol* (2005) 153(2):324–30. doi: 10.1111/j.1365-2133.2005.06755.x

24. Agar NS, Wedgeworth E, Crichton S, Mitchell TJ, Cox M, Ferreira S, et al. Survival outcomes and prognostic factors in mycosis fungoides/Sezary syndrome: validation of the revised international society for cutaneous Lymphomas/European organisation for research and treatment of cancer staging proposal. *J Clin Oncol* (2010) 28(31):4730–9. doi: 10.1200/JCO.2009.27.7665

25. Novelli S, Garcia-Muret P, Mozos A, Sierra J, Briones J. Total body-surface area as a new prognostic variable in mycosis fungoides and sezary syndrome. *Leuk Lymphoma* (2016) 57(5):1060–6. doi: 10.3109/10428194.2015.1057894

26. Nikolaou V, Papadavid E, Patsatsi A, Siakantaris M, Economidi A, Marinos L, et al. Prognostic indicators for mycosis fungoides in a Greek population. *Br J Dermatol* (2017) 176(5):1321–30. doi: 10.1111/bjd.15000

27. Arulogun SO, Prince HM, Ng J, Lade S, Ryan GF, Blewitt O, et al. Long-term outcomes of patients with advanced-stage cutaneous T-cell lymphoma and large cell transformation. *Blood* (2008) 112(8):3082–7. doi: 10.1182/blood-2008-05-154609

28. Scarisbrick JJ, Quaglino P, Prince HM, Kim YH, Willemze Rinvestigators P. Ethnicity in mycosis fungoides: white patients present at an older age and with more advanced disease. *Br J Dermatol* (2019) 180(5):1264–5. doi: 10.1111/bjd.17602

29. Allen PB, Goyal S, Niyogusaba T, O'Leary C, Ayers A, Tarabadkar ES, et al. Clinical presentation and outcome differences between black patients and patients of other races and ethnicities with mycosis fungoides and sezary syndrome. *JAMA Dermatol* (2022) 158(11):1293–9. doi: 10.1001/jamadermatol.2022.3601

30. Geller S, Lebowitz E, Pulitzer M, Myskowski PL. Understanding racial disparities in mycosis fungoides through international collaborative studies. *Br J Dermatol* (2019) 180(5):1263–4. doi: 10.1111/bjd.17598

31. Litvinov IV, Shtreis A, Kobayashi K, Glassman S, Tsang M, Woetmann A, et al. Investigating potential exogenous tumor initiating and promoting factors for cutaneous T-cell lymphomas (CTCL), a rare skin malignancy. *Oncoimmunology* (2016) 5(7): e1175799. doi: 10.1080/2162402X.2016.1175799

32. Sibbald C, Pope E. Systematic review of cases of cutaneous T-cell lymphoma transformation in pityriasis lichenoides and small plaque parapsoriasis. *Br J Dermatol* (2016) 175(4):807–9. doi: 10.1111/bjd.14605

33. Cerroni L. Past, present and future of cutaneous lymphomas. *Semin Diagn Pathol* (2017) 34(1):3–14. doi: 10.1053/j.semdp.2016.11.001

34. Mirvish JJ, Pomerantz RG, Falo LDJr., Geskin LJ. Role of infectious agents in cutaneous T-cell lymphoma: facts and controversies. *Clin Dermatol* (2013) 31(4):423–31. doi: 10.1016/j.clindermatol.2013.01.009

35. Abrams JT, Balin BJ, Vonderheid EC. Association between sézary T cell-activating factor, chlamydia pneumoniae, and cutaneous T cell lymphoma. *Ann N Y Acad Sci* (2001) 941:69–85. doi: 10.1111/j.1749-6632.2001.tb03712.x

36. Rijlaarsdam U, Scheffer E, Meijer CJ, Kruyswijk MR, Willemze R. Mycosis fungoides-like lesions associated with phenytoin and carbamazepine therapy. *J Am Acad Dermatol* (1991) 24(2 Pt 1):216–20. doi: 10.1016/0190-9622(91)70029-2

37. Jahan-Tigh RR, Huen AO, Lee GL, Pozadzides JV, Liu P, Duvic M. Hydrochlorothiazide and cutaneous T cell lymphoma: prospective analysis and case series. *Cancer* (2013) 119(4):825–31. doi: 10.1002/cncr.27740

38. Peck JR, Frank MP, Peck LR. Was treatment the trigger? mycosis fungoides. *Am J Med* (2013) 126(12):1048–9. doi: 10.1016/j.amjmed.2013.08.007

39. Talpur R, Cox KM, Hu M, Geddes ER, Parker MK, Yang BY, et al. Vitamin d deficiency in mycosis fungoides and sezary syndrome patients is similar to other cancer patients. *Clin Lymphoma Myeloma Leuk* (2014) 14(6):518–24. doi: 10.1016/j.clml.2014.06.023

40. Litvinov IV, Tetzlaff MT, Rahme E, Habel Y, Risser DR, Gangar P, et al. Identification of geographic clustering and regions spared by cutaneous T-cell lymphoma in Texas using 2 distinct cancer registries. *Cancer* (2015) 121(12):1993–2003. doi: 10.1002/cncr.29301

41. Moreau JF, Buchanich JM, Geskin JZ, Akilov OE, Geskin LJ. Non-random geographic distribution of patients with cutaneous T-cell lymphoma in the greater Pittsburgh area. *Dermatol Online J* (2014) 20(7):13030/qt4nw7592w. doi: 10.5070/D3207023128

42. Ghazawi FM, Netchiporouk E, Rahme E, Tsang M, Moreau L, Glassman S, et al. Comprehensive analysis of cutaneous T-cell lymphoma (CTCL) incidence and mortality in Canada reveals changing trends and geographic clustering for this malignancy. *Cancer* (2017) 123(18):3550–67. doi: 10.1002/cncr.30758

43. Clough L, Bayakly AR, Ward KC, Khan MK, Chen SC, Lechowicz MJ, et al. Clustering of cutaneous T-cell lymphoma is associated with increased levels of the environmental toxins benzene and trichloroethylene in the state of Georgia. *Cancer* (2020) 126(8):1700–7. doi: 10.1002/cncr.32665

44. Jones CL, Degasperi A, Grandi V, Amarante TDGenomics England Research C, , Mitchell TJ, et al. Spectrum of mutational signatures in T-cell lymphoma reveals a key role for UV radiation in cutaneous T-cell lymphoma. *Sci Rep* (2021) 11(1):3962. doi: 10.1038/s41598-021-83352-4

45. Campbell JJ, Clark RA, Watanabe R, Kupper TS. Sezary syndrome and mycosis fungoides arise from distinct T-cell subsets: a biologic rationale for their distinct clinical behaviors. *Blood* (2010) 116(5):767–71. doi: 10.1182/blood-2009-11-251926

46. Damsky WE, Choi J. Genetics of cutaneous T cell lymphoma: From bench to bedside. *Curr Treat Options Oncol* (2016) 17(7):33. doi: 10.1007/s11864-016-0410-8

47. Larocca C, Kupper T. Mycosis fungoides and sezary syndrome: An update. *Hematol Oncol Clin North Am* (2019) 33(1):103–20. doi: 10.1016/j.hoc.2018.09.001

48. Buus TB, Willerslev-Olsen A, Fredholm S, Blümel E, Nastasi C, Gluud M, et al. Single-cell heterogeneity in sézary syndrome. *Blood Adv* (2018) 2(16):2115–26. doi: 10.1182/bloodadvances.2018022608

49. Furlan FC, Pereira BA, Sotto MN, Sanches JA. Hypopigmented mycosis fungoides versus mycosis fungoides with concomitant hypopigmented lesions: same disease or different variants of mycosis fungoides? *Dermatology* (2014) 229(3):271–4. doi: 10.1159/000363319

50. Deonizio JMD, Ascef RD, Sanches JA. Folliculotropic mycosis fungoides: clinical and epidemiological evaluation in a single center in Brazil. *Int J Dermatol* (2016) 55(5): e256–61. doi: 10.1111/ijd.13149

51. Vasconcelos Berg R, Valente NYS, Fanelli C, Wu I, Pereira J, Zatz R, et al. Poikilodermatous mycosis fungoides: Comparative study of clinical, histopathological and immunohistochemical features. *Dermatology* (2020) 236(2):117–22. doi: 10.1111/ijd.13149

52. Molloy K, Jonak C, Woei AJF, Guenova E, Busschots AM, Bervoets A, et al. Characteristics associated with significantly worse quality of life in mycosis fungoides/Sezary syndrome from the prospective cutaneous lymphoma international prognostic index (PROCLIPI) study. *Br J Dermatol* (2020) 182(3):770–9. doi: 10.1111/bjd.18089

53. Morris L, Tran J, Duvic M. Non-classic signs of sezary syndrome: A review. *Am J Clin Dermatol* (2020) 21(3):383–91. doi: 10.1007/s40257-020-00501-7

54. Scarisbrick JJ, Quaglino P, Prince HM, Papadavid E, Hodak E, Bagot M, et al. The PROCLIPI international registry of early-stage mycosis fungoides identifies substantial diagnostic delay in most patients. *Br J Dermatol* (2019) 181(2):350–7. doi: 10.1111/bjd.17258

55. Quaglino P, Pimpinelli N, Berti E, Calzavara-Pinton P, Alfonso Lombardo G, Rupoli S, et al. Time course, clinical pathways, and long-term hazards risk trends of disease progression in patients with classic mycosis fungoides: a multicenter, retrospective follow-up study from the Italian group of cutaneous lymphomas. *Cancer* (2012) 118(23):5830–9. doi: 10.1002/cncr.27627

56. Guitart J, Kennedy J, Ronan S, Chmiel J, Hsiegh Y, Variakojis D. Histologic criteria for the diagnosis of mycosis fungoides: proposal for a grading system to standardize pathology reporting. *J Cutan Pathol* (2001) 28(4):174–83. doi: 10.1034/j.1600-0560.2001.028004174.x

57. Scarisbrick JJ, Prince HM, Vermeer MH, Quaglino P, Horwitz S, Porcu P, et al. Cutaneous lymphoma international consortium study of outcome in advanced stages of mycosis fungoides and sezary syndrome: Effect of specific prognostic markers on survival and development of a prognostic model. *J Clin Oncol* (2015) 33(32):3766–73. doi: 10.1200/JCO.2015.61.7142

58. Hodak E, Amitay-Laish I, Atzmony L, Prag-Naveh H, Yanichkin N, Barzilai A, et al. New insights into folliculotropic mycosis fungoides (FMF): A single-center experience. *J Am Acad Dermatol* (2016) 75(2):347–55. doi: 10.1016/j.jaad.2016.03.009

59. Mitteldorf C, Stadler R, Sander CA, Kempf W. Folliculotropic mycosis fungoides. *J Dtsch Dermatol Ges.* (2018) 16(5):543–57. doi: 10.1111/ddg.13514

60. Demirkesen C, Esirgen G, Engin B, Songur A, Oguz O. The clinical features and histopathologic patterns of folliculotropic mycosis fungoides in a series of 38 cases. *J Cutan Pathol* (2015) 42(1):22–31. doi: 10.1111/cup.12423

61. Hodak E, Amitay-Laish I, Feinmesser M, Davidovici B, David M, Zvulunov A, et al. Juvenile mycosis fungoides: cutaneous T-cell lymphoma with frequent follicular involvement. *J Am Acad Dermatol* (2014) 70(6):993–1001. doi: 10.1016/j.jaad.2013.12.029

62. Deonizio JMD, Guitart J, Yazdan P, Mulinari-Brenner F, Sotto MN, Sanches JA. Immune privilege disruption in folliculotropic mycosis fungoides: investigation of major histocompatibility complex antigen expression. *Int J Dermatol* (2018) 57(6):675–80. doi: 10.1111/ijd.13967

63. Heymann WR. Predicting the nature of follicular mucinosis: Still a sticky situation. *J Am Acad Dermatol* (2019) 80(6):1524–5. doi: 10.1016/j.jaad.2019.04.005

64. Martinez-Escala ME, Gonzalez BR, Guitart J. Mycosis fungoides variants. *Surg Pathol Clin* (2014) 7(2):169–89. doi: 10.1016/j.path.2014.02.003

65. Kempf W, Ostheeren-Michaelis S, Paulli M, Lucioni M, Wechsler J, Audring H, et al. Granulomatous mycosis fungoides and granulomatous slack skin: a multicenter study of the cutaneous lymphoma histopathology task force group of the European organization for research and treatment of cancer (EORTC). *Arch Dermatol* (2008) 144 (12):1609–17. doi: 10.1001/archdermatol.2008.46

66. Castano E, Glick S, Wolgast L, Naeem R, Sunkara J, Elston D, et al. Hypopigmented mycosis fungoides in childhood and adolescence: a long-term retrospective study. *J Cutan Pathol* (2013) 40(11):924–34. doi: 10.1111/cup.12217

67. Valencia Ocampo OJ, Julio L, Zapata V, Correa LA, Vasco C, Correa S, et al. Mycosis fungoides in children and adolescents: A series of 23 cases. *Actas Dermo-Sifiliográficas* (2020) 111(2):149–56. doi: 10.1016/j.adengl.2020.01.007

68. Abbott RA, Sahni D, Robson A, Agar N, Whittaker S, Scarisbrick JJ. Poikilodermatous mycosis fungoides: a study of its clinicopathological, immunophenotypic, and prognostic features. *J Am Acad Dermatol* (2011) 65(2):313–9. doi: 10.1016/j.jaad.2010.05.041

69. Cesar A, Cruz M, Mota A, Azevedo F. Erythroderma. a clinical and etiological study of 103 patients. *J Dermatol Case Rep* (2016) 10(1):1–9. doi: 10.3315/jdcr.2016.1222

70. Miyashiro D, Sanches JA. Erythroderma: a prospective study of 309 patients followed for 12 years in a tertiary center. *Sci Rep* (2020) 10(1):9774. doi: 10.1038/s41598-020-66040-7

71. Vonderheid E. On the diagnosis of erythrodermic cutaneous T-cell lymphoma. *J Cutan Pathol* (2006) 33(Suppl. 1):27–42. doi: 10.1111/j.0303-6987.2006.00541.x

72. Kamijo H, Sugaya M. Two distinct variants of mycosis fungoides (MF): Folliculotropic MF and erythrodermic MF. *J Dermatol* (2019) 46(12):1136–40. doi: 10.1111/1346-8138.15114

73. Olsen E, Vonderheid E, Pimpinelli N, Willemze R, Kim Y, Knobler R, et al. Revisions to the staging and classification of mycosis fungoides and sezary syndrome: a proposal of the international society for cutaneous lymphomas (ISCL) and the cutaneous lymphoma task force of the European organization of research and treatment of cancer (EORTC). *Blood* (2007) 110(6):1713–22. doi: 10.1182/blood-2007-03-055749

74. Hristov AC, Tejasvi T, Wilcox RA. Cutaneous T-cell lymphomas: 2023 update on diagnosis, risk-stratification, and management. *Am J Hematol* (2023) 98(1):193–209. doi: 10.1002/ajh.26760

75. Dewar R, Andea AA, Guitart J, Arber DA, Weiss LM. Best practices in diagnostic immunohistochemistry: workup of cutaneous lymphoid lesions in the diagnosis of primary cutaneous lymphoma. *Arch Pathol Lab Med* (2015) 139(3):338–50. doi: 10.5858/arpa.2014-0018-CP

76. Pimpinelli N, Olsen EA, Santucci M, Vonderheid E, Haeffner AC, Stevens S, et al. Defining early mycosis fungoides. *J Am Acad Dermatol* (2005) 53(6):1053–63. doi: 10.1016/j.jaad.2005.08.057

77. Kim EJ, Hess S, Richardson SK, Newton S, Showe LC, Benoit BM, et al. Immunopathogenesis and therapy of cutaneous T cell lymphoma. *J Clin Invest.* (2005) 115(4):798–812. doi: 10.1172/JCI24826

78. Jawed SI, Myskowski PL, Horwitz S, Moskowitz A, Querfeld C. Primary cutaneous T-cell lymphoma (mycosis fungoides and sezary syndrome): part i. diagnosis: clinical and histopathologic features and new molecular and biologic markers. *J Am Acad Dermatol* (2014) 70(2):205 e1–16. doi: 10.1016/j.jaad.2013.07.049

79. Vonderheid EC, Kantor GR, Telang GH, Bujanouskas P, Kadin ME. A histo-immunopathologic and prognostic study of erythrodermic cutaneous T-cell lymphoma. *J Cutan Pathol* (2019) 46(12):913–24. doi: 10.1111/cup.13564

80. Cerroni L. Mycosis fungoides-clinical and histopathologic features, differential diagnosis, and treatment. *Semin Cutan Med Surg* (2018) 37(1):2–10. doi: 10.12788/j.sder.2018.002

81. Shamim H, Johnson EF, Gibson LE, Comfere N. Mycosis fungoides with spongiosis: A potential diagnostic pitfall. *J Cutan Pathol* (2019) 46(9):645–52. doi: 10.1111/cup.13477

82. Hossain C, Jennings T, Duffy R, Knoblauch K, Gochoco A, Chervoneva I, et al. The histological prevalence and clinical implications of folliculotropism and syringotropism in mycosis fungoides. *Chin Clin Oncol* (2019) 8(1):6. doi: 10.21037/cco.2018.10.02

83. Gru AA, Kim J, Pulitzer M, Guitart J, Battistella M, Wood GS, et al. The use of central pathology review with digital slide scanning in advanced-stage mycosis fungoides and sézary syndrome: A multi-institutional and international pathology study. *Am J Surg Pathol* (2018) 42(6):726–34. doi: 10.1097/PAS.0000000000001041

84. Wang RC, Sakata S, Chen BJ, Chang ST, Hsieh PP, Yang CS, et al. Mycosis fungoides in Taiwan shows a relatively high frequency of large cell transformation and CD56 expression. *Pathology* (2018) 50(7):718–24. doi: 10.1016/j.pathol.2018.08.008

85. Furmanczyk PS, Wolgamot GM, Kussick SJ, Sabath DE, Olerud JE, Argenyi ZB. Diagnosis of mycosis fungoides with different algorithmic approaches. *J Cutan Pathol* (2010) 37(1):8–14. doi: 10.1111/j.1600-0560.2009.01289.x

86. Ahn CS, AL A, Sangueza OP. Mycosis fungoides: an updated review of clinicopathologic variants. *Am J Dermatopathol.* (2014) 36(12):933–48. doi: 10.1097/DAD.0000000000000207

87. Prince HM, Kim YH, Horwitz SM, Dummer R, Scarisbrick J, Quaglino P, et al. Brentuximab vedotin or physician's choice in CD30-positive cutaneous T-cell lymphoma (ALCANZA): an international, open-label, randomised, phase 3, multicentre trial. *Lancet* (2017) 390(10094):555–66. doi: 10.1016/S0140-6736(17)31266-7

88. Nielsen PR, Eriksen JO, Wehkamp U, Lindahl LM, Gniadecki R, Fogh H, et al. Clinical and histological characteristics of mycosis fungoides and sezary syndrome: A retrospective, single-centre study of 43 patients from Eastern Denmark. *Acta Derm Venereol.* (2019) 99(13):1231–6. doi: 10.2340/00015555-3351

89. Raghavan SS, Hong EK, Kim YH, Kim J. Utility of CD30, ki-67, and p53 in assisting with the diagnosis of mycosis fungoides with large cell transformation. *J Cutan Pathol* (2019) 46(1):33–43. doi: 10.1111/cup.13375

90. Kash N, Massone C, Fink-Puches R, Cerroni L. Phenotypic variation in different lesions of mycosis fungoides biopsied within a short period of time from the same patient. *Am J Dermatopathol.* (2016) 38(7):541–5. doi: 10.1097/DAD.0000000000000493

91. Ponti R, Fierro MT, Quaglino P, Lisa B, Paola F, Michela O, et al. TCRgamma-chain gene rearrangement by PCR-based GeneScan: diagnostic accuracy improvement and clonal heterogeneity analysis in multiple cutaneous T-cell lymphoma samples. *J Invest Dermatol* (2008) 128(4):1030–8. doi: 10.1038/sj.jid.5701109

92. Schafernak KT, Variakojis D, Goolsby CL, Tucker RM, Martinez-Escala ME, Smith FA, et al. Clonality assessment of cutaneous b-cell lymphoid proliferations: a

comparison of flow cytometry immunophenotyping, molecular studies, and immunohistochemistry/*in situ* hybridization and review of the literature. *Am J Dermatopathol.* (2014) 36(10):781–95. doi: 10.1097/DAD.0000000000000022

93. Sufficool KE, Lockwood CM, Abel HJ, Hagemann IS, Schumacher JA, Kelley TW, et al. T-Cell clonality assessment by next-generation sequencing improves detection sensitivity in mycosis fungoides. *J Am Acad Dermatol* (2015) 73(2):228–36 e2. doi: 10.1016/j.jaad.2015.04.030

94. Vega F, Luthra R, Medeiros LJ, Dunmire V, Lee SJ, Duvic M, et al. Clonal heterogeneity in mycosis fungoides and its relationship to clinical course. *Blood* (2002) 100(9):3369–73. doi: 10.1182/blood.V100.9.3369

95. Penāte Y, Servitje O, Machan S, Fernández-de-Misa R, Estrach MT, Acebo E, et al. The first year of the AEVD primary cutaneous lymphoma registry. *Actas Dermosifiliogr* (2018) 109(7):610–6. doi: 10.1016/j.ad.2018.03.006

96. Lim HLJ, Tan EST, Tee SI, Ho ZY, Boey JJ, Tan WP, et al. Epidemiology and prognostic factors for mycosis fungoides and sezary syndrome in a multi-ethnic Asian cohort: a 12-year review. *J Eur Acad Dermatol Venereol.* (2019) 33(8):1513–21. doi: 10.1111/jdv.15526

97. Bunn PAJr., Lamberg SI. Report of the committee on staging and classification of cutaneous T-cell lymphomas. *Cancer Treat Rep* (1979) 63(4):725–8.

98. Olsen EA, Whittaker S, Willemze R, Pinter-Brown L, Foss F, Geskin L, et al. Primary cutaneous lymphoma: recommendations for clinical trial design and staging update from the ISCL, USCLC, and EORTC. *Blood* (2022) 140(5):419–37. doi: 10.1182/blood.2021012057

99. Zackheim HS, Amin S, Kashani-Sabet M, McMillan A. Prognosis in cutaneous T-cell lymphoma by skin stage: long-term survival in 489 patients. *J Am Acad Dermatol* (1999) 40(3):418–25. doi: 10.1016/S0190-9622(99)70491-3

100. Vonderheid EC, Diamond LW, Lai SM, Au F, Dellavecchia MA. Lymph node histopathologic findings in cutaneous T-cell lymphoma. a prognostic classification system based on morphologic assessment. *Am J Clin Pathol* (1992) 97(1):121–9. doi: 10.1093/ajcp/97.1.121

101. Bunn PAJr., Ihde DC, Foon KA. Recombinant interferon alfa-2a, an active agent in advanced cutaneous T-cell lymphomas. *Int J Cancer Suppl* (1987) 1:9–13. doi: 10.1002/ijc.2910390704

102. Garces S, Yin CC, Miranda RN, Patel KP, Li S, Xu J, et al. Clinical, histopathologic, and immunoarchitectural features of dermatopathic lymphadenopathy: an update. *Mod Pathol* (2020) 33(6):1104–21. doi: 10.1038/s41379-019-0440-4

103. Haththotuwa R, Zilinskiene L, Oliff J, Vydianath B, Amel-Kashipaz R, Stevens A, et al. Biopsy correlation of surface area vs. single-axis measurements on computed tomography scan of lymph nodes in patients with erythrodermic mycosis fungoides and sezary syndrome. *Br J Dermatol* (2017) 177(3):877–8. doi: 10.1111/bjd.15266

104. Fraser-Andrews EA, Mitchell T, Ferreira S, Seed PT, Russell-Jones R, Calonje E, et al. Molecular staging of lymph nodes from 60 patients with mycosis fungoides and sezary syndrome: correlation with histopathology and outcome suggests prognostic relevance in mycosis fungoides. *Br J Dermatol* (2006) 155(4):756–62. doi: 10.1111/j.1365-2133.2006.07428.x

105. Burg G. Systemic involvement in mycosis fungoides. *Clin Dermatol* (2015) 33 (5):563–71. doi: 10.1016/j.clindermatol.2015.05.008

106. Rappaport H, Thomas LB. Mycosis fungoides: the pathology of extracutaneous involvement. *Cancer* (1974) 34(4):1198–229. doi: 10.1002/1097-0142(197410)34:4<1198::AID-CNCR2820340431>3.0.CO;2-E

107. Epstein EHJ, Levin DL, Croft JDJ, Lutzner MA. Mycosis fungoides. survival, prognostic features, response to therapy, and autopsy findings. *Med (Baltimore).* (1972) 51(1):61–72. doi: 10.1097/00005792-197201000-00004

108. Graham SJ, Sharpe RW, Steinberg SM, Cotelingam JD, Sausville EA, Foss FM. Prognostic implications of a bone marrow histopathologic classification system in mycosis fungoides and the sézary syndrome. *Cancer* (1993) 72(3):726–34. doi: 10.1002/1097-0142(19930801)72:3<726::AID-CNCR2820720316>3.0.CO;2-P

109. Beylot-Barry M, Parrens M, Delaunay M, Thiebault R, Vergier B, DeMascarel A, et al. Is bone marrow biopsy necessary in patients with mycosis fungoides and sezary syndrome? a histological and molecular study at diagnosis and during follow-up. *Br J Dermatol* (2005) 152(6):1378–9. doi: 10.1111/j.1365-2133.2005.06621.x

110. Vonderheid EC, Bernengo MG. The sezary syndrome: hematologic criteria. *Hematol Oncol Clin North Am* (2003) 17(6):1367–89. doi: 10.1016/S0889-8588(03)00120-5

111. Guitart J. Sezary syndrome and mycosis fungoides flow cytometric evaluation: The clinicians' perspective. *Cytometry B Clin Cytom* (2021) 100(2):129–31. doi: 10.1002/cyto.b.21870

112. Vaughan J, Harrington AM, Hari PN, Kroft SH, Olteanu H. Immunophenotypic stability of sezary cells by flow cytometry: usefulness of flow cytometry in assessing response to and guiding alemtuzumab therapy. *Am J Clin Pathol* (2012) 137(3):403–11. doi: 10.1309/AJCP7QHH5XASTJPL

113. Trautinger F, Eder J, Assaf C, Bagot M, Cozzio A, Dummer R, et al. European Organisation for research and treatment of cancer consensus recommendations for the treatment of mycosis fungoides/Sezary syndrome - update 2017. *Eur J Cancer.* (2017) 77:57–74. doi: 10.1016/j.ejca.2017.02.027

114. Scarisbrick JJ, Hodak E, Bagot M, Stranzenbach R, Stadler R, Ortiz-Romero PL, et al. Blood classification and blood response criteria in mycosis fungoides and sezary syndrome using flow cytometry: recommendations from the EORTC cutaneous lymphoma task force. *Eur J Cancer.* (2018) 93:47–56. doi: 10.1016/j.ejca.2018.01.076

115. Hurabielle C, Michel L, Ram-Wolff C, Battistella M, Jean-Louis F, Beylot-Barry M, et al. Expression of sézary biomarkers in the blood of patients with erythrodermic mycosis fungoides. *J Invest Dermatol* (2016) 136(1):317–20. doi: 10.1038/JID.2015.360

116. Manfrere KCG, Torrealba MP, Miyashiro DR, Pereira NZ, Yoshikawa FSY, de M Oliveira L, et al. Profile of differentially expressed toll-like receptor signaling genes in the natural killer cells of patients with sézary syndrome. *Oncotarget* (2017) 8 (54):92183–94. doi: 10.18632/oncotarget.21006

117. Torrealba MP, Manfrere KC, Miyashiro DR, Lima JF, de M Oliveira L, Pereira NZ, et al. Chronic activation profile of circulating CD8+ T cells in sézary syndrome. *Oncotarget* (2017) 9(3):3497–506. doi: 10.18632/oncotarget.23334

118. Boonk SE, Zoutman WH, Marie-Cardine A, van der Fits L, Out-Luiting JJ, Mitchell TJ, et al. Evaluation of immunophenotypic and molecular biomarkers for sezary syndrome using standard operating procedures: A multicenter study of 59 patients. *J Invest Dermatol* (2016) 136(7):1364–72. doi: 10.1016/j.jid.2016.01.038

119. Delfau-Larue MH, Laroche L, Wechsler J, Lepage E, Lahet C, Asso-Bonnet M, et al. Diagnostic value of dominant T-cell clones in peripheral blood in 363 patients presenting consecutively with a clinical suspicion of cutaneous lymphoma. *Blood* (2000) 96(9):2987–92. doi: 10.1182/blood.V96.9.2987

120. Haynes L, Maue AC. Effects of aging on T cell function. *Curr Opin Immunol* (2009) 21(4):414–7. doi: 10.1016/j.coi.2009.05.009

121. Livesey A, Garty F, Shipman AR, Shipman KE. Lactate dehydrogenase in dermatology practice. *Clin Exp Dermatol* (2020) 45(5):539–43. doi: 10.1111/ced.14134

122. Alberti-Violetti S, Talpur R, Schlichte M, Sui D, Duvic M. Advanced-stage mycosis fungoides and sezary syndrome: survival and response to treatment. *Clin Lymphoma Myeloma Leuk.* (2015) 15(6):e105–12. doi: 10.1016/j.clml.2015.02.027

123. Danish HH, Liu S, Jhaveri J, Flowers CR, Lechowicz MJ, Esiashvili N, et al. Validation of cutaneous lymphoma international prognostic index (CLIPI) for mycosis fungoides and sezary syndrome. *Leuk Lymphoma* (2016) 57(12):2813–9. doi: 10.3109/10428194.2016.1173210

124. Yoo C, Yoon DH, Suh C. Serum beta-2 microglobulin in malignant lymphomas: an old but powerful prognostic factor. *Blood Res* (2014) 49(3):148–53. doi: 10.5045/br.2014.49.3.148

125. Kaito S, Kanemasa Y, Sasaki Y, Okuya T, Yamaguchi T, Funasaka C, et al. A new prognostic score comprising lactate dehydrogenase, albumin and neutrophil to lymphocyte ratio to predict sensitivity to first-line chemotherapy in patients with peripheral T-cell lymphomas. *Int J Hematol* (2018) 107(4):451–9. doi: 10.1007/s12185-017-2362-6

126. Rothe MJ, Bernstein ML, Grant-Kels JM. Life-threatening erythroderma: diagnosing and treating the "red man". *Clin Dermatol* (2005) 23(2):206–17. doi: 10.1016/j.clindermatol.2004.06.018

127. Li J, Zheng HY. Erythroderma: a clinical and prognostic study. *Dermatology* (2012) 225(2):154–62. doi: 10.1159/000342365

128. Kim YH, Hoppe RT. Mycosis fungoides and the sezary syndrome. *Semin Oncol* (1999) 26(3):276–89.

129. Diamandidou E, Colome-Grimmer M, Fayad L, Duvic M, Kurzrock R. Transformation of mycosis fungoides/Sezary syndrome: clinical characteristics and prognosis. *Blood* (1998) 92(4):1150–9. doi: 10.1182/blood.V92.4.1150

130. Sanz-Bueno J, Lora D, Monsalvez V, Maronas-Jimenez L, Postigo C, Rodriguez-Peralto JL, et al. The new cutaneous lymphoma international prognostic index (CLIPi) for early mycosis fungoides failed to identify prognostic groups in a cohort of Spanish patients. *Br J Dermatol* (2016) 175(4):794–6. doi: 10.1111/bjd.14559

131. van Doorn R, Scheffer E, Willemze R. Follicular mycosis fungoides, a distinct disease entity with or without associated follicular mucinosis: a clinicopathologic and follow-up study of 51 patients. *Arch Dermatol* (2002) 138(2):191–8. doi: 10.1001/archderm.138.2.191

132. Gerami P, Rosen S, Kuzel T, Boone SL, Guitart J. Folliculotropic mycosis fungoides: an aggressive variant of cutaneous T-cell lymphoma. *Arch Dermatol* (2008) 144(6):738–46. doi: 10.1001/archderm.144.6.738

133. Muniesa C, Estrach T, Pujol RM, Gallardo F, Garcia-Muret P, Climent J, et al. Folliculotropic mycosis fungoides: clinicopathological features and outcome in a series of 20 cases. *J Am Acad Dermatol* (2010) 62(3):418–26. doi: 10.1016/j.jaad.2009.03.014

134. Benner MF, Jansen PM, Vermeer MH, Willemze R. Prognostic factors in transformed mycosis fungoides: a retrospective analysis of 100 cases. *Blood* (2012) 119 (7):1643–9. doi: 10.1182/blood-2011-08-376319

135. Scarisbrick JJ. Survival in mycosis fungoides and sezary syndrome: How can we predict outcome? *J Invest Dermatol* (2020) 140(2):281–3. doi: 10.1016/j.jid.2019.08.440

136. Scarisbrick JJ, Kim YH, Whittaker SJ, Wood GS, Vermeer MH, Prince HM, et al. Prognostic factors, prognostic indices and staging in mycosis fungoides and sezary syndrome: where are we now? *Br J Dermatol* (2014) 170(6):1226–36. doi: 10.1111/bjd.12909

137. Sanches JA, Cury-Martins J, Abreu RM, Miyashiro D, Pereira J. Mycosis fungoides and sezary syndrome: focus on the current treatment scenario. *Bras Dermatol* (2021) 96(4):458–71. doi: 10.1016/j.abd.2020.12.007

138. Zackheim HS. Treatment of patch-stage mycosis fungoides with topical corticosteroids. *Dermatol Ther* (2003) 16(4):283–7. doi: 10.1111/j.1396-0296.2003.01639.x

139. Tarabadkar ES, Shinohara MM. Skin directed therapy in cutaneous T-cell lymphoma. *Front Oncol* (2019) 9:260. doi: 10.3389/fonc.2019.00260

140. Gilson D, Whittaker SJ, Child FJ, Scarisbrick JJ, Illidge TM, Parry EJ, et al. British Association of dermatologists and U.K. cutaneous lymphoma group guidelines for the management of primary cutaneous lymphomas 2018. *Br J Dermatol* (2019) 180 (3):496–526. doi: 10.1111/bjd.17240

141. Quaglino P, Maule M, Prince HM, Porcu P, Horwitz S, Duvic M, et al. Global patterns of care in advanced stage mycosis fungoides/Sezary syndrome: a multicenter retrospective follow-up study from the cutaneous lymphoma international consortium. *Ann Oncol* (2017) 28(10):2517–25. doi: 10.1093/annonc/mdx352

142. Bhabha FK, McCormack C, Wells J, Campbell BA, Newland K, Lade S, et al. Mycosis fungoides and sezary syndrome: Australian clinical practice statement. *Australas J Dermatol* (2021) 62(1):e8–18. doi: 10.1111/ajd.13467

143. Iqbal M, Reljic T, Ayala E, Sher T, Murthy H, Roy V, et al. Efficacy of allogeneic hematopoietic cell transplantation in cutaneous T cell lymphoma: Results of a systematic review and meta-analysis. *Biol Blood Marrow Transplant* (2020) 26(1):76–82. doi: 10.1016/j.bbmt.2019.08.019

144. Knobler R, Berlin G, Calzavara-Pinton P, Greinix H, Jaksch P, Laroche L, et al. Guidelines on the use of extracorporeal photopheresis. *J Eur Acad Dermatol Venereol* (2014) 28(Suppl 1):1–37. doi: 10.1111/jdv.12311

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 26 April 2023
DOI 10.3389/fonc.2023.1177590

OPEN ACCESS

EDITED BY
Alessandro Isidori,
AORMN Hospital, Italy

REVIEWED BY
Mattia Novo,
AOU Città della Salute e della Scienza di Torino, Italy
Pier Paolo Piccaluga,
IRCCS Azienda Opedaliera-Universitaria S. Orsola-Malpighi Hospital, Italy

*CORRESPONDENCE
Luís Alberto de Pádua Covas Lage
luis.lage@hc.fm.usp.br
luisalberto_lage@yahoo.com.br

SPECIALTY SECTION
This article was submitted to Hematologic Malignancies, a section of the journal Frontiers in Oncology

RECEIVED 01 March 2023
ACCEPTED 13 April 2023
PUBLISHED 26 April 2023

CITATION
Lage LAdPC, Culler HF, Reichert CO, da Siqueira SAC and Pereira J (2023) Angioimmunoblastic T-cell lymphoma and correlated neoplasms with T-cell follicular helper phenotype: from molecular mechanisms to therapeutic advances. *Front. Oncol.* 13:1177590. doi: 10.3389/fonc.2023.1177590

Angioimmunoblastic T-cell lymphoma and correlated neoplasms with T-cell follicular helper phenotype: from molecular mechanisms to therapeutic advances

Luís Alberto de Pádua Covas Lage[1,2]*, Hebert Fabricio Culler[1,2], Cadiele Oliana Reichert[1,2], Sheila Aparecida Coelho da Siqueira[3] and Juliana Pereira[1,2,4]

[1]Department of Hematology, Hemotherapy & Cell Therapy, University of São Paulo (USP), São Paulo, SP, Brazil, [2]Laboratory of Medical Investigation in Pathogenesis and Directed Therapy in Onco-Immuno-Hematology (LIM-31), University of São Paulo (USP), São Paulo, SP, Brazil, [3]Department of Pathology, University of São Paulo (USP), São Paulo, SP, Brazil, [4]Department of Hematology and Oncology, Hospital Alemão Oswaldo Cruz (HAOC), São Paulo, SP, Brazil

Angioimmunoblastic T-cell lymphoma (AITL) is the second most frequent subtype of mature T-cell lymphoma (MTCL) in the Western world. It derives from the monoclonal proliferation of T-follicular helper (TFH) cells and is characterized by an exacerbated inflammatory response and immune dysregulation, with predisposition to autoimmunity phenomena and recurrent infections. Its genesis is based on a multistep integrative model, where age-related and initiator mutations involve epigenetic regulatory genes, such as *TET-2* and *DNMT3A*. Subsequently, driver-mutations, such as *RhoA* G17V and *IDH-2* R172K/S promote the expansion of clonal TFH-cells ("second-hit"), that finally begin to secrete cytokines and chemokines, such as IL-6, IL-21, CXCL-13 and VEGF, modulating a network of complex relationships between TFH-cells and a defective tumor microenvironment (TME), characterized by expansion of follicular dendritic cells (FDC), vessels and EBV-positive immunoblasts. This unique pathogenesis leads to peculiar clinical manifestations, generating the so-called "*immunodysplastic syndrome*", typical of AITL. Its differential diagnosis is broad, involving viral infections, collagenosis and adverse drug reactions, which led many authors to use the term "*many-faced lymphoma*" when referring to AITL. Although great advances in its biological knowledge have been obtained in the last two decades, its treatment is still an unmet medical need, with highly reserved clinical outcomes. Outside the setting of clinical trials, AITL patients are still treated with multidrug therapy based on anthracyclines (CHOP-like), followed by up-front consolidation with autologous stem cell transplantation (ASCT). In this setting, the estimated 5-year overall survival (OS) is around 30-40%. New drugs, such as hypomethylating agents (HMAs) and histone deacetylase inhibitors (HDAi), have been used for relapsed/refractory (R/

R) disease with promising results. Such agents have their use based on a biological rationale, have significant potential to improve the outcomes of patients with AITL and may represent a paradigm shift in the therapeutic approach to this lymphoma in the near future.

KEYWORDS

angioimmunoblastic T-cell lymphoma (AITL), T-cell follicular helper phenotype (TFH), epigenetic dysregulation, *RhoA* G17V mutation, *immunodysplastic syndrome*, hypomethylating agents (HMAs), histone deacetylase inhibitors (HDAi)

1 Introduction

Angioimmunoblastic T-cell lymphoma (AITL) is a rare mature T-cell lymphoid malignancy characterized by an intense inflammatory reaction and immune dysregulation, with a broad spectrum of clinical manifestations and distinct molecular findings (1–4). Its normal counterpart is the T-follicular helper cell (TFH), a subtype of effector T-lymphocyte that plays a crucial role in the activation of B-cells and in their differentiation within the germinal center (2–4). Under physiological conditions, TFH-lymphocytes actively regulate the maturation of centroblasts into centrocytes, and subsequently, their differentiation into memory B-cells and plasma cells, in addition to ensuring homeostasis of immune tolerance mechanisms. Therefore, the clonal proliferation of TFH-cells generates an imbalance in the germinal center, characterized by pro-inflammatory phenomena, autoimmunity and hypersecretion of immunoglobulins, biological hallmarks of AITL (4–6).

Classically, mature T-cell lymphomas (MTCL), previously known as peripheral T-cell lymphomas (PTCL), derive from activated, post-thymic T-lymphocytes and are classified according to their dominant clinical presentation into predominantly nodal (nMTCL), extranodal (enMTCL), primary cutaneous (pcMTCL), and disseminated or leukemic (lMTCL) (7–9). AITL is the second most frequent subtype of nMTCL, accounting for 10-15% of all MTCL (2, 10). The 5th. edition of the Classification of Hematopoietic and Lymphoid Tissue Neoplasms proposed by the World Health Organization in 2022 (WHO-HAEM5) recognizes AITL as a clinical-pathological entity that belongs to the group of nodal lymphomas with T-cell follicular helper phenotype (nTFHL) (11). This family comprises three main entities that share the gene signature and antigenic expression of TFH-cells, including the angioimmunoblastic-type nTFHL (nTFHL-AI); the follicular-type nTFHL (nTFHL-F), associated with recurrent chromosomal rearrangement t(5;9)(q33;q22) - *ITK/SYK*; and nTFHL not otherwise specified (nTFHL-NOS). Although nTFHL-AI is the prototype of these tumors, great overlap in clinical, immunophenotypic and molecular-genetic features exists among these entities (11–13).

Described by Frizzera et al. in 1974 as a new lymphoma-like disease affecting the elderly, with a clinical-pathological pattern similar to the graft-versus-host reaction, with immunological abnormalities and a frequently fatal course, AITL was initially called "*angioimmunoblastic lymphadenopathy with dysproteinemia*" (14). Subsequently, other authors reported series of similar cases under the designations "*immunoblastic lymphadenopathy*" and "*lymphogranulomatosis X*" (15, 16). Although most cases had a recurrent and fatal course, it was initially considered as a non-neoplastic hyperimmune reaction triggered by an unknown stimulus. However, the clonal nature of AITL was definitively demonstrated in the 1980s by studies that revealed recurrent chromosomal abnormalities and monoclonal pattern rearrangement of the T-cell receptor (TCR) gene in tissue samples from its patients (17, 18). This led to its recognition as a subtype of PTCL by the *Revised European American Lymphoma Classification* (REAL) in 1994 (19).

In the 2000s, studies involving gene expression profiling (GEP) determined that the normal counterpart of AITL was the TFH-cell (4, 6, 20). After 2010, different groups have described recurrent and inactivating mutations of epigenetic regulatory genes, involved in DNA-methylation and histone-deacetylation, in tumor samples from AITL patients. These findings have contributed significantly to understanding the biology of AITL and suggest a central role for recurrent somatic mutations involving epigenetic modulation in nTFHL oncogenesis (21–23). Finally, in 2014, the G17V somatic mutation of the *RhoA* gene (*Ras-homolog family member A*) was described as a molecular biomarker of AITL, being found in up to 60-70% of MTCL cases with TFH-phenotype (24, 25).

2 Epidemiology

AITL is a rare lymphoproliferative disorder, accounting for 1-2% of all non-Hodgkin's lymphomas (NHL) and 15-20% of MTCL (3). According to the *International Peripheral T-cell and natural-killer/T-cell Lymphoma Project* (ITCLP), a pioneering study that described the pathological characteristics and clinical outcomes of more than 1,300 patients with MTCL from different regions of the world, AITL was the second most prevalent subtype of MTCL in the Western world, just behind peripheral T-cell lymphoma, not otherwise specified (PTCL, NOS) (10). Characteristically, AITL presents an inverse geographical tropism to other subtypes of T-cell lymphomas, being more common in Europe, particularly in the British Islands and Nordic countries, comprising 28.7% of MTCL,

and less prevalent in Asia and North America, where it accounts for 17.9% and 16.0% of T-cell lymphomas, respectively (10). In the United States, its incidence is low, with 0.05 new cases per 100,000 inhabitants per year (26). On the other hand, epidemiological data from the *French National Cancer Agency* pointed to AITL as the most prevalent subtype of MTCL, accounting for 36.1% of cases diagnosed between 2010-2013 (27).

Epidemiological data from a subgroup analysis of the prospective *T-Cell Project* study, involving 282 AITL patients recorded between 2006 and 2018, revealed lower prevalence of this lymphoma in South America compared to other geographic regions (7% of MTCL, p=0.002) (28). This prevalence was similar to that reported by our group, which demonstrated 10.4% of AITL cases among 124-Brazilian patients with nMTCL diagnosed and treated at the Hematology Service of the University of São Paulo between 2000 and 2019 (29). This observation supports that regional differences in the incidence of AITL may indeed exist, which can be explained in part by the high incidence of other MTCL subtypes in Central and South America, such as extranodal NK/T-cell lymphoma (ENKTL) and adult T-cell leukemia/lymphoma (ATLL), both subtypes associated with oncovirus endemic in these areas of the globe.

AITL preferentially affects elderly patients, with a median age of 60 to 65 years. Although there is no clear predilection for gender and ethnicity, some studies indicate a slight predominance in Caucasian males (10, 28–30).

3 Etiopathogenesis

The oncogenesis of AITL occurs in an integrative model characterized by close interaction among neoplastic cells, the tissue immune microenvironment (TIME) and Epstein-Barr virus (EBV) (4, 31, 32). AITL originates from the neoplastic transformation of TFH cell, a subtype of T-CD4+ effector lymphocytes that reside in the germinal center and are characterized by high expression of the chemokine receptor CXCR5 (*C-X-C motif receptor 5*), chemokine CXCL-13 (*C-X-C motif ligand 13*), ICOS (*CD28-related inducible T-cell co-stimulator*), CD154, CD40L and NFATC1 (6, 33–36). TFH-cells participate in the formation of the germinal center, leading to the expansion of lymphoid B-cells and promoting their differentiation into plasma cells and memory B-cells, as well as promoting the proliferation of follicular dendritic cells (FDC). These phenomena are regulated by the secretion of CXCL-13 and IL-21 by the TFH-cells (31, 32, 37).

Under normal conditions, naïve T-CD4+ lymphocytes interact with dendritic cells in the germinal center, promoting the activation of ICOS protein on T-cells and consequently activation of the PI3K pathway, leading to up-regulation of BCL-6, a critical transcriptional factor for TFH-cell differentiation (38, 39). Subsequently, activated T-cells will overexpress PD-1 and CXCR5, transforming into precursor TFH-cells, which will migrate to the periphery of the follicle to interact with antigen-specific B-cells. This interaction promotes a reaction in the germinal center that leads to the terminal maturation of TFH-cells, with activation of the JAK-STAT pathway through the secretion of IL-6 and IL-21 (32, 40).

The TIME elements represent the majority of the cellular component in a lymph node involved by AITL. Similar to classic Hodgkin's lymphoma (cHL), reactive T-lymphocytes, B-lymphocytes, plasma cells, macrophages, dendritic cells, and endothelial cells make up the AITL's tumor microenvironment. Such cells promote an exacerbated inflammatory response and immune dysregulation, considered as biological hallmarks of AITL and responsible for the recurrent infectious and autoimmune complications observed in this tumor. The exacerbated secretion of CXCL-13 and VEGF-1 (*vascular endothelial growth factor-1*) by mutated TFH-cells promote the expansion and proliferation of FDC and high endothelial venules (HEV) commonly found in AITL (31). Studies in animal models demonstrated that in AITL, B-cells are retained for a longer time in the germinal center and have a greater opportunity to interact with TFH-cells. IL-21 secretion by TFH-cells induce expansion of B-lymphocytes, which terminally differentiate into immunoglobulin-secreting plasma cells. Polyclonal plasmacytosis recurrently occurs in tissues affected by AITL, and polyclonal gammopathy is a characteristic laboratory finding of this neoplasm (31, 41). Dendritic cells and macrophages of the tumor microenvironment are hyperactivated and secrete high levels of IL-6, an agent with pro-inflammatory and pro-proliferative activity on the TFH malignant clone (4).

The Epstein-Barr virus (EBV) is an oncogenic infectious agent implicated in the development of several subtypes of lymphomas. In up to 80% of AITL cases, EBV is detected by EBER-ISH (Epstein-Barr encoded RNAs–*in situ* hybridization) inside large B-immunoblasts morphologically similar to Reed-Sternberg cells (RS-cells) (42, 43). Additionally, monoclonal rearrangement of the immunoglobulin gene (IgVH) is seen in up to one third of AITL (44). Therefore, some authors were able to establish an association between high-tissue EBV viral load, the occurrence of B-monoclonality, and the development of diffuse large B-cell lymphoma (DLBCL), which can develop concomitantly or in a late evolutionary phase of AITL (4, 31, 45). It is speculated that EBV B-cell infection results from the characteristic immune dysregulation observed in AITL, however EBV may be able to modulate disease progression as well as the development of truly B-cell malignancies, such as DLBCL or plasmablastic lymphoma (PL).

Concerning to molecular aspects, AITL is characterized by the presence of recurrent mutations involving the *RhoA* gene and epigenetic regulatory genes, implicated in the processes of DNA-methylation, histone-deacetylation, and regulation of nuclear chromatin remodeling, such as *IDH-2*, *DNMT3A* and *TET-2* (3). The *RhoA* gene, located on chromosome 3, encodes a small GTPase that regulates cell migration, intracellular signaling, proliferation and survival. It also participates in the conformation of the cytoskeleton, signaling of the T-cell receptor (TCR) pathway, and plays a central role in the ontogenesis of T-lymphocytes (46–49). In its active state, the RhoA protein binds to guanine-triphosphate (GTP) and in its inactive state it binds to guanine- diphosphate (GDP). Mutant RhoA protein (RhoA-mut) has compromised GTP binding, which leads to alterations in the *RhoA* signaling pathway

and impairment of its biological functions (24, 50). The *RhoA* G17V mutation, associated with loss of GTPase function, is recurrently found in up to 60-70% of AITL cases, and is currently considered a diagnostic biomarker for nMTCL-TFH-phenotype (24, 25, 50–52). Recent studies using animal models point to a clear relationship between the *RhoA* G17V mutation and TFH-cell differentiation, establishing a pathogenic link between this molecular alteration and AITL development (49, 53, 54). Based on these findings, it is postulated that the *RhoA* G17V mutation is a driver event for the development of nMTCL-TFH-phenotype, although it can still be found in neoplasms of different histogenesis, such as Burkitt's lymphoma (BL), adult T-cell leukemia/lymphoma (ATLL) and gastric adenocarcinomas (24, 50, 55–57).

TET-2 mutations (*Ten-eleven translocation 2*) were originally described in myeloid malignancies, such as myelodysplastic syndromes (MDS), chronic myelomonocytic leukemias (CMML), and acute myeloid leukemias (AML). However, later, these mutations have been identified recurrently in MTCL, particularly in those with TFH-phenotype (22, 58–60). Loss-of-function mutations on *TET-2* gene occur in up to 80% of AITL. This gene encodes an oxyglutarate-oxygenase that catalyzes the oxidation of DNA 5-methylcytosine (5-mC) to 5-hydroxy-methylcytosine (5-hmC) (61, 62). Experimental studies demonstrated that *TET-2*-mutant mices were prone to developing CMML and nMTCL-TFH-phenotype. These same studies associate suppression of the *TET-2* gene function with upregulation of the BCL-6 transcription factor and selection for differentiation of näive CD4+ T-cells for mature TFH-cells (21, 63, 64). Although *TET-2* mutations are considered secondary events in nMTCL-TFH-phenotype, different studies demonstrate an association of *TET-2* and *RhoA* G17V mutations in these neoplasms, suggesting a biological cooperation between both mutations to promote AITL development (49, 65). Recently, our research group demonstrated a high-rate of co-occurrence between *TET-2* and *RhoA* mutations in Brazilian patients with non-anaplastic nMTCL, confirming the data found in previous experimental studies (51, 52). In our population, composed of 59 patients with nMTCL, the *RhoA*-mut/*TET-2*-mut association was demonstrated in 42.8% of cases with nMTCL-TFH-phenotype, also being associated with a high-tumor volume represented by bulky disease ≥ 7 cm and decreased overall response rates (ORR) to primary treatment based on anthracyclines (51).

Up to 30-40% of AITL have a mutation in the *IDH-2* gene (*isocitrate dehydrogenase 2*) (23, 66). Physiologically, IDH enzymes catalyze the conversion of isocitric acid to 2-alpha-ketoglutarate (2-KG). Mutant IDH-2 enzymes will contribute to the malignant phenotype through the production of the aberrant metabolite 2-hydroxy-glutarate (2-HG) (23, 67). 2-HG inhibits the activity of TET family proteins and histone deacetylases, resulting in the production of 5-hmC and subsequent repression of tumor suppressor genes, resulting in the promotion of the malignant phenotype (68). Although it occurs recurrently in other neoplasms, such as AML and high-grade gliomas, *IDH-2* mutations, when present in MTCL, are practically exclusive to AITL and are usually restricted to the arginine residue 172 (R172K and R172S) (66, 69). Unlike cases of AML, where *IDH-2* and *TET-2* mutations are mutually exclusive and associated with adverse prognosis, in AITL they are usually seen concurrently, which suggests a cooperative effect between both mutations for expansion and differentiation of malignant TFH-cells (66).

Many other loss-of-function mutations, such as those involving the *DNMT3A* (*DNA methyl-transferase 3A*) gene, found in 10-40% of AITL, and mutations in genes associated with the TCR signaling pathway, such as *PLCG1*, *CD28*, *VAV1* and *FYN*, found in up to 50% of AITL also contribute to the development of the malignant phenotype in nMTCL-TFH-phenotype, although the latter are also frequently observed in PTCL, NOS and ATLL cases (25, 66, 70, 71).

Therefore, we conclude that the tumorigenesis of AITL and other nMTCL-THF-phenotype is a complex process, involving multiple steps of mutational phenomena associated with epigenetic regulation, leading to differentiation and expansion of malignant TFH-cells. These clonal TFH-cells, in turn, interact with a defective immune microenvironment that is permissive to the development of the neoplasm. In the meantime, self-reactivity phenomena and the EBV oncogenic activity, which proliferates in an immunodeficient microenvironment, favor tumor propagation and generate the classic immunodysplastic findings, typical of this group of lymphomas. Currently, it is speculated that age-related loss-of-function mutations in the *TET-2* and *DNMT3A* genes, which occur at an early-stage of hematopoietic development, constitute the initial events that promote the tumor (*first hit*), since they are found in clonal TFH-cells, in B-cells and in CD34+ myeloid precursors. Subsequently, the occurrence of specific-mutations, such as *RhoA* G17V and *IDH-2* R172K/S, observed exclusively in TFH-cells, lead to a second hit that modulates the differentiation and proliferation of clonal TFH-cells, orchestrating the development of AITL (32). It is now known that AITL, CMML and AITL-derived DLBCL are neoplasms that share common age-related ancestral mutations (*TET-2* and *DNMT3A*). Following this multi-step process, specific mutations such as *RhoA* G17V/*IDH-2*, *NPM-1* and *NOTCH-1* will predispose to the emergence and expansion of malignant clones of AITL, CMML and DLBCL, respectively, all of them considered as correlated hematopoietic neoplasms (46, 72–74). Figure 1 demonstrates the previously summarized integrative pathogenic model of AITL.

4 Histopathological diagnosis

As for other MTCL, excisional lymph node biopsy is the preferred procedure for establishing the diagnosis of AITL and other nMTCL-TFH-phenotype. Although incisional or core needle biopsies can establish this diagnosis, a complete architectural evaluation of the affected lymphoid tissue in the AITL is usually required, with a detailed analysis of the tumor compartment and the exuberant immune microenvironment, which many times can only be achieved with histopathological analysis of the entire lymph node. Additionally, large amounts of tumor tissue are needed to carry out a broad immunohistochemical panel, *in situ* hybridization (ISH) for EBV and complementary molecular tests, such as T-cell clonality assays by PCR and mutational profiling involving the *RhoA* gene and other epigenetic regulators by Sanger sequencing or next-generation sequencing (NGS) techniques.

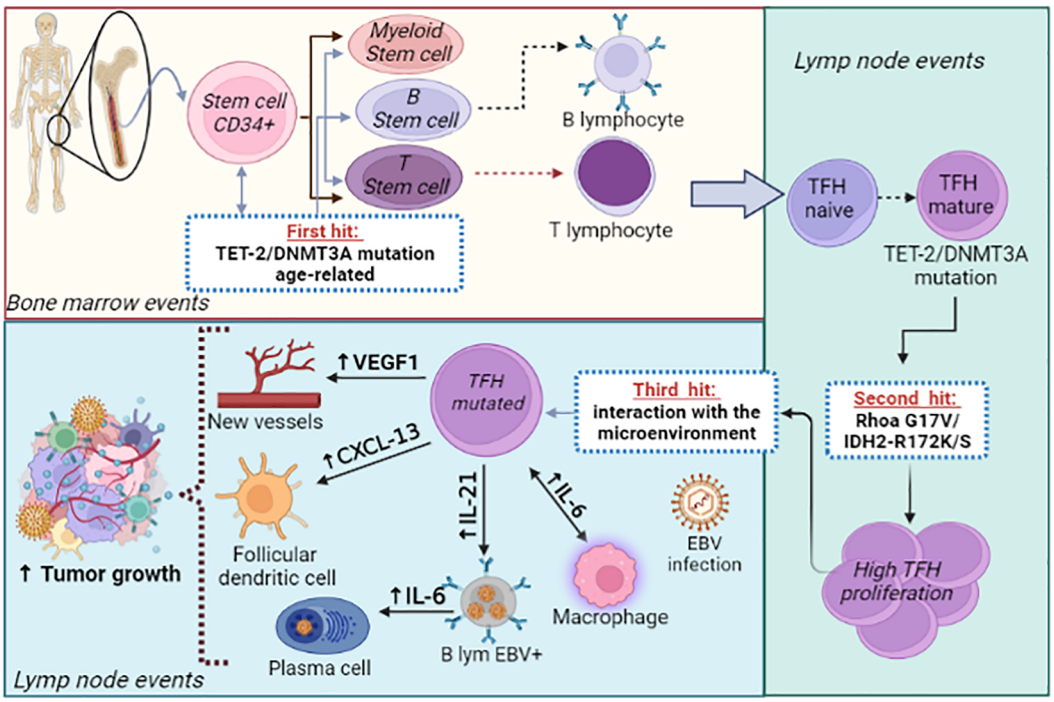


FIGURE 1
Integrative biologic model for AITL pathogenesis. AITL development follows a "*multi-step*" process. The initiating event of neoplasia ("*first-hit*") involves age-related mutations in epigenetic regulatory genes (*TET-2* and/or *DNMT3A*) that compromise the pluripotent hematopoietic stem cells-CD34+ with propagation to myeloid precursor cells, B- and T-lymphoid cells. Subsequently, the naïve T-CD4+ cell matures to TFH-cell in the germinal center and experiences specific-disease ("driver") mutations, such as the *RhoA* G17V and/or *IDH-2* R172 K/S ("*second-hit*"). Finally, mature and mutated TFH-cells expand in the germinal center from the interaction with several elements of the tumor immune microenvironment (FDC, macrophages, B-cells, endothelial-cells and EBV), which provide an environment marked by immune-dysregulation and pro-inflammatory activity, highly permissive for the propagation and dissemination of the neoplasm ("*third -hit*").

Classically, AITL can be characterized by partial lymph node involvement, or more commonly by diffuse nodal infiltration with complete rupture of the normal tissue architecture, as well as capsular and perinodal infiltration frequently sparing the peripheral sinus, and absence of residual B-cell lymphoid follicles (2, 75). According to Attygalle et al., three distinct architectural patterns can be observed in the AITL. Pattern I, or AITL with hyperplastic follicles, found in 15% of cases, exhibits partially preserved nodal architecture and hyperplastic lymphoid follicles with poorly developed mantle zones. In pattern II, or AITL with depleted follicles, observed in 25% of cases, occasionally depleted follicles are present and the FDC meshwork is unchanged or minimally expanded (20). In pattern III, the so-called classic AITL without follicles, which occurs in 60% of cases, there is complete loss of the architecture of the affected lymph node, capsular and perinodal infiltration sparing the peripheral sinus, and absence of residual B-cell lymphoid follicles (20). Different histopathological patterns have been documented in consecutive biopsies and seem to represent progressive stages of the neoplasia. This purely reflects a morphological evolution and is not associated with clinical progression, since patients with pattern I usually present with symptomatic and advanced-stage disease (20, 76).

The histopathological findings of classic AITL (pattern III) include: (I) diffuse and polymorphic infiltrate, composed of a variable proportion of tumor cells with TFH-phenotype, interspersed with small reactive lymphocytes, histiocytes or epithelioid cells, large B-immunoblasts, eosinophils and plasma cells; (II) prominent proliferation of high endothelium venules (HEV) in an arborescent pattern; (III) expansion of the FDC meshwork, often accompanying the proliferated vessels; and (IV) expansion of large immunoblasts of B-lymphoid phenotype in the paracortical area, with frequent Reed-Sternberg-like (RS) morphology and expressing small-RNAs encoded by the EBV (EBERs) (2, 11, 20, 77).

In AITL, the neoplastic cells with TFH-phenotype are usually small to medium-sized and have mild atypia. Its cytoplasm is abundantly clear ("*clear cells*"), with a tendency to form small aggregates surrounding HEV (78). Expansion of B-immunoblasts is usually present in the paracortex and may mimic reactive immunoblastic hyperplasia or cHL by its RS-like morphology (79). Tissue plasmacytosis can represent an exuberant finding in several cases, obscuring the tumor cells (2, 77). The amount and distribution of neoplastic cells and the different components of the tumor microenvironment differ widely between cases; however, usually clear tumor cells comprise the minority (about 30%) of the nucleated elements in AITL (2).

In the immunohistochemical study, the lymph nodes involved by AITL show expansion of the paracortex by a diffuse infiltrate of CD4+ T-cells. B-cell lymphoid areas are usually reduced, although large speckled B-immunoblasts, often CD30+ and EBV+ are found

among tumor cells (2, 77). Usually, neoplastic cells show expression of pan-T markers, such as CD2+, CD3+ and CD5+, although loss of expression of these antigens, particularly surface CD3 (sCD3) and CD7 can occur in up to 50% of cases (27, 80). By flow cytometry, loss of sCD3 is commonly observed and can be an important diagnostic clue (81, 82). Tumor cells are CD4+, although the CD4/CD8 ratio is usually preserved in lymph nodes involved by AITL (83). A portion of AITL cases (20-30%) may exhibit partial expression of the Ki-1 marker (CD30) (2, 77).

Being a neoplasm of TFH-origin, AITL displays multiple TFH-related antigens, including PD-1/CD279 (*programmed death-1*), CD10 (a metalloendopeptidase called *common acute lymphoblastic leukemia antigen*/CALLA), BCL-6 (*B-cell lymphoma 6 protein*), CXCL-13 (*C-X-C motif chemokine ligand 13*), ICOS (*inducible T-cell costimulator*), SAP (*signaling lymphocyte activation molecule [SLAM]-associated protein*) and CXCR-5 (*C-X-C motif chemokine receptor 5*) (11, 35, 36, 77, 84, 85). It should be noted that these markers are not specific, so it is recommended that at least two of them, or preferably three, be expressed by the neoplastic cell to define that a nMTCL has a TFH-phenotype (2, 77). Among the TFH-markers, PD-1/CD279 and ICOS are reported to be more sensitive, while CXCL-13 and CD10 are less sensitive but more specific (86).

In the immunohistochemical study, the exuberant vascular proliferation characteristically observed in AITL may be better evidenced by the endothelial markers CD31 and CD34, as well as the expanded FDC meshwork, evidenced by staining for CD21, CD23 and CD35 antigens. CD138 staining will help to show exuberant plasmacytosis, often seen in AITL. Although plasma cells are usually polyclonal, truly clonal plasma cell proliferations have rarely been described (87, 88).

Polyclonal or truly monoclonal EBV+ lymphoid B-cell proliferation will often be seen in AITL and appear to result from immunoderegulation mediated by tumor TFH-cells. Different studies demonstrate that up to 80% of AITL cases may contain a variable number of EBV+ B-cells, ranging from isolated foci of paracortical immunoblasts to true EBV-associated mature B-cell lymphoproliferation, such as EBV +DLBCL, which can be observed both in the initial diagnosis of AITL (synchronous neoplasms) or during disease progression (89–91).

Skin and bone marrow are also commonly affected in AITL. In the skin, histological findings are usually subtle, including perivascular and periadnexal lymphoid infiltrates with mild atypia, conditions that are difficult to distinguish from inflammatory dermatoses. Immunohistochemistry for TFH-markers and T-cell clonality assays (PCR) can be very useful to distinguish reactive paraneoplastic skin rash from a cutaneous tumor infiltration by AITL (77). In the bone marrow, neoplastic infiltration by AITL can be characterized by nodular lymphoid infiltrates of paratrabecular or interstitial distribution, commonly accompanied by trilineage dysplasia, fibrosis, and plasmacytosis. TFH-associated markers usually have a lower yield in the bone marrow than in the lymph nodes, possibly due to impaired antigenic recovery influenced by sample decalcification (2, 77). Figures 2, 3 summarize the main histological and immunohistochemical findings observed in classic cases of AITL in lymph node and bone marrow.

AITL shares many histopathological findings with other nMTCL-TFH-phenotype, so-called AITL-related disorders, due to the overlap of phenotypic and molecular aspects. In follicular-type nTFHL (nTFHL-F), the affected lymph nodes present a predominantly nodular growth pattern, similar to that observed in follicular lymphoma. However, the exuberant inflammatory background observed in AITL, as well as vascular proliferation with HEV and expansion of the FDC meshwork are not observed (2, 11, 12, 77).

5 Clinical and laboratory features

Although up to 20-25% of AITL cases may have an indolent and fluctuating course, the disease usually has a clinical spectrum marked by high aggressiveness (1, 10, 28, 80). Clinically, the disease affects elderly men, with a median age around the sixth decade of life, as generalized lymphadenopathy associated with manifestations of autoimmunity and immunodeficiency (31, 80). The clinical manifestations resulting from hyper-inflammatory reaction, autoimmunity, and immunodeficiency, causing greater susceptibility to infections, constitute the so-called "*angioimmunoblastic lymphoma syndrome*" or "*immunodysplastic syndrome*" (3, 31). Prototypically, AITL manifests as an acute or subacute systemic disease after administration of drugs or viral infections, establishing a wide differential diagnosis with pharmacodermias, collagenosis and exanthematic viral diseases, which has led some authors to recognize this disorder as "*the many-faced lymphoma*" (3).

The majority of AITL patients have advanced-stage disease (Ann Arbor III or IV), although rare cases of early-disease (I or II) have been reported and constitute less than 10% of clinical presentations (3, 10, 31). Although considered within the scope of predominantly nodal MTCL, extranodal involvement is not infrequent in AITL. Bone marrow infiltration has been reported in up to 70% of cases, as well as involvement of other extranodal sites, such as the lungs and the gastrointestinal tract (3, 31). Peripheral blood involvement by small circulating clones, particularly identified by flow cytometry, usually with sCD3-/CD4+/CD10+ phenotype can be identified (81).

Characteristically, AITL patients present with constitutional symptoms (B-symptoms), such as fever, weight loss, and night sweats. Lymphadenopathy is usually generalized and small (< 1-3 cm), although our group observed a high occurrence of bulky disease ≥ 7 cm in South American patients (29, 31). Hepato-splenomegaly is frequently observed, as well as skin rash and pruritus, the latter occurring in up to 50% of cases, and may be reactive or correspond to frank cutaneous tumor infiltration by the TFH-clone (3, 31, 77). The typical immunoderegulation of AITL predisposes to immunodeficiency and greater susceptibility to bacterial, fungal, and opportunistic infections (3, 31, 89). Manifestations associated with increased vascular permeability, resulting from the hypersecretion of VEGF-1 (92, 93), such as peripheral edema, pleural effusion and ascites are observed in up

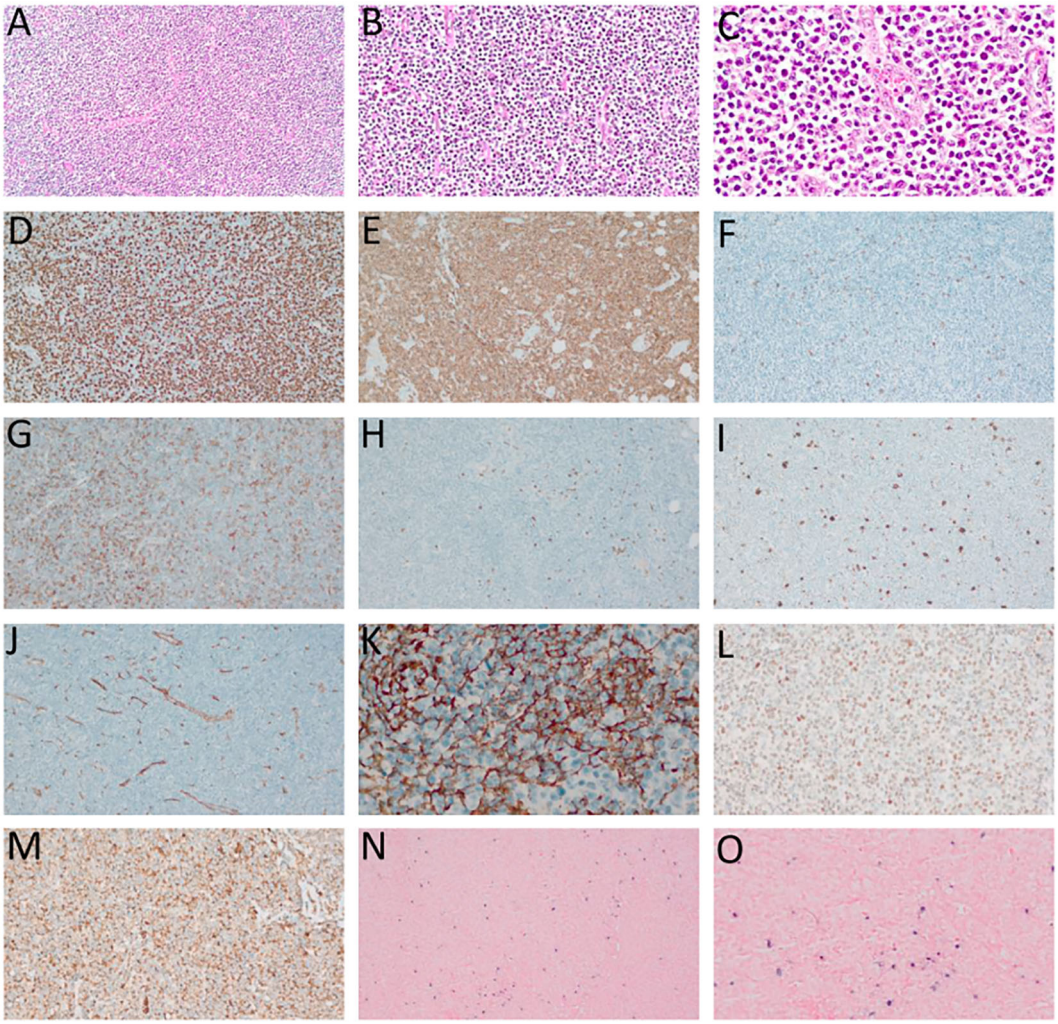


FIGURE 2
Angioimmunoblastic T-cell lymphoma (AITL). **(A–C)** – Hematoxylin-eosin (HE), optical microscopy, magnifications of 10 x **(A)**, 20 x **(B)** and 40 x **(C)**. Proliferation of small/medium sized atypical lymphoid cells diffusely infiltrating the lymph node with marked vascular proliferation. **(D)** high-rate of cell proliferation index - positive Ki-67 in more than 80% of the nuclei of atypical lymphoid cells. **(E)** Strong and diffuse labeling for the pan-T CD3 antigen. **(F)** Rare large CD20-positive cells in the paracortical region ("immunoblasts"). **(G)** CD4-positive in most neoplastic cells. **(H)** CD8-negative in neoplastic cells and positive in rare small reactive lymphocytes. **(I)** CD30-positive in large, expanded cells in the paracortical region with RS-like morphology. **(J)** Exuberant vascular proliferation with high endothelial venules (VEA) enhanced by CD34. **(K)** Expanded follicular dendritic cell (FDC) meshwork, labeling for CD21. **(L)** BCL6 positive and **(M)** CD10 positive, both markers of TFH origin. **(N, O)** *In situ* hybridization (ISH) for EBV, positivity for EBERs (Epstein-Barr small encoded-RNAs), staining in black, "speckled" pattern, revealing EBV staining in RS-like immunoblasts in the paracortex. **(D–F, H, L–N)** – Optical microscopy, 10 x magnification. **(I, J, O)** – Optical microscopy, 20 x magnification; **(K)** – Optical microscopy, 40 x magnification.

to 30% of cases (3, 30). Autoimmune phenomena such as leukocytoclastic vasculitis, thyroiditis, arthralgia and/or arthritis are recurrently observed (3, 31). Laboratory findings include autoimmune hemolytic anemia (Coombs positive), polyclonal hypergammaglobulinemia, elevated erythrocyte sedimentation rate (ESR), eosinophilia, lymphopenia, and thrombocytopenia, although some patients may present with atypical lymphocytosis with circulating hyper basophilic lymphoid cells (3, 31, 94, 95). Serum markers of autoimmunity, such as antinuclear antibodies (ANA), rheumatoid factor, anti-smooth muscle antibodies, circulating immune complexes, cryoglobulins and cryoagglutinins are described in up to 20-30% of cases (3, 30, 31). Figure 4 summarizes the spectrum of clinical and laboratory manifestations recurrently observed in AITL and related disorders.

In opposition to other MTCL, such as ALK1-positive anaplastic large-cell lymphoma (ALK1+ ALCL), associated with the t(2;5)(p23;q35), and T-cell prolymphocytic leukemia (T-PLL), associated with inv (14)(q11;q32)/t(14;14)(q11;q32), AITL does not present a characteristic or recurrent chromosomal abnormality. However, cases of nTFHL-F are recurrently associated with t(5;9)(q33;q22) and consequent *splenic tyrosine kinase* (SYK) upregulation (96). In AITL, more than 90% of cases demonstrate clonal aberrations, the most common being trisomy of chromosomes +3, +5, +21, +X and deletion of chromosome 6q-. Inactivation of the *TP53*, associated with the abnormalities del(17p-)/-17 and complex karyotype are

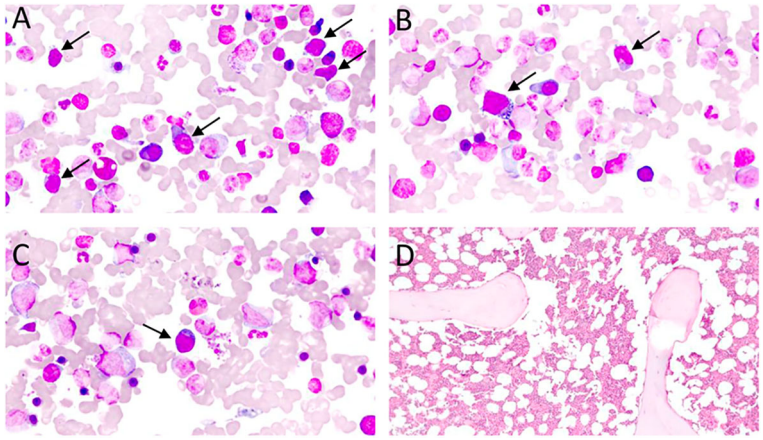


FIGURE 3
(A–C) – Bone marrow aspirate, Leishman staining, optic microscopy, 70 x magnification: subtle bone marrow infiltration by atypical lymphoid cells (black arrows) in an AITL case. Atypical lymphoid cells are small/medium-sizes, have loose chromatin, cytoplasm with pronounced basophilia, and some forms have microvacuolation, making up about 10% of the nucleated elements in the bone marrow specimen. **(D)** – Bone marrow biopsy, Hematoxylin-Eosin (HE), optical microscopy, 10 x magnification. Usually, lymphomatous infiltration of the bone marrow in AITL is subtle and difficult to characterize in HE, requiring immunohistochemical staining for TFH-associated antigens for better revelation of neoplastic cells.

rare clonogenic alterations in AITL associated with poor outcomes (94, 97). T-cell and B-cell clonality assays using PCR techniques to assess clonal rearrangements in the TCR and immunoglobulin heavy-chain (IgVH) gene occur in 80-100% and up to 50% of AITL, respectively (31, 98).

Recent studies involving NGS techniques have demonstrated variable proportions of recurrent mutations involving the *RhoA* GTPase, epigenetic regulatory genes (*IDH-2*, *TET-2* and *DNMT3A*) and genes involved in the TCR signaling pathway (*PLCγ*, *CD28*, *FYN* and *VAV-1*) in patients with AITL (99, 100). *RhoA* mutations, particularly G17V have been reported in 60-70% of cases, *TET-2* in 47-83%, *DNMT3A* in 20-30% and *IDH-2* R172K/S in 20-45% of cases. Mutations in genes associated with the TCR signaling pathway are rarer, occurring in 14% of cases, 9-11%, 3-4% and 5%, respectively, for *PLCγ*, *CD28*, *FYN* and *VAV-1* genes (13, 23–25, 50, 60, 71, 101).

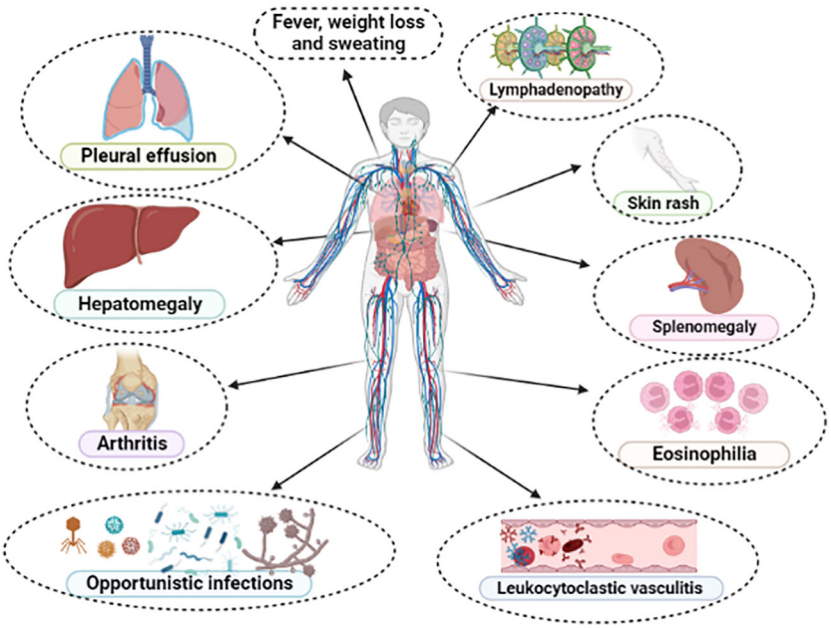


FIGURE 4
Main clinical and laboratory findings presented in typical cases of angioimmunoblastic T-cell lymphoma (AITL). All these manifestations compose the so-called "*angioimmunoblastic lymphoma syndrome*" or "*immunodysplastic syndrome*", establishing differential diagnosis with drug-reactions, collagenoses and exanthematic viral infections.

6 Outcomes and prognostic factors:

Although it is a tumor with a highly fatal course, 32-41% and 18-38% of AITL patients will survive after 5 years of diagnosis and will remain event-free during this period, respectively, according to data from the *International T-cell Lymphoma Project* (ITCLP), as well as to other large collaborative studies (10, 27, 30, 80, 102). In the last two decades, since the initial report by the *ITCLP* in 2008, the outcomes of patients with AITL have not presented significant increments, despite the great advances in its biological knowledge. Newly published data involving a subgroup of 282 AITL cases recorded in the *ITCLP* from 2006 to 2018 demonstrate OS and progression-free survival (PFS) at 5 years of 44% and 32%, respectively (28). In parallel, this same study pointed to progression of disease within 24 months from diagnosis (POD-24) as a powerful predictor of prognosis in patients with AITL, with an estimated 5-year OS of 63% versus only 6% for patients without POD-24 and with POD-24, respectively, $p<0.001$ (28). Data from our research group, involving 124- Brazilian cases with nMTCL diagnosed and treated at the University of São Paulo, from 2000 to 2019, with 10.4% (13/124) of AITL among all cases, revealed similar outcomes. In our cohort, the estimated 2-year OS and PFS for South American patients with AITL was 53.8% and 45.5%, respectively, corroborating the poor clinical outcomes associated with this tumor, as previously reported in other studies conducted in Europe and North America (29).

Prognostic scores, such as the *International Prognostic Index* (IPI) and the *Prognostic Index for Peripheral T-cell Lymphoma Unspecified* (PTCL-U score), the latter including the variables age ≥ 60 years, LDH ≥ normal value, performance status according to the scale *Eastern Cooperative Oncology Group* (ECOG) ≥ 2 and histopathological bone marrow involvement by NHL, in addition to reduced platelet count, have been able to predict prognosis in AITL patients (10, 30, 80). Recently, Advani et al. reported a new risk-score, denominated *AITL score*, contemplating the independent variables age ≥ 60 years, ECOG performance status ≥ 2, high C-reactive protein levels, and β2-microglobulin ≥ normal value, as having a high ability to discriminate clinical outcomes in cases of AITL (28). According to this risk-score, patients categorized as low-, intermediate-, and high-risk had 5-year OS estimates of 63%, 54%, and 21%, respectively (28). Similarly, Hong et al., retrospectively analyzing 115 patients with AITL, identified as independent predictors associated with poor survival the histological involvement of bone marrow, involvement of > 1 extranodal site by NHL and performance status > 1. In this prognostic score, patients were categorized into three groups, with 5-year OS estimates of 86.9%, 46.3% and 16.2%, respectively, $p<0.0001$. According to the authors, this score showed better predictive discrimination to determine survival for AITL patients when compared to the IPI and PTCL-U scores (103).

Laboratory and molecular-genetic biomarkers have emerged in recent decades as methods for predicting prognosis in several hematological malignancies, including AITL. Therefore, high peripheral monocyte count at diagnosis, thrombocytopenia, high-levels of immunoglobulin A (IgA), serum albumin < 35 g/L, high-levels of β2-microglobulin, reduced lymphocyte/monocyte ratio, presence of *TET-2* mutations in diagnostic biopsies, the *RhoA* G17V mutation, high tumor mutational burden (TMB) involving a gene panel composed by the *RhoA* GTPase and epigenetic regulators genes (*IDH-2*, *TET-2* and *DNMT3A*), gene signature pattern related to monocytes and *TP53*, as well as tumor status for EBV assessed by EBER-ISH in lymph nodes (EBER-negative status) adversely impacted the survival of AITL patients according to data published by different research groups in the last ten years (51, 52, 104–109).

7 Treatment

Up-front therapy for AITL and other nMTCL-TFH-phenotype may be focused on different objectives, from curative proposals to strategies with a merely palliative aim. Young patients aged less than 60-65 years, with good performance status (ECOG ≤ 2) and without significant comorbidities will experience multidrug therapy based on anthracyclic agents with curative intent, preferably followed by consolidation in first complete remission/partial response (CR/PR) with high-dose therapy (HDT) and autologous hematopoietic stem cell transplantation (ASCT). Although it is considered the gold-standard therapy outside the clinical trial scenario, this approach is still not able to promote high-rates of long-term sustained remission, being associated with an estimated 5-year OS around 30-40% (3, 10, 28, 110). On the other hand, elderly patients (≥ 60-65 years old) with poor clinical conditions and/or severe comorbidities ("frail patients") will be treated with palliative intent, due to the high morbidity and mortality rates presented by this population when submitted to intensified therapeutic strategies. Therefore, this subgroup of patients may be approached with a combination of corticosteroids and low-dose chemotherapy, as well as the isolated use of steroids, calcineurin inhibitors (cyclosporine A), immunomodulators (lenalidomide), use of the immunoconjugate drug-monoclonal antibody anti-CD30 (brentuximab-vedotin/BV), gemcitabine-based monochemotherapy, or strategies focused on epigenetic regulation, such as the association of hypomethylating agents (HMAs) and histone deacetylase inhibitors (HDAi) (110, 111). At the same time, AITL elderly patients (≥ 60 years) without serious comorbidities and presenting good performance status, categorized as an "intermediate-age" population (between 60 – 75 to 80 years), may benefit from a full course of cytotoxic chemotherapy (e.g. CHOP or BV-CHP regimens), even if ineligible for consolidation with ASCT.

As observed in other nMTCL, patients with AITL exhibit high-rates of resistance to anthracyclic-based chemotherapy, primarily centered on the CHOP regimen (cyclophosphamide, doxorubicin, vincristine, and prednisone) (3, 10, 29, 110, 111). Currently, it is known that this is due to the high-concentration of P-glycoprotein (Pgp) present in nMTCL tumor cells, which confers a multidrug resistance (MDR) phenotype. By generating an efflux pump mechanism for chemotherapeutic agents, notably against anthracyclines and vinca alkaloids, nMTCL tumor cells block the cytoplasmic internalization of these drugs, thus preventing their

antineoplastic effect (112–114). Although CHOP-like regimens are still considered the "standard of care" for AITL patients treated with curative intent, the responses obtained with this strategy are considered poor, with overall response rates (ORR) around 70-79% and complete response (CR) around 35-39% (10, 115, 116). Similarly, long-term outcomes have been highly unsatisfactory for AITL patients treated with anthracycline-based regimens as reported by different groups, with 5-year PFS estimates of 18%, 13%, and 20% according to the *ITCLP*, *British Columbia Cancer Agency* (BCCA) group, and *Swedish National Lymphoma Registry*, respectively (10, 115, 116). This has led to several guidelines, including that of the *National Comprehensive Cancer Network* (NCCN), to recommend enlisting patients with AITL in clinical trials as preferred primary therapeutic strategy (117).

Due to the rarity of AITL, which represents only 2% of all NHL, the majority of studies that aim to analyze the effectiveness of different therapeutic strategies include AITL patients together with others subtypes of nMTCL, such as PTCL, NOS and ALCL, which makes it difficult to interpret the results. Therefore, most of the studies presented in the next paragraphs do not exclusively involve patients with AITL, although those with greater scientific robustness present results for patients with AITL in the form of subgroup analysis.

Aiming to improve the outcomes provided by the CHOP regimen in patients with nMTCL, several strategies have been conducted over the last two decades. Among the main ones, we highlight the addition of other drugs to the CHOP protocol, such as etoposide (CHOEP regimen), the anti-CD52 monoclonal antibody alemtuzumab (CHOP plus alemtuzumab), the proteasome inhibitor bortezomib (V-CAP regimen), association with epigenetic modulators (CHOP plus 5-azacitidin or romidepsin), in addition to more intensified multi-drug regimens such as ICE/ABVD (ifosfamide, carboplatin, etoposide/adriamycin, bleomycin, vinblastine and dacarbazine), PEGS (cisplatin, etoposide, gemcitabine and solumedrol), ACVBP (doxorubicin, cyclophosphamide, vindesine, bleomycin and prednisone), among many others. Overall, although some of these combinations promoted higher overall response rates, toxicity was notably increased and there was no unequivocal benefit in overall survival (29, 116, 118–124).

The first strategy used to intensify the CHOP regimen in the scenario of nMTCL was based on the addition of etoposide at a dose of 100 mg/sqm I.V. on the first three days of the regimen with dosing intervals every 14 or 21 days (CHOEP-14/21). Although a study conducted by the *German High-Grade NHL Study Group* involving 289 patients with nMTCL demonstrated a 3-year event-free survival (EFS) benefit for patients with ALK1+ ALCL, specifically for those younger than 60 years and with normal levels of LDH (p=0.003), this benefit was only marginal (p=0.057) for non-ALK1+ subtypes, including AITL patients. In this same cohort, elderly patients (aged > 60 years) experienced prohibitive toxicity with the addition of etoposide to the CHOP regimen. Furthermore, even in the group of younger patients, CHOP plus etoposide did not demonstrate an unequivocal benefit in terms of overall survival for any of the nMTCL histological subtypes (118). Similar data from the *Swedish National Lymphoma Registry*, with 252 cases of nMTCL treated over a period of more than 10 years showed that the addition of etoposide to CHOP caused higher ORR and increased PFS in patients younger than 60 years (HR 0.49, p=0.008), without, however, leading to greater overall survival (116).

In opposition to previous studies, a recent publication involving 1427 patients enrolled in the *Netherlands Cancer Registry* between 1989 and 2018 demonstrated for the first time a 5-year OS advantage for the CHOEP versus CHOP regimen (64% and 44%, respectively, p<0.01). However, after adjustment for histological subtype, this benefit was maintained only for patients with ALK1+ ALCL, with a 6.3-fold increased risk of mortality among those treated with CHOP. In this analysis, patients < 65 years with ALK1-negative ALCL, AITL, and PTCL, NOS subtypes did not have increased OS with addition of etoposide to CHOP (125). In 2021, Kim J et al., conducted a meta-analysis involving 34 cohorts from 28 studies and a pool of 1424 patients with nMTCL. This meta-analysis demonstrated OS benefit for the CHOEP regimen versus the CHOP regimen (126). However, in contrast, another recent meta-analysis involving 1560 patients with nMTCL from five retrospective and prospective studies was unable to demonstrate differences in CR, PR, and survival between CHOP and CHOEP regimens. As expected, in this meta-analysis patients with nMTCL treated with CHOEP-21 had higher hematologic toxicity, including higher rates of anemia and thrombocytopenia (127).

In a recent retrospective analysis published by our group, involving 124-Brazilian patients with nMTCL, treated under real-life conditions between 2000 and 2019, we demonstrated similar ORR between cases treated with CHOP-21 and CHOEP-21 (76.6% and 65.9%, respectively, p=0.259). Although CHOEP-21 was associated with a lower rate of primary refractoriness (4.5% vs 21.2%, p=0.018), the combination of CHOP plus etoposide was correlated with higher rates of delay between chemotherapy cycles (p=0.0004), definitive interruption of treatment (p=0.003) and toxicities, including severe neutropenia (p=0.001), febrile neutropenia (p=0.003) and severe thrombocytopenia (p=0.0007). Therefore, we assume that the high toxicities of the CHOEP-21 regimen, when applied to a population of particularly frail patients with nMCTL, may explain the absence of OS and PFS benefit, as demonstrated in our study (29). In summary, even today, the addition of etoposide to the CHOP regimen presents conflicting results regarding its potential benefit in the scenario of nMTCL, particularly in non-ALK1+ ALCL cases, such as AITL and correlated neoplasms with TFH-phenotype. Specifically for the population with AITL, no study was able to demonstrate an OS advantage for the CHOEP regimen compared to CHOP.

Based on the biological rationale associated to VEGF-1 overexpression, with consequent increase in tumor vascular density, as well as the immune dysfunction commonly observed in AITL, the use of agents with anti-angiogenic and immunomodulatory properties have been tested as potential new therapeutic weapons in this lymphoma. In this regard, the effects of adding the humanized anti-VEGF1 monoclonal antibody bevacizumab to the CHOP regimen followed by maintenance with monodrug bevacizumab was tested in a phase II study involving 46 patients with nMTCL. Despite promoting high ORR,

the CHOP plus bevacizumab regimen failed to provide durable remissions and was associated with high-rates of toxicity, particularly myelotoxicity, and an increased risk of serious cardiovascular events (128). Following the same rationale, addition of the immunomodulatory and anti-angiogenic agent lenalidomide to the CHOP-21 regimen was recently tested in a phase II multicenter study including 80 treatment-naïve elderly AITL patients. In this study, the primary endpoint (complete metabolic response [CMR] at the end of treatment) was reached in only 41% of cases, being below the CMR rate of 55%, pre-specified as the adopted success rate. The 2-year PFS and OS were 42.1% and 59.2%, respectively, with non-negligible hematological toxicity, which resulted in discontinuation of therapy in 15% of cases. Interestingly, in this prospective study, with broad molecular characterization of the included cases, mutations in the *DNMT3A* gene were associated with shortened PFS and *IDH-2* mutations were associated with specific pathological findings, including FDC expansion, presence of clear neoplastic cells and bone marrow infiltration (129).

Due to the frequent interactions between TFH-tumor cells and CD20+ B-cells that make up the AITL tumor microenvironment, together with the fact that large CD20+ B-immunoblasts containing EBV are expanding in the paracortex of AITL lymph nodes, there is a rationale for the use of therapies that deplete B-lymphocytes, such as the anti-CD20 monoclonal antibody rituximab in this tumor. Based on this premise, Delfau-Larue et al. conducted a phase II study using 8 cycles of R-CHOP-21 in 25 patients newly diagnosed with AITL. With a CR rate of only 44% and an estimated 2-year PFS of 42%, this study did not demonstrate a clear benefit of adding rituximab to conventional anthracycline-based chemotherapy in patients with AITL (106). A recent publication, involving 335 AITL patients registered in the *Netherlands Cancer Registry* between 2014 and 2020, confirmed the findings previously cited by the French group. In the Dutch study, although addition of rituximab to CHOP-like regimens (CHOP-21 or CHOEP-21) improved ORR, there was no increase in OS or PFS with adoption of this strategy (130).

Expression of the CD30 antigen has been described in 43-90% of AITL cases, which opened a precedent for the use of the anti-CD30 drug-antibody immunoconjugate brentuximab vedotin (BV) in its primary treatment. In the phase III, multicenter, prospective Echelon-2 study, 452 patients with CD30+ nMTCL were up-front treated with BV-CHP regimen (Brentuximab-vedotin, cyclophosphamide, doxorubicin, and prednisone) versus CHOP for 6 to 8 cycles with a 21-day interval between administrations. We highlight the fact that more than 70% of the patients included in the study had ALCL, a universally CD30+ neoplasm associated with a high density of antigen expression. On the other hand, only 13% of this cohort involved patients with AITL. The BV-CHP group had substantially higher rates of complete response ($p<0.01$), as well as unequivocal benefit of OS and PFS, with a median PFS of 48 months versus 20 months for BV-CHP and CHOP, respectively, $p=0.01$. Furthermore, the rate of serious adverse events, including myelotoxicity and peripheral neuropathy, did not differ statistically significantly between both treatment arms. The data presented by Echelon-2 represented a paradigm shift in the primary therapy of CD30+ nMTCL, particularly for ALCL, where the BV-CHP regimen is currently considered the gold standard of care. However, we highlight the fact that in subgroup analysis, in opposition to what was evidenced for CD30+ PTCL, NOS and ALCL (ALK1+/ALK1-), patients with CD30+ AITL treated with the BV-CHP regimen had decreased PFS than those treated with CHOP (HR: 1.40, 95% CI 0.64-3.07) (131).

The discovery of recurrent mutations involving epigenetic regulatory genes in patients with AITL and in other nMTCL-TFH-phenotype opened the opportunity for the incorporation of epigenetic modifying drugs in the therapeutic arsenal of these lymphomas (132). Therefore, HMAs, particularly 5-azacytidine, and several HDAi, such as romidepsin, belinostat and vorinostat have been used in monotherapy or in combination for AITL therapy. Recent studies have shown promising results and relative safety for these drugs, both when used in the first-line setting or in R/R disease (120, 133–136).

In 2018, Lemmonier et al., published a report demonstrating the effectiveness of 5-azacytidine in inducing sustained responses in AITL. In this study, the authors used 5-azacytidine in monotherapy, applied subcutaneously in a retrospective series of 12 AITL patients treated with HMAs for concomitant myeloid neoplasia or in the setting of R/R disease. The ORR was 75%, with 50% CR and 25% PR. With a median follow-up of 27 months, the median OS and PFS were 21 months and 15 months, respectively. Due to sample limitations, the authors could not establish the impact of the mutational status of the *RhoA*, *TET-2*, *DNMT3A* and *IDH-2* genes in therapeutic response and survival (133). Subsequently, O'Connor et al. conducted a phase I study to assess the efficacy and safety of combining oral azacytidine with intravenous romidepsin in patients with advanced R/R lymphoid malignancies, with an emphasis on MTCL. This study determined the maximum tolerated dose of this association (300 mg of azacytidine P.O. on days 1 to 14 and romidepsin 14 mg/sqm I.V. on days 1, 8, 15 and 22, with an interval of 35 days between cycles) and revealed the efficacy and safety of the combination. The overall response and complete response rate were 73% and 55% in patients with MTCL, however the authors did not find association between response and level of demethylation or tumor mutational profile (135). In 2021, the same group published data from a phase II multicenter study testing the same association in 25 cases of R/R MTCL. In that study, ORR and CR were 61% and 48%, respectively. Interestingly, cases with TFH-phenotype were particularly susceptible to the combination of epigenetic modifiers, with ORR and CR of 80% and 67%, respectively. This regimen was shown to be safe and capable of inducing long-lasting responses in patients with R/R disease, with a median response duration of 20.3 months and median OS not reached. In a pioneering way, the authors were able to establish an association between response and greater mutational burden involving epigenetic regulatory genes (134).

Although the use of epigenetic modifiers has shown promising results in R/R disease, their use in the first-line setting, particularly for fit patients, is premature and has led to controversial results. In this sense, studies using the up-front CHOP regimen in association with epigenetic modifiers have had their results recently published. In 2020, Jia Ruan et al. reported results from a phase II study

involving 21 previously untreated MTCL patients receiving 6 cycles of CHOP plus oral azacytidine. More than 80% of the sample had the TFH-phenotype. At the end of therapy, ORR was 76.5% for all patients and 86.7% for cases with TFH-phenotype. Estimates of PFS and OS at 1 year were 61.1% and 88.9%, respectively, for nMTCL-TFH. *TET-2* mutations were associated with higher rates of CR (p=0.014), PFS (p=0.012) and OS (p=0.042). In contrast, *DNMT3A* mutations were associated with decreased OS (p=0.028) (120). On the other hand, in 2015, French researchers published results of a phase Ib/II study testing the combination CHOP plus romidepsin (Ro-CHOP) in 37 previously untreated patients with nMTCL. The regimen was initially shown to be effective and safe, allowing for escalation to a phase III trial (136). However, the results of the phase III study, involving 421 patients with newly diagnosed MTCL (Ro-CHOP = 211, CHOP = 210) have just been released. In addition to being associated with higher rates of grade 3/4 hematologic toxicity, the addition of romidepsin to CHOP did not increase response rates, PFS, and OS. Therefore, the authors concluded that, although based on a biological rationale, the Ro-CHOP association did not represent a significant advance in the standard of care for primary therapy of MTCL (121). Table 1 summarizes the results of different studies comparing CHOP regimen and other approaches for treatment of AITL in different clinical settings.

The role of consolidation therapy with autologous hematopoietic stem cell transplantation (ASCT) in first remission has been evaluated in different retrospective and prospective studies. The two largest retrospective studies were conducted by the Swedish (*Swedish Lymphoma Registry*) and French (*LYSA study*) groups (116, 137). Both studies excluded patients with ALK1+ ALCL, and evaluated 252 and 269 patients with nodal MTCL, respectively. While the French study did not demonstrate a significant difference in OS and PFS between transplanted and non-transplanted cases, the Swedish study demonstrated that patients undergoing upfront ASCT had superior PFS (HR: 0.56, p=0.002) and increased OS (HR: 0.58, p=0.004) (116, 137). Another large retrospective study was conducted by Abramson JS et al. (2014), aiming to evaluate the optimal frontline therapy for PTCL, as well as assess the impact of upfront ASCT in these malignancies. This trial included 341 newly diagnosed PTCL patients from 2000 to 2011, and 23% of them had a diagnosis of AITL. Most patients (70%) received CHOP-like therapy and only 10% experienced upfront ASCT. Although ORR was 73%, 24% of cases were chemo-refractory and the 3-year PFS estimate was only 32%, significantly lower than DLBCL-matched patients. In this study, early-stage disease and response to initial therapy were the main predictors of favorable outcomes. However, no difference in OS was observed based on choice of primary chemotherapy regimen or consolidation with ASCT in first remission (138).Among prospective studies, the first to assess the role of upfront consolidation with ASCT in PTCL was conducted by Reimer et al. (2008) (139). Eighty-three patients with PTCL were included in this trial, with 32.5% (n=27) having AITL diagnosis, but only 55 (66%) received upfront ASCT consolidation. The main reason for not receiving the transplant was disease progression. ORR was 66% (56% CR and 8% PR). With a median follow-up of 33 months, the estimated 3-year OS, DFS and PFS were 48%, 53%, and 36%, respectively. The results obtained by

TABLE 1 Main studies evaluating alternative therapeutic strategies to the CHOP regimen for MCTL in different clinical settings.

Author	Study design	Population	Therapeutic strategies	Outcomes
Schmitz N, 2010	Retrospective	289 nMTCL (28 AITL)	CHOP vs CHOEP	3y EFS: 50.0 vs 67.5% (for AITL)
Ellin F, 2014	Retrospective	755 nMTCL (104 AITL)	CHOP vs CHOEP	5y PFS: 23.0 vs 40.0% (for AITL)
Lage L, 2022	Retrospective	124 nMTCL (13 AITL)	CHOP vs CHOEP	2y PFS: 69.7 vs 25.0% (for all nMTCL)
Brink M, 2022	Retrospective	1427 nMTCL (294 AITL)	CHOP vs CHOEP	5y OS: 44.0 vs 64.0% (for all nMTCL)
Gallamini A, 2007	Prospective, Phase II	24 MTCL (6 AITL)	CHOP plus alemtuzumab	2y FFS: 48%
Ganjoo K, 2014	Prospective, Phase II	46 MTCL (17 AITL)	CHOP plus bevacizumab	1y PFS: 44% (57% for AITL)
Lemonnier F, 2021	Prospective, Phase II	80 nMTCL (67 AITL)	CHOP plus lenalidomide	2y PFS: 42% for AITL
Meewes FO, 2022	Retrospective	335 AITL	R-CHO(E)P vs R-CHOP	2y PFS: 45.0 vs 40.0%
Horwitz S, 2019	Prospective, Phase III	452 MTCL (> 70% ALCL)	BV-CHP vs CHOP	Median PFS: 48.2 vs 20.8 months
Falchi L, 2021	Prospective, Phase II	25 R/R nMTCL (20 nMTCL-TFH)	5-azacytidine plus romidepsin	Median PFS: 8.0 months
Ruan J, 2020	Prospective, Phase II	21 nMTCL (16 nMTCL-TFH)	5-azacytidine plus CHOP	1y PFS: 56.8% (61.1% for AITL)
Bachy E, 2022	Prospective, Phase III	421 MTCL	CHOP vs Romidepsin plus CHOP (Ro-CHOP)	Median PFS: 10.2 vs 12.0 months

nMTCL, nodal mature T-cell lymphoma; AITL, angioimmunoblastic T-cell lymphoma; EFS, event-free survival; PFS, progression-free survival; OS, overall survival; FFS, failure-free survival; ALCL, anaplastic large-cell lymphoma; nMTCL-TFH, nodal mature T-cell lymphoma with follicular T-helper phenotype; CHOP, cyclophosphamide, doxorubicin, vincristine and prednisone; R, rituximab; BV, brentuximab-vedotin; (E), etoposide; Ro, romidepsin.

the German group in this prospective and multicenter trial suggested, for the first time, a substantial impact on outcomes for upfront ASCT in PTCL (139).

The largest prospective study addressing the role of upfront ASCT in PTCL was the *NLG-T-01* trial, conducted by the Nordic group (140). In this phase 2, prospective and multicenter study, 160 patients with non-ALK1+ nodal MTCL were treated with 6 cycles of CHOEP-14. Patients on first CR/PR were consolidated with BEAM (carmustine, etoposide, cytarabine, and melphalan) followed by ASCT. A total of 71% of cases (115/160) underwent ASCT, with estimated 5-year PFS and OS of 44% and 51%, respectively. A significant benefit was particularly seen in the ALK1-negative ALCL group, with 5-year OS and PFS estimates of 70% and 61%, respectively. AITL patients had 5-year OS and PFS estimates of 51% and 49%, respectively (140).

Two other recent large prospective studies evaluated the impact of upfront ASCT in patients with PTCL including AITL cases. In the study conducted by investigators from the COMPLETE group (*Comprehensive Oncology Measures for Peripheral T-Cell Lymphoma Treatment*) 119 patients with non-ALK1-positive nodal PTCL were included (36 experienced ASCT at first CR and 83 did not receive ASCT). With a median follow-up of 2.8 years, the median OS was not reached in patients who underwent ASCT and was 57.6 months in those who did not receive the transplant (p=0.06). ASCT has been associated with increased survival in patients with advanced-stage disease and in those with high-risk IPI, with particular benefit of OS and PFS for AITL cases (141). In the study of the Korean group, 191 patients with PTCL were prospectively enrolled, with 31.4% (n=60) diagnosed with AITL. Among transplant-eligible patients (n=59), 54.2% (n=32) had experienced this therapeutic modality, but there were no significant differences in OS and PFS between the transplanted and non-transplanted arms. However, in patients with AITL, the ASCT promoted PFS benefit, without offering significant differences in OS. Although up-front consolidation with ASCT still offers contradictory results in the therapeutic management of PTCL in the modern era, data from both studies suggest that ASCT may provide a real survival benefit in AITL, particularly when applied in selected cases (142).

Few studies have evaluated the role of upfront allogeneic stem cell transplantation (alloSCT) in nodal MTCL. In this sense, the *AATT* study was the only phase 3 trial to assess the role of upfront stem cell transplantation in patients with nodal MTCL, excluding cases of ALK1+ ALCL. This study compared outcomes of patients undergoing first-line ASCT and alloSCT. The study was prematurely discontinued because the interim analysis revealed a high-rate of transplant-related mortality (TRM) in the alloSCT arm. After a follow-up of 42 months, the 3-year EFS was 43% and 38% for alloSCT and ASCT, respectively. The 3-year OS was 57% and 70% for alloSCT and ASCT, respectively. Although no patient who underwent alloSCT relapsed versus 36% (13/36) in the ASCT group, the TRM was 31% (8/26) in the alloSCT group versus 0% in the ASCT group (0/41) (143).

In the context of R/R disease, the therapeutic strategy for AITL patients should be determined in view of the objective of the treatment, whether with curative or palliative intent. Patients with R/R disease treated with curative intuit are usually rescued with polychemotherapeutic regimens based on platinum derivatives, such as DHAP (dexamethasone, cytarabine, cisplatin), DHAOX (dexamethasone, cytarabine, oxaliplatin), ICE (ifosfamide, carboplatin, etoposide), ESHAP (etoposide, methylprednisolone, cytarabine and cisplatin), GDP (gemcitabine, dexamethasone and cisplatin) or GEMOX (gemcitabine, oxaliplatin) as bridges to ASCT (if not previously submitted to this therapeutic strategy) or alloSCT (if previously experienced ASCT). In this context, in general, the 5-year OS estimate with ASCT is around 45-53%, with TRM oscillating between 0-10% (144–147). For the alloSCT, the 5-year OS ranges from 50-81%, with 5-year PFS ranging from 40-64%, and TRM between 12-33%, according to different studies (146, 148–150). Patients with R/R disease who will be treated with non-curative intent should preferably experience regimens with agents that do not cause cumulative toxicity, in low-doses and "non-finite". For this purpose, several options may be considered, including monotherapy with romidepsin, belinostat, 5-azacytidine, pralatrexate, bendamustine, brentuximab-vedotin, lenalidomide and cyclosporine A, with ORR ranging from 8-75%, and a short median duration of response (3.5-17 months) (110). However, we must highlight that some of these compounds are not approved by many regulatory agencies for AITL management; therefore, the availability of these products might vary in different countries. Among combination regimens tested in phase 1/2 studies, the most promising appear to be romidepsin/pralatrexate (ORR 71%; CR 29%), oral 5-azacitidinb/romidepsin (ORR 71%; CR 71%) and gemcitabine/copanlisib (ORR 72%; CR 32%) (135, 151, 152).

Although chimeric antigen T-cell receptor (CAR-T) therapy has proved to be an interesting form of immunotherapy in several B-cell lymphoid malignancies, such as R/R diffuse large B-cell lymphoma, follicular lymphoma, B-cell acute lymphoblastic leukemia and multiple myeloma, its use has been extremely limited in the setting of PTCL. While encouraging results were seen with some novel agents, there is still very few data about CAR-T therapy in PTCL; limited experience and lack of controls preclude critical analyses. One of the main challenges in the use of CAR-T therapy in T-cell malignancies is due to the fact that neoplastic cells share a series of common antigens with normal T-lymphocytes, which can lead to fratricide and serious T-cell lymphoid aplasia in the receptor. To mitigate this effect, a selection of appropriate antigenic targets is essential. Currently, specific antigens have been selected for the construction of chimeric products, among which CD30, CD37, TRBC1, CCR4 and CCR9 stand out. Use of nanobody-derived or naturally selected CAR-T are attractive strategies to overcome fratricide. Another problem intrinsic to the use of this therapeutic modality in T-cell malignancies refers to the potential contamination of the product collected for the construction of CAR-T with clonal T-cells, however, the use of allogeneic CAR-T products or CAR-NK-cells are possible strategies with ability to mitigate this contamination. Currently, data about the use of CAR-T therapy in PTCL, particularly for AITL are very scarce, although it may constitute an interesting therapeutic option for R/R disease in the next future (153). Figure 5 summarizes the proposal for a

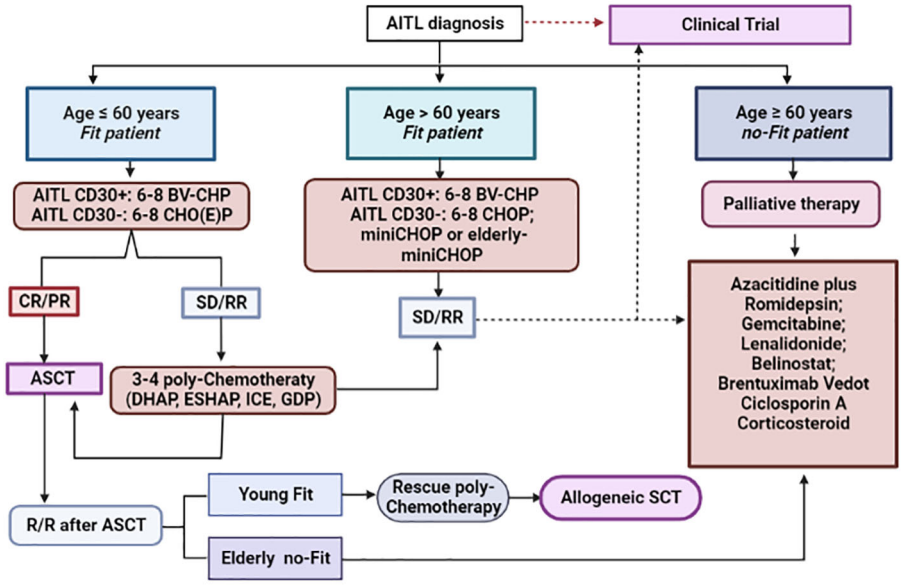


FIGURE 5
Therapeutic algorithm for the management of AITL patients. Up-front therapy and treatment of relapsed/refractory cases. *AITL, angioimmunoblastic T-cell lymphoma; BV-CHP, brentuximab-vedotin plus cyclophosphamide, doxorubicin and prednisone; CHO(E)P, cyclophosphamide, doxorubicin, vincristine, etoposide and prednisone; CR, complete response; PR, partial response; SD, stable disease; R/R, relapsed/refractory disease; ASCT, autologous stem cell transplantation; DHAP, dexamethasone, high-doses of cytarabine, cisplatin; ESHAP, etoposide, methylprednisolone, high-doses of cytarabine, cisplatin; ICE, ifosfamide, carboplatin, etoposide; GDP, gemcitabine, dexamethasone and cisplatin.

therapeutic algorithm to approach patients with AITL in first line and in the context of R/R disease.

8 Conclusion

AITL is a peculiar subtype of nodal MTCL derived from monoclonal proliferation of TFH-cells and is associated with a poor overall prognosis. Recent studies, involving modern technologies, such as GEP and NGS, have contributed to determine its cell of origin, define its unique gene-signature, and revealed a specific-pattern of recurrent mutations, which have contributed in a decisive way for its biological understanding. Studies conducted in xenograft models have elucidated the multi-step neoplastic transformation process of AITL, from acquisition of premalignant epigenetic mutations to the development of disease-specific mutations ("driver-mutations"), such as *RhoA* G17V. However, such biological progress has not yet been translated objectively into the therapeutic field. Currently, the treatment of patients with AITL is still based on the wide use of anthracyclic agents, with dismal outcomes, being an unmet medical need. Hopefully, new agents, with employment based on biological rationale, such as HMAs and HDAi may represent a paradigm shift in the approach to this lymphoma in the near future. For this, collaborative multicenter studies involving large cohorts from different parts of the globe are necessary.

Author contributions

LL, HC, CR, SdS, and JP reviewed the literature, organized, and wrote the article. All authors contributed to the article and approved the submitted version.

Acknowledgments

We thank Fabio Alessandro de Freitas and Samuel Campanelli Couto Freitas for layout of the article and English revision, respectively.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Lage LA, Cabral TC, Costa Rde O, Gonçalves Mde C, Levy D, Zerbini MC, et al. Primary nodal peripheral T-cell lymphomas: diagnosis and therapeutic considerations. *Rev Bras hematologia e hemoterapia* (2015) 37(4):277–84. doi: 10.1016/j.bjhh.2015.03.017

2. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, et al. The 2016 revision of the world health organization classification of lymphoid neoplasms. *Blood* (2016) 127(20):2375–90. doi: 10.1182/blood-2016-01-643569

3. Lunning MA, Vose JM. Angioimmunoblastic T-cell lymphoma: the many-faced lymphoma. *Blood* (2017) 129(9):1095–102. doi: 10.1182/blood-2016-09-692541

4. Chiba S, Sakata-Yanagimoto M. Advances in understanding of angioimmunoblastic T-cell lymphoma. *Leukemia* (2020) 34(10):2592–606. doi: 10.1038/s41375-020-0990-y

5. Vinuesa CG, Tangye SG, Moser B, Mackay CR. Follicular b helper T cells in antibody responses and autoimmunity. *Nat Rev Immunol* (2005) 5(11):853–65. doi: 10.1038/nri1714

6. de Leval L, Rickman DS, Thielen C, Reynies A, Huang YL, Delsol G, et al. The gene expression profile of nodal peripheral T-cell lymphoma demonstrates a molecular link between angioimmunoblastic T-cell lymphoma (Aitl) and follicular helper T (Tfh) cells. *Blood* (2007) 109(11):4952–63. doi: 10.1182/blood-2006-10-055145

7. Armitage JO, Vose JM, Weisenburger DD. Towards understanding the peripheral T-cell lymphomas. *Ann Oncol Off J Eur Soc Med Oncol* (2004) 15(10):1447–9. doi: 10.1093/annonc/mdh409

8. Rodríguez J, Gutiérrez A, Martínez-Delgado B, Perez-Manga G. Current and future aggressive peripheral T-cell lymphoma treatment paradigms, biological features and therapeutic molecular targets. *Crit Rev oncology/hematology* (2009) 71(3):181–98. doi: 10.1016/j.critrevonc.2008.10.011

9. Campo E, Swerdlow SH, Harris NL, Pileri S, Stein H, Jaffe ES. The 2008 who classification of lymphoid neoplasms and beyond: evolving concepts and practical applications. *Blood* (2011) 117(19):5019–32. doi: 10.1182/blood-2011-01-293050

10. Vose J, Armitage J, Weisenburger D. International peripheral T-cell and natural Killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol Off J Am Soc Clin Oncol* (2008) 26(25):4124–30. doi: 10.1200/jco.2008.16.4558

11. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

12. Huang Y, Moreau A, Dupuis J, Streubel B, Petit B, Le Gouill S, et al. Peripheral T-cell lymphomas with a follicular growth pattern are derived from follicular helper T cells (Tfh) and may show overlapping features with angioimmunoblastic T-cell lymphomas. *Am J Surg Pathol* (2009) 33(5):682–90. doi: 10.1097/PAS.0b013e3181971591

13. Dobay MP, Lemonnier F, Missiaglia E, Bastard C, Vallois D, Jais JP, et al. Integrative clinicopathological and molecular analyses of angioimmunoblastic T-cell lymphoma and other nodal lymphomas of follicular helper T-cell origin. *Haematologica* (2017) 102(4):e148–e51. doi: 10.3324/haematol.2016.158428

14. Frizzera G, Moran EM, Rappaport H. Angio-immunoblastic lymphadenopathy with dysproteinaemia. *Lancet (London England)* (1974) 1(7866):1070–3. doi: 10.1016/s0140-6736(74)90553-4

15. Lukes RJ, Tindle BH. Immunoblastic lymphadenopathy. a hyperimmune entity resembling hodgkin's disease. *New Engl J Med* (1975) 292(1):1–8. doi: 10.1056/nejm197501022920101

16. Lennert K. [Nature, prognosis and nomenclature of angioimmunoblastic (Lymphadenopathy (Lymphogranulomatosis X or T-zone lymphoma)]. *Deutsche medizinische Wochenschrift (1946)* (1979) 104(35):1246–7.

17. Kaneko Y, Maseki N, Sakurai M, Takayama S, Nanba K, Kikuchi M, et al. Characteristic karyotypic pattern in T-cell lymphoproliferative disorders with reactive "Angioimmunoblastic lymphadenopathy with dysproteinemia-type" features. *Blood* (1988) 72(2):413–21.

18. Tobinai K, Minato K, Ohtsu T, Mukai K, Kagami Y, Miwa M, et al. Clinicopathologic, immunophenotypic, and immunogenotypic analyses of immunoblastic lymphadenopathy-like T-cell lymphoma. *Blood* (1988) 72(3):1000–6.

19. Harris NL. A practical approach to the pathology of lymphoid neoplasms: a revised European-American classification from the international lymphoma study group. *Important Adv Oncol* (1995) 111–40.

20. Attygalle A, Al-Jehani R, Diss TC, Munson P, Liu H, Du MQ, et al. Neoplastic T cells in angioimmunoblastic T-cell lymphoma express Cd10. *Blood* (2002) 99(2):627–33. doi: 10.1182/blood.v99.2.627

21. Quivoron C, Couronné L, Della Valle V, Lopez CK, Plo I, Wagner-Ballon O, et al. Tet2 inactivation results in pleiotropic hematopoietic abnormalities in mouse and is a recurrent event during human lymphomagenesis. *Cancer Cell* (2011) 20(1):25–38. doi: 10.1016/j.ccr.2011.06.003

22. Couronné L, Bastard C, Bernard OA. Tet2 and Dnmt3a mutations in human T-cell lymphoma. *New Engl J Med* (2012) 366(1):95–6. doi: 10.1056/NEJMc1111708

23. Cairns RA, Iqbal J, Lemonnier F, Kucuk C, de Leval L, Jais JP, et al. Idh2 mutations are frequent in angioimmunoblastic T-cell lymphoma. *Blood* (2012) 119(8):1901–3. doi: 10.1182/blood-2011-11-391748

24. Sakata-Yanagimoto M, Enami T, Yoshida K, Shiraishi Y, Ishii R, Miyake Y, et al. Somatic rhoa mutation in angioimmunoblastic T cell lymphoma. *Nat Genet* (2014) 46(2):171–5. doi: 10.1038/ng.2872

25. Palomero T, Couronné L, Khiabanian H, Kim MY, Ambesi-Impiombato A, Perez-Garcia A, et al. Recurrent mutations in epigenetic regulators, rhoa and fyn kinase in peripheral T cell lymphomas. *Nat Genet* (2014) 46(2):166–70. doi: 10.1038/ng.2873

26. Morton LM, Wang SS, Devesa SS, Hartge P, Weisenburger DD, Linet MS. Lymphoma incidence patterns by who subtype in the united states, 1992-2001. *Blood* (2006) 107(1):265–76. doi: 10.1182/blood-2005-06-2508

27. de Leval L, Parrens M, Le Bras F, Jais JP, Fataccioli V, Martin A, et al. Angioimmunoblastic T-cell lymphoma is the most common T-cell lymphoma in two distinct French information data sets. *Haematologica* (2015) 100(9):e361–4. doi: 10.3324/haematol.2015.126300

28. Advani RH, Skrypets T, Civallero M, Spinner MA, Manni M, Kim WS, et al. Outcomes and prognostic factors in angioimmunoblastic T-cell lymphoma: final report from the international T-cell project. *Blood* (2021) 138(3):213–20. doi: 10.1182/blood.2020010387%JBlood

29. Lage L, Brito CV, Barreto GC, Culler HF, Reichert CO, Levy D, et al. Up-front therapy with chop plus etoposide in Brazilian nodal ptcl patients: increased toxicity and no survival benefit compared to chop regimen-results of a real-life study from a middle-income country. *Clin Lymphoma Myeloma Leuk* (2022) 22(11):812–24. doi: 10.1016/j.clml.2022.06.012

30. Federico M, Rudiger T, Bellei M, Nathwani BN, Luminari S, Coiffier B, et al. Clinicopathologic characteristics of angioimmunoblastic T-cell lymphoma: analysis of the international peripheral T-cell lymphoma project. *J Clin Oncol Off J Am Soc Clin Oncol* (2013) 31(2):240–6. doi: 10.1200/jco.2011.37.3647

31. de Leval L, Gisselbrecht C, Gaulard P. Advances in the understanding and management of angioimmunoblastic T-cell lymphoma. *Br J haematology* (2010) 148(5):673–89. doi: 10.1111/j.1365-2141.2009.08003.x

32. Cortés JR, Palomero T. The curious origins of angioimmunoblastic T-cell lymphoma. *Curr Opin Hematol* (2016) 23(4):434–43. doi: 10.1097/moh.0000000000000261

33. Schaerli P, Willimann K, Lang AB, Lipp M, Loetscher P, Moser B. Cxc chemokine receptor 5 expression defines follicular homing T cells with b cell helper function. *J Exp Med* (2000) 192(11):1553–62. doi: 10.1084/jem.192.11.1553

34. Kim CH, Campbell DJ, Butcher EC. Nonpolarized memory T cells. *Trends Immunol* (2001) 22(10):527–30. doi: 10.1016/s1471-4906(01)02029-4

35. Dupuis J, Boye K, Martin N, Copie-Bergman C, Plonquet A, Fabiani B, et al. Expression of Cxcl13 by neoplastic cells in angioimmunoblastic T-cell lymphoma (Aitl): a new diagnostic marker providing evidence that aitl derives from follicular helper T cells. *Am J Surg Pathol* (2006) 30(4):490–4. doi: 10.1097/00000478-200604000-00009

36. Grogg KL, Attygalle AD, Macon WR, Remstein ED, Kurtin PJ, Dogan A. Expression of Cxcl13, a chemokine highly upregulated in germinal center T-helper cells, distinguishes angioimmunoblastic T-cell lymphoma from peripheral T-cell lymphoma, unspecified. *Modern Pathol an Off J United States Can Acad Pathology Inc* (2006) 19(8):1101–7. doi: 10.1038/modpathol.3800625

37. Vinuesa CG, Linterman MA, Yu D, MacLennan IC. Follicular helper T cells. *Annu Rev Immunol* (2016) 34:335–68. doi: 10.1146/annurev-immunol-041015-055605

38. Stone EL, Pepper M, Katayama CD, Kerdiles YM, Lai CY, Emslie E, et al. Icos coreceptor signaling inactivates the transcription factor Foxo1 to promote tfh cell differentiation. *Immunity* (2015) 42(2):239–51. doi: 10.1016/j.immuni.2015.01.017

39. Crotty S. T Follicular helper cell differentiation, function, and roles in disease. *Immunity* (2014) 41(4):529–42. doi: 10.1016/j.immuni.2014.10.004

40. Ma CS, Tangye SG. Helping the helpers! *Immunity* (2009) 31(1):12–4. doi: 10.1016/j.immuni.2009.06.009

41. Nagoshi H, Kuroda J, Kobayashi T, Maegawa S, Chinen Y, Kiyota M, et al. Clinical manifestation of angioimmunoblastic T-cell lymphoma with exuberant plasmacytosis. *Int J Hematol* (2013) 98(3):366–74. doi: 10.1007/s12185-013-1411-z

42. Bräuninger A, Spieker T, Willenbrock K, Gaulard P, Wacker HH, Rajewsky K, et al. Survival and clonal expansion of mutating "Forbidden" (Immunoglobulin receptor-deficient) Epstein-Barr virus-infected b cells in angioimmunoblastic T cell lymphoma. *J Exp Med* (2001) 194(7):927–40. doi: 10.1084/jem.194.7.927

43. Nicolae A, Pittaluga S, Venkataraman G, Vijnovich-Baron A, Xi L, Raffeld M, et al. Peripheral T-cell lymphomas of follicular T-helper cell derivation with Hodgkin/Reed-sternberg cells of b-cell lineage: both ebv-positive and ebv-negative variants exist. *Am J Surg Pathol* (2013) 37(6):816–26. doi: 10.1097/PAS.0b013e3182785610

44. de Leval L. Approach to nodal-based T-cell lymphomas. *Pathology* (2020) 52(1):78–99. doi: 10.1016/j.pathol.2019.09.012

45. Suefuji N, Niino D, Arakawa F, Karube K, Kimura Y, Kiyasu J, et al. Clinicopathological analysis of a composite lymphoma containing both T- and b-cell lymphomas. *Pathol Int* (2012) 62(10):690–8. doi: 10.1111/j.1440-1827.2012.02858.x

46. Mhaidly R, Krug A, Gaulard P, Lemonnier F, Ricci JE, Verhoeyen E. New preclinical models for angioimmunoblastic T-cell lymphoma: filling the gap. *Oncogenesis* (2020) 9(8):73. doi: 10.1038/s41389-020-00259-x

47. Jaffe AB, Hall A. Rho gtpases: biochemistry and biology. *Annu Rev Cell Dev Biol* (2005) 21:247–69. doi: 10.1146/annurev.cellbio.21.020604.150721

48. Etienne-Manneville S, Hall A. Rho gtpases in cell biology. *Nature* (2002) 420 (6916):629–35. doi: 10.1038/nature01148

49. Cortes JR, Ambesi-Impiombato A, Couronné L, Quinn SA, Kim CS, da Silva Almeida AC, et al. Rhoa G17v induces T follicular helper cell specification and promotes lymphomagenesis. *Cancer Cell* (2018) 33(2):259–73.e7. doi: 10.1016/j.ccell.2018.01.001

50. Yoo HY, Sung MK, Lee SH, Kim S, Lee H, Park S, et al. A recurrent inactivating mutation in rhoa gtpase in angioimmunoblastic T cell lymphoma. *Nat Genet* (2014) 46 (4):371–5. doi: 10.1038/ng.2916

51. de Pádua Covas Lage LA, Barreto GC, Culler HF, Cavalcante JB, de Oliveira Alves LB, Nardinelli L, et al. Tet-2 mutations predict poor outcomes and are associated with unfavorable clinical-biological features in ptcl, not otherwise specified and angioimmunoblastic T-cell lymphoma in Brazilian patients. *Cancer Biomarkers section A Dis Markers* (2022) 35(2):179–91. doi: 10.3233/cbm-220013

52. de Pádua Covas Lage LA, Culler HF, Barreto GC, Reichert CO, Levy D, de Oliveira Costa R, et al. Tumor mutation burden involving epigenetic regulatory genes and the rhoa gtpase predicts overall survival in nodal mature T-cell lymphomas. *Clin Epigenet* (2022) 14(1):180. doi: 10.1186/s13148-022-01395-4

53. Zang S, Li J, Yang H, Zeng H, Han W, Zhang J, et al. Mutations in 5-methylcytosine oxidase Tet2 and rhoa cooperatively disrupt T cell homeostasis. *J Clin Invest* (2017) 127(8):2998–3012. doi: 10.1172/jci92026

54. Fujisawa M, Sakata-Yanagimoto M, Nishizawa S, Komori D, Gershon P, Kiryu M, et al. Activation of rhoa-Vav1 signaling in angioimmunoblastic T-cell lymphoma. *Leukemia* (2018) 32(3):694–702. doi: 10.1038/leu.2017.273

55. Rohde M, Richter J, Schlesner M, Betts MJ, Claviez A, Bonn BR, et al. Recurrent rhoa mutations in pediatric burkitt lymphoma treated according to the nhl-bfm protocols. *Genes Chromosomes Cancer* (2014) 53(11):911–6. doi: 10.1002/gcc.22202

56. Kakiuchi M, Nishizawa T, Ueda H, Gotoh K, Tanaka A, Hayashi A, et al. Recurrent gain-of-Function mutations of rhoa in diffuse-type gastric carcinoma. *Nat Genet* (2014) 46(6):583–7. doi: 10.1038/ng.2984

57. Nagata Y, Kontani K, Enami T, Kataoka K, Ishii R, Totoki Y, et al. Variegated rhoa mutations in adult T-cell Leukemia/Lymphoma. *Blood* (2016) 127(5):596–604. doi: 10.1182/blood-2015-06-644948

58. Pronier E, Delhommeau F. Role of Tet2 mutations in myeloproliferative neoplasms. *Curr Hematol Malig Rep* (2012) 7(1):57–64. doi: 10.1007/s11899-011-0108-8

59. Abdel-Wahab O, Mullally A, Hedvat C, Garcia-Manero G, Patel J, Wadleigh M, et al. Genetic characterization of Tet1, Tet2, and Tet3 alterations in myeloid malignancies. *Blood* (2009) 114(1):144–7. doi: 10.1182/blood-2009-03-210039

60. Lemonnier F, Couronné L, Parrens M, Jaïs JP, Travert M, Lamant L, et al. Recurrent Tet2 mutations in peripheral T-cell lymphomas correlate with tfh-like features and adverse clinical parameters. *Blood* (2012) 120(7):1466–9. doi: 10.1182/blood-2012-02-408542

61. Tahiliani M, Koh KP, Shen Y, Pastor WA, Bandukwala H, Brudno Y, et al. Conversion of 5-methylcytosine to 5-hydroxymethylcytosine in mammalian DNA by mll partner Tet1. *Sci (New York NY)* (2009) 324(5929):930–5. doi: 10.1126/science.1170116

62. Ito S, D'Alessio AC, Taranova OV, Hong K, Sowers LC, Zhang Y. Role of tet proteins in 5mc to 5hmc conversion, Es-cell self-renewal and inner cell mass specification. *Nature* (2010) 466(7310):1129–33. doi: 10.1038/nature09303

63. Moran-Crusio K, Reavie L, Shih A, Abdel-Wahab O, Ndiaye-Lobry D, Lobry C, et al. Tet2 loss leads to increased hematopoietic stem cell self-renewal and myeloid transformation. *Cancer Cell* (2011) 20(1):11–24. doi: 10.1016/j.ccr.2011.06.001

64. Muto H, Sakata-Yanagimoto M, Nagae G, Shiozawa Y, Miyake Y, Yoshida K, et al. Reduced Tet2 function leads to T-cell lymphoma with follicular helper T-Cell-Like features in mice. *Blood Cancer J* (2014) 4(12):e264. doi: 10.1038/bcj.2014.83

65. Ng SY, Yoshida N, Christie AL, Ghandi M, Dharia NV, Dempster J, et al. Targetable vulnerabilities in T- and nk-cell lymphomas identified through preclinical models. *Nat Commun* (2018) 9(1):2024. doi: 10.1038/s41467-018-04356-9

66. Wang C, McKeithan TW, Gong Q, Zhang W, Bouska A, Rosenwald A, et al. Idh2r172 mutations define a unique subgroup of patients with angioimmunoblastic T-cell lymphoma. *Blood* (2015) 126(15):1741–52. doi: 10.1182/blood-2015-05-644591

67. Ward PS, Patel J, Wise DR, Abdel-Wahab O, Bennett BD, Coller HA, et al. The common feature of leukemia-associated Idh1 and Idh2 mutations is a neomorphic enzyme activity converting alpha-ketoglutarate to 2-hydroxyglutarate. *Cancer Cell* (2010) 17(3):225–34. doi: 10.1016/j.ccr.2010.01.020

68. Figueroa ME, Abdel-Wahab O, Lu C, Ward PS, Patel J, Shih A, et al. Leukemic Idh1 and Idh2 mutations result in a hypermethylation phenotype, disrupt Tet2 function, and impair hematopoietic differentiation. *Cancer Cell* (2010) 18(6):553–67. doi: 10.1016/j.ccr.2010.11.015

69. Dang L, Jin S, Su SM. Idh mutations in glioma and acute myeloid leukemia. *Trends Mol Med* (2010) 16(9):387–97. doi: 10.1016/j.molmed.2010.07.002

70. Heavican TB, Bouska A, Yu J, Lone W, Amador C, Gong Q, et al. Genetic drivers of oncogenic pathways in molecular subgroups of peripheral T-cell lymphoma. *Blood* (2019) 133(15):1664–76. doi: 10.1182/blood-2018-09-872549

71. Vallois D, Dobay MP, Morin RD, Lemonnier F, Missiaglia E, Juilland M, et al. Activating mutations in genes related to tcr signaling in angioimmunoblastic and other follicular helper T-Cell-Derived lymphomas. *Blood* (2016) 128(11):1490–502. doi: 10.1182/blood-2016-02-698977

72. Nguyen TB, Sakata-Yanagimoto M, Asabe Y, Matsubara D, Kano J, Yoshida K, et al. Identification of cell-Type-Specific mutations in nodal T-cell lymphomas. *Blood Cancer J* (2017) 7(1):e516. doi: 10.1038/bcj.2016.122

73. Lemonnier F, Cairns RA, Inoue S, Li WY, Dupuy A, Broutin S, et al. The Idh2 R172k mutation associated with angioimmunoblastic T-cell lymphoma produces 2hg in T cells and impacts lymphoid development. *Proc Natl Acad Sci USA* (2016) 113 (52):15084–9. doi: 10.1073/pnas.1617929114

74. Tiacci E, Venanzi A, Ascani S, Marra A, Cardinali V, Martino G, et al. High-risk clonal hematopoiesis as the origin of aitl and Npm1-mutated aml. *New Engl J Med* (2018) 379(10):981–4. doi: 10.1056/NEJMc1806413

75. Jaffe ES, Nicolae A, Pittaluga S. Peripheral T-cell and nk-cell lymphomas in the who classification: pearls and pitfalls. *Modern Pathol an Off J United States Can Acad Pathology Inc* (2013) 26 Suppl 1(Suppl 1):S71–87. doi: 10.1038/modpathol.2012.181

76. Rodriguez-Justo M, Attygalle AD, Munson P, Roncador G, Marafioti T, Piris MA. Angioimmunoblastic T-cell lymphoma with hyperplastic germinal centres: a neoplasia with origin in the outer zone of the germinal centre? clinicopathological and immunohistochemical study of 10 cases with follicular T-cell markers. *Modern Pathol an Off J United States Can Acad Pathology Inc* (2009) 22(6):753–61. doi: 10.1038/modpathol.2009.12

77. Xie Y, Jaffe ES. How I diagnose angioimmunoblastic T-cell lymphoma. *Am J Clin Pathol* (2021) 156(1):1–14. doi: 10.1093/ajcp/aqab090

78. Willenbrock K, Renné C, Gaulard P, Hansmann ML. In angioimmunoblastic T-cell lymphoma, neoplastic T cells may be a minor cell population. a molecular single-cell and immunohistochemical study. *Virchows Archiv an Int J Pathol* (2005) 446 (1):15–20. doi: 10.1007/s00428-004-1114-1

79. Sachsida-Colombo E, Barbosa Mariano LC, Bastos FQ, Rassi AB, Lage LA, Barreto A, et al. A difficult case of angioimmunoblastic T-cell lymphoma to diagnose. *Rev Bras hematologia e hemoterapia* (2016) 38(1):82–5. doi: 10.1016/j.bjhh.2015.11.002

80. Mourad N, Mounier N, Brière J, Raffoux E, Delmer A, Feller A, et al. Clinical, biologic, and pathologic features in 157 patients with angioimmunoblastic T-cell lymphoma treated within the groupe d'etude des lymphomes de l'adulte (Gela) trials. *Blood* (2008) 111(9):4463–70. doi: 10.1182/blood-2007-08-105759

81. Singh A, Schabath R, Ratei R, Stroux A, Klemke CD, Nebe T, et al. Peripheral blood $Scd3^{-}$ $Cd4^{+}$ T cells: a useful diagnostic tool in angioimmunoblastic T cell lymphoma. *Hematological Oncol* (2014) 32(1):16–21. doi: 10.1002/hon.2080

82. Alikhan M, Song JY, Sohani AR, Moroch J, Plonquet A, Duffield AS, et al. Peripheral T-cell lymphomas of follicular helper T-cell type frequently display an aberrant Cd3(-/Dim)Cd4(+) population by flow cytometry: an important clue to the diagnosis of a Hodgkin lymphoma mimic. *Modern Pathol an Off J United States Can Acad Pathology Inc* (2016) 29(10):1173–82. doi: 10.1038/modpathol.2016.113

83. Merchant SH, Amin MB, Viswanatha DS. Morphologic and immunophenotypic analysis of angioimmunoblastic T-cell lymphoma: emphasis on phenotypic aberrancies for early diagnosis. *Am J Clin Pathol* (2006) 126(1):29–38. doi: 10.1309/28yp-0del-gkej-grxg

84. Dorfman DM, Brown JA, Shahsafaei A, Freeman GJ. Programmed death-1 (Pd-1) is a marker of germinal center-associated T cells and angioimmunoblastic T-cell lymphoma. *Am J Surg Pathol* (2006) 30(7):802–10. doi: 10.1097/01.pas.0000209855.28282.ce

85. Roncador G, García Verdes-Montenegro JF, Tedoldi S, Paterson JC, Klapper W, Ballabio E, et al. Expression of two markers of germinal center T cells (Sap and pd-1) in angioimmunoblastic T-cell lymphoma. *Haematologica* (2007) 92(8):1059–66. doi: 10.3324/haematol.10864

86. Gaulard P, de Leval L. Follicular helper T cells: implications in neoplastic hematopathology. *Semin Diagn Pathol* (2011) 28(3):202–13. doi: 10.1053/j.semdp.2011.03.003

87. Balagué O, Martínez A, Colomo L, Roselló E, Garcia A, Martínez-Bernal M, et al. Epstein-Barr Virus negative clonal plasma cell proliferations and lymphomas in peripheral T-cell lymphomas: a phenomenon with distinctive clinicopathologic features. *Am J Surg Pathol* (2007) 31(9):1310–22. doi: 10.1097/PAS.0b013e3180339f18

88. Huppmann AR, Roullet MR, Raffeld M, Jaffe ES. Angioimmunoblastic T-cell lymphoma partially obscured by an Epstein-Barr virus-negative clonal plasma cell proliferation. *J Clin Oncol Off J Am Soc Clin Oncol* (2013) 31(2):e28–30. doi: 10.1200/jco.2012.43.3797

89. Dunleavy K, Wilson WH, Jaffe ES. Angioimmunoblastic T cell lymphoma: pathobiological insights and clinical implications. *Curr Opin Hematol* (2007) 14 (4):348–53. doi: 10.1097/MOH.0b013e328186ffbf

90. Weiss LM, Jaffe ES, Liu XF, Chen YY, Shibata D, Medeiros LJ. Detection and localization of Epstein-Barr viral genomes in angioimmunoblastic lymphadenopathy and angioimmunoblastic lymphadenopathy-like lymphoma. *Blood* (1992) 79(7):1789–95.

91. Willenbrock K, Bräuninger A, Hansmann ML. Frequent occurrence of b-cell lymphomas in angioimmunoblastic T-cell lymphoma and proliferation of Epstein-Barr virus-infected cells in early cases. *Br J Haematol* (2007) 138(6):733–9. doi: 10.1111/j.1365-2141.2007.06725.x

92. Konstantinou K, Yamamoto K, Ishibashi F, Mizoguchi Y, Kurata M, Nakagawa Y, et al. Angiogenic mediators of the angiopoietin system are highly expressed by Cd10-positive lymphoma cells in angioimmunoblastic T-cell lymphoma. *Br J Haematol* (2009) 144(5):696–704. doi: 10.1111/j.1365-2141.2008.07534.x

93. Zhao WL, Mourah S, Mounier N, Leboeuf C, Daneshpouy ME, Legrès L, et al. Vascular endothelial growth factor-a is expressed both on lymphoma cells and endothelial cells in angioimmunoblastic T-cell lymphoma and related to lymphoma progression. *Lab investigation; J Tech Methods Pathol* (2004) 84(11):1512–9. doi: 10.1038/labinvest.3700145

94. Dogan A, Attygalle AD, Kyriakou C. Angioimmunoblastic T-cell lymphoma. *Br J Haematol* (2003) 121(5):681–91. doi: 10.1046/j.1365-2141.2003.04335.x

95. Alizadeh AA, Advani RH. Evaluation and management of angioimmunoblastic T-cell lymphoma: a review of current approaches and future strategies. *Clin Adv Hematol Oncol H&O* (2008) 6(12):899–909.

96. Streubel B, Vinatzer U, Willheim M, Raderer M, Chott A. Novel T(5;9)(Q33; Q22) fuses itk to syk in unspecified peripheral T-cell lymphoma. *Leukemia* (2006) 20 (2):313–8. doi: 10.1038/sj.leu.2404045

97. Nelson M, Horsman DE, Weisenburger DD, Gascoyne RD, Dave BJ, Loberiza FR, et al. Cytogenetic abnormalities and clinical correlations in peripheral T-cell lymphoma. *Br J Haematol* (2008) 141(4):461–9. doi: 10.1111/j.1365-2141.2008.07042.x

98. Tan BT, Warnke RA, Arber DA. The frequency of b- and T-cell gene rearrangements and Epstein-Barr virus in T-cell lymphomas: a comparison between angioimmunoblastic T-cell lymphoma and peripheral T-cell lymphoma, unspecified with and without associated b-cell proliferations. *J Mol Diagn JMD* (2006) 8(4):466–75. doi: 10.2353/jmoldx.2006.060016

99. Hathuc V, Kreisel F. Genetic landscape of peripheral T-cell lymphoma. *Life (Basel Switzerland)* (2022) 12(3):1–22. doi: 10.3390/life12030410

100. Wang M, Zhang S, Chuang SS, Ashton-Key M, Ochoa E, Bolli N, et al. Angioimmunoblastic T cell lymphoma: novel molecular insights by mutation profiling. *Oncotarget* (2017) 8(11):17763–70. doi: 10.18632/oncotarget.14846

101. Rohr J, Guo S, Huo J, Bouska A, Lachel C, Li Y, et al. Recurrent activating mutations of Cd28 in peripheral T-cell lymphomas. *Leukemia* (2016) 30(5):1062–70. doi: 10.1038/leu.2015.357

102. Tokunaga T, Shimada K, Yamamoto K, Chihara D, Ichihashi T, Oshima R, et al. Retrospective analysis of prognostic factors for angioimmunoblastic T-cell lymphoma: a multicenter cooperative study in Japan. *Blood* (2012) 119(12):2837–43. doi: 10.1182/blood-2011-08-374371

103. Hong H, Fang X, Wang Z, Huang H, Lam ST, Li F, et al. Angioimmunoblastic T-cell lymphoma: a prognostic model from a retrospective study. *Leukemia Lymphoma* (2018) 59(12):2911–6. doi: 10.1080/10428194.2018.1459610

104. de Pádua Covas Lage LA, Hamasaki DT, Moreira FR, Rocha V, Zerbini MCN, Pereira J. Absolute monocyte count is a predictor of overall survival and progression-free survival in nodal peripheral T cell lymphoma. *Ann Hematol* (2019) 98(9):2097–102. doi: 10.1007/s00277-019-03731-w

105. Iqbal J, Wright G, Wang C, Rosenwald A, Gascoyne RD, Weisenburger DD, et al. Gene expression signatures delineate biological and prognostic subgroups in peripheral T-cell lymphoma. *Blood* (2014) 123(19):2915–23. doi: 10.1182/blood-2013-11-536359

106. Delfau-Larue MH, de Leval L, Joly B, Plonquet A, Challine D, Parrens M, et al. Targeting intratumoral b cells with rituximab in addition to chop in angioimmunoblastic T-cell lymphoma. *A Clinicobiological Study Gela. Haematologica* (2012) 97(10):1594–602. doi: 10.3324/haematol.2011.061507

107. Eladl AE, Shimada K, Suzuki Y, Takahara T, Kato S, Kohno K, et al. Ebv status has prognostic implication among young patients with angioimmunoblastic T-cell lymphoma. *Cancer Med* (2020) 9(2):678–88. doi: 10.1002/cam4.2742

108. Wei C, Li W, Qin L, Liu S, Xue C, Ren K, et al. Clinicopathologic characteristics, outcomes, and prognostic factors of angioimmunoblastic T-cell lymphoma in China. *Cancer Med* (2022) 12:3987–98. doi: 10.1002/cam4.5248

109. Shang Y, Fu X, Chang Y, Li Y, Zhang M. B2 microglobulin is a novel prognostic marker of angioimmunoblastic T-cell lymphoma. *Sci Rep* (2018) 8(1):12907. doi: 10.1038/s41598-018-31212-z

110. Moskowitz AJ. Practical treatment approach for angioimmunoblastic T-cell lymphoma. *J Oncol Pract* (2019) 15(3):137–43. doi: 10.1200/jop.18.00511

111. Mohammed Saleh MF, Kotb A, Abdallah GEM, Muhsen IN, El Fakih R, Aljurf M. Recent advances in diagnosis and therapy of angioimmunoblastic T cell lymphoma. *Curr Oncol (Toronto Ont)* (2021) 28(6):5480–98. doi: 10.3390/curroncol28060456

112. Majumdar G. Singh AK. p-glycoprotein expression in drug-resistant chronic lymphoproliferative disorder. *Leukemia Lymphoma* (1991) 5(5-6):387–90. doi: 10.3109/10428199109067633

113. Nooter K, Sonneveld P. Clinical relevance of p-glycoprotein expression in haematological malignancies. *Leukemia Res* (1994) 18(4):233–43. doi: 10.1016/0145-2126(94)90025-6

114. Kansara R, Savage KJ. The problem with cyclophosphamide, doxorubicin, vincristine and prednisone for the treatment of peripheral T-cell lymphoma. *Leukemia Lymphoma* (2014) 55(4):727–9. doi: 10.3109/10428194.2013.858154

115. Savage KJ, Chhanabhai M, Gascoyne RD, Connors JM. Characterization of peripheral T-cell lymphomas in a single north American institution by the who classification. *Ann Oncol Off J Eur Soc Med Oncol* (2004) 15(10):1467–75. doi: 10.1093/annonc/mdh392

116. Ellin F, Landström J, Jerkeman M, Relander T. Real-world data on prognostic factors and treatment in peripheral T-cell lymphomas: a study from the Swedish lymphoma registry. *Blood* (2014) 124(10):1570–7. doi: 10.1182/blood-2014-04-573089

117. Horwitz SM, Ansell S, Ai WZ, Barnes J, Barta SK, Brammer J, et al. T-Cell lymphomas, version 2.2022, nccn clinical practice guidelines in oncology. *J Natl Compr Cancer Network JNCCN* (2022) 20(3):285–308. doi: 10.6004/jnccn.2022.0015

118. Schmitz N, Trümper L, Ziepert M, Nickelsen M, Ho AD, Metzner B, et al. Treatment and prognosis of mature T-cell and nk-cell lymphoma: an analysis of patients with T-cell lymphoma treated in studies of the German high-grade non-Hodgkin lymphoma study group. *Blood* (2010) 116(18):3418–25. doi: 10.1182/blood-2010-02-270785

119. Gallamini A, Zaja F, Patti C, Billio A, Specchia MR, Tucci A, et al. Alemtuzumab (Campath-1h) and chop chemotherapy as first-line treatment of peripheral T-cell lymphoma: results of a gitil (Gruppo italiano terapie innovative nei linfomi) prospective multicenter trial. *Blood* (2007) 110(7):2316–23. doi: 10.1182/blood-2007-02-074641

120. Ruan J, Moskowitz AJ, Mehta-Shah N, Sokol L, Chen Z, Kotlov N, et al. Multicenter phase 2 study of oral azacitidine (Cc-486) plus chop as initial treatment for peripheral T-cell lymphoma. *Blood* (2023). doi: 10.1182/blood.2022018254

121. Bachy E, Camus V, Thieblemont C, Sibon D, Casasnovas RO, Ysebaert L, et al. Romidepsin plus chop versus chop in patients with previously untreated peripheral T-cell lymphoma: results of the ro-chop phase iii study (Conducted by lysa). *J Clin Oncol Off J Am Soc Clin Oncol* (2022) 40(3):242–51. doi: 10.1200/jco.21.01815

122. Simon A, Peoch M, Casassus P, Deconinck E, Colombat P, Desablens B, et al. Upfront vip-Reinforced-Abvd (Vip-rabvd) is not superior to Chop/21 in newly diagnosed peripheral T cell lymphoma. results of the randomized phase iii trial goelams-Ltp95. *Br J Haematol* (2010) 151(2):159–66. doi: 10.1111/j.1365-2141.2010.08329.x

123. Mahadevan D, Unger JM, Spier CM, Persky DO, Young F, LeBlanc M, et al. Phase 2 trial of combined cisplatin, etoposide, gemcitabine, and methylprednisolone (Pegs) in peripheral T-cell non-Hodgkin lymphoma: southwest oncology group study S0350. *Cancer* (2013) 119(2):371–9. doi: 10.1002/cncr.27733

124. Gisselbrecht C, Lepage E, Molina T, Quesnel B, Fillet G, Lederlin P, et al. Shortened first-line high-dose chemotherapy for patients with poor-prognosis aggressive lymphoma. *J Clin Oncol Off J Am Soc Clin Oncol* (2002) 20(10):2472–9. doi: 10.1200/jco.2002.02.125

125. Brink M, Meeuwes FO, van der Poel MWM, Kersten MJ, Wondergem M, Mutsaers P, et al. Impact of etoposide and asct on survival among patients aged <65 years with stage ii to iv ptcl: a population-based cohort study. *Blood* (2022) 140 (9):1009–19. doi: 10.1182/blood.2021015114

126. Kim J, Cho J, Byeon S, Kim WS, Kim SJ. Comparison of first-line treatments of peripheral T-cell lymphoma according to regimen: a systematic review and meta-analysis. *Hematological Oncol* (2021) 39(5):664–73. doi: 10.1002/hon.2924

127. Deng S, Lin S, Shen J, Zeng Y. Comparison of chop vs chope for treatment of peripheral T-cell lymphoma: a meta-analysis. *OncoTargets Ther* (2019) 12:2335–42. doi: 10.2147/ott.S189825

128. Ganjoo K, Hong F, Horning SJ, Gascoyne RD, Natkunam Y, Swinnen LJ, et al. Bevacizumab and cyclosphosphamide, doxorubicin, vincristine and prednisone in combination for patients with peripheral T-cell or natural killer cell neoplasms: an Eastern cooperative oncology group study (E2404). *Leukemia Lymphoma* (2014) 55 (4):768–72. doi: 10.3109/10428194.2013.816700

129. Lemonnier F, Safar V, Beldi-Ferchiou A, Cottereau AS, Bachy E, Cartron G, et al. Integrative analysis of a phase 2 trial combining lenalidomide with chop in angioimmunoblastic T-cell lymphoma. *Blood Adv* (2021) 5(2):539–48. doi: 10.1182/bloodadvances.2020003081

130. Meeuwes FO, Brink M, van der Poel MWM, Kersten MJ, Wondergem M, Mutsaers P, et al. Impact of rituximab on treatment outcomes of patients with angioimmunoblastic T-cell lymphoma; a population-based analysis. *Eur J Cancer (Oxford Engl 1990)* (2022) 176:100–9. doi: 10.1016/j.ejca.2022.09.008

131. Horwitz S, O'Connor OA, Pro B, Illidge T, Fanale M, Advani R, et al. Brentuximab vedotin with chemotherapy for Cd30-positive peripheral T-cell lymphoma (Echelon-2): a global, double-blind, randomised, phase 3 trial. *Lancet (London England)* (2019) 393(10168):229–40. doi: 10.1016/s0140-6736(18)32984-2

132. Timmins MA, Wagner SD, Ahearne MJ. The new biology of ptcl-nos and aitl: current status and future clinical impact. *Br J Haematol* (2020) 189(1):54–66. doi: 10.1111/bjh.16428

133. Lemonnier F, Dupuis J, Sujobert P, Tournillhac O, Cheminant M, Sarkozy C, et al. Treatment with 5-azacytidine induces a sustained response in patients with angioimmunoblastic T-cell lymphoma. *Blood* (2018) 132(21):2305–9. doi: 10.1182/blood-2018-04-840538

134. Falchi L, Ma H, Klein S, Lue JK, Montanari F, Marchi E, et al. Combined oral 5-azacytidine and romidepsin are highly effective in patients with ptcl: a multicenter phase 2 study. *Blood* (2021) 137(16):2161–70. doi: 10.1182/blood.2020009004

135. O'Connor OA, Falchi L, Lue JK, Marchi E, Kinahan C, Sawas A, et al. Oral 5-azacytidine and romidepsin exhibit marked activity in patients with ptcl: a multicenter phase 1 study. *Blood* (2019) 134(17):1395–405. doi: 10.1182/blood.2019001285

136. Dupuis J, Morschhauser F, Ghesquières H, Tilly H, Casasnovas O, Thieblemont C, et al. Combination of romidepsin with cyclophosphamide, doxorubicin, vincristine, and prednisone in previously untreated patients with peripheral T-cell lymphoma: a non-randomised, phase 1b/2 study. *Lancet Haematol* (2015) 2(4):e160–5. doi: 10.1016/s2352-3026(15)00023-x

137. Fossard G, Broussais F, Coelho I, Bailly S, Nicolas-Virelizier E, Toussaint E, et al. Role of up-front autologous stem-cell transplantation in peripheral T-cell lymphoma for patients in response after induction: an analysis of patients from lysa centers. *Ann Oncol Off J Eur Soc Med Oncol* (2018) 29(3):715–23. doi: 10.1093/annonc/mdx787

138. Abramson JS, Feldman T, Kroll-Desrosiers AR, Muffly LS, Winer E, Flowers CR, et al. Peripheral T-cell lymphomas in a Large us multicenter cohort: prognostication in the modern era including impact of frontline therapy. *Ann Oncol Off J Eur Soc Med Oncol* (2014) 25(11):2211–7. doi: 10.1093/annonc/mdu443

139. Reimer P, Rüdiger T, Geissinger E, Weissinger F, Nerl C, Schmitz N, et al. Autologous stem-cell transplantation as first-line therapy in peripheral T-cell lymphomas: results of a prospective multicenter study. *J Clin Oncol Off J Am Soc Clin Oncol* (2009) 27(1):106–13. doi: 10.1200/jco.2008.17.4870

140. d'Amore F, Relander T, Lauritzsen GF, Jantunen E, Hagberg H, Anderson H, et al. Up-front autologous stem-cell transplantation in peripheral T-cell lymphoma: nlg-T-01. *J Clin Oncol Off J Am Soc Clin Oncol* (2012) 30(25):3093–9. doi: 10.1200/jco.2011.40.2719

141. Park SI, Horwitz SM, Foss FM, Pinter-Brown LC, Carson KR, Rosen ST, et al. The role of autologous stem cell transplantation in patients with nodal peripheral T-cell lymphomas in first complete remission: report from complete, a prospective, multicenter cohort study. *Cancer* (2019) 125(9):1507–17. doi: 10.1002/cncr.31861

142. Cho H, Yoon DH, Shin DY, Koh Y, Yoon SS, Kim SJ, et al. Current treatment patterns and the role of upfront autologous stem cell transplantation in patients with peripheral T-cell lymphoma: a Korean nationwide, multicenter prospective registry study (Cisl 1404). *Cancer Res Treat* (2023) 55(2):684–92. doi: 10.4143/crt.2022.1434

143. Schmitz N, Truemper L, Bouabdallah K, Ziepert M, Leclerc M, Cartron G, et al. A randomized phase 3 trial of autologous vs allogeneic transplantation as part of first-line therapy in poor-risk peripheral T-nhl. *Blood* (2021) 137(19):2646–56. doi: 10.1182/blood.2020008825

144. Rodríguez J, Caballero MD, Gutiérrez A, Marín J, Lahuerta JJ, Sureda A, et al. High-dose chemotherapy and autologous stem cell transplantation in peripheral T-cell lymphoma: the gel-tamo experience. *Ann Oncol Off J Eur Soc Med Oncol* (2003) 14 (12):1768–75. doi: 10.1093/annonc/mdg459

145. Nademanee A, Palmer JM, Popplewell L, Tsai NC, Delioukina M, Gaal K, et al. High-dose therapy and autologous hematopoietic cell transplantation in peripheral T cell lymphoma (Ptcl): analysis of prognostic factors. *Biol Blood marrow Transplant J Am Soc Blood Marrow Transplant* (2011) 17(10):1481–9. doi: 10.1016/j.bbmt.2011.02.008

146. Smith SM, Burns LJ, van Besien K, Lerademacher J, He W, Fenske TS, et al. Hematopoietic cell transplantation for systemic mature T-cell non-Hodgkin lymphoma. *J Clin Oncol Off J Am Soc Clin Oncol* (2013) 31(25):3100–9. doi: 10.1200/jco.2012.46.0188

147. El-Asmar J, Reljic T, Ayala E, Hamadani M, Nishihori T, Kumar A, et al. Efficacy of high-dose therapy and autologous hematopoietic cell transplantation in peripheral T cell lymphomas as front-line consolidation or in the Relapsed/Refractory setting: a systematic Review/Meta-analysis. *Biol Blood marrow Transplant J Am Soc Blood Marrow Transplant* (2016) 22(5):802–14. doi: 10.1016/j.bbmt.2015.12.004

148. Corradini P, Dodero A, Zallio F, Caracciolo D, Casini M, Bregni M, et al. Graft-Versus-Lymphoma effect in relapsed peripheral T-cell non-hodgkin's lymphomas after reduced-intensity conditioning followed by allogeneic transplantation of hematopoietic cells. *J Clin Oncol Off J Am Soc Clin Oncol* (2004) 22(11):2172–6. doi: 10.1200/jco.2004.12.050

149. Le Gouill S, Milpied N, Buzyn A, De Latour RP, Vernant JP, Mohty M, et al. Graft-Versus-Lymphoma effect for aggressive T-cell lymphomas in adults: a study by the société francaise de greffe de moëlle et de thérapie cellulaire. *J Clin Oncol Off J Am Soc Clin Oncol* (2008) 26(14):2264–71. doi: 10.1200/jco.2007.14.1366

150. Dodero A, Spina F, Narni F, Patriarca F, Cavattoni I, Benedetti F, et al. Allogeneic transplantation following a reduced-intensity conditioning regimen in Relapsed/Refractory peripheral T-cell lymphomas: long-term remissions and response to donor lymphocyte infusions support the role of a graft-Versus-Lymphoma effect. *Leukemia* (2012) 26(3):520–6. doi: 10.1038/leu.2011.240

151. Amengual JE, Lichtenstein R, Lue J, Sawas A, Deng C, Lichtenstein E, et al. A phase 1 study of romidepsin and pralatrexate reveals marked activity in relapsed and refractory T-cell lymphoma. *Blood* (2018) 131(4):397–407. doi: 10.1182/blood-2017-09-806737

152. Yhim HY, Kim T, Kim SJ, Shin HJ, Koh Y, Kim JS, et al. Combination treatment of copanlisib and gemcitabine in Relapsed/Refractory ptcl (Cosmos): an open-label phase I/Ii trial. *Ann Oncol Off J Eur Soc Med Oncol* (2021) 32(4):552–9. doi: 10.1016/j.annonc.2020.12.009

153. Luo L, Zhou X, Zhou L, Liang Z, Yang J, Tu S, et al. Current state of car-T therapy for T-cell malignancies. *Ther Adv Hematol* (2022) 13:20406207221143025. doi: 10.1177/20406207221143025

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 28 April 2023
DOI 10.3389/fonc.2023.1150715

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Ali Sakhdari,
University Health Network (UHN), Canada
Carlos Murga-Zamalloa,
University of Illinois at Chicago,
United States

*CORRESPONDENCE
Jasmine Zain
jazain@coh.org

RECEIVED 24 January 2023
ACCEPTED 06 March 2023
PUBLISHED 28 April 2023

CITATION
Zain J and Kallam A (2023) Challenges in nodal peripheral T-cell lymphomas: from biological advances to clinical applicability. *Front. Oncol.* 13:1150715. doi: 10.3389/fonc.2023.1150715

Challenges in nodal peripheral T-cell lymphomas: from biological advances to clinical applicability

Jasmine Zain* and Avyakta Kallam

Department of Hematology/Hematopoietic Cell Transplantation, City of Hope Medical Center, Duarte, CA, United States

T cell lymphomas are a heterogenous group with varying biological and clinical features that tend to have poor outcomes with a few exceptions. They account for 10-15% of all non-Hodgkin lymphomas (NHL), and 20% of aggressive NHL. There has been little change in the overall prognosis of T cell lymphomas over the last 2 decades. Most subtypes carry an inferior prognosis when compared to the B cell lymphomas, with a 5-year OS of 30%. Gene expression profiling and other molecular techniques has enabled a deeper understanding of these differences in the various subtypes as reflected in the latest 5th WHO and ICC classification of T cell lymphomas. It is becoming increasingly clear that therapeutic approaches that target specific cellular pathways are needed to improve the clinical outcomes of T cell lymphomas. This review will focus on nodal T cell lymphomas and describe novel treatments and their applicability to the various subtypes.

KEYWORDS

T cell lymphoma, updated classification, PTCL-NOS, TFH-cell lymphoma, anaplastic large cell lymphoma, novel therapies

1 Introduction

A diagnosis of Peripheral T cell lymphomas (PTCL) remains challenging and confusing for physicians as well as patients. This has to do with the rarity of this group of diseases with clinical features that overlap other illnesses and an overall poor prognosis. PTCL are a heterogenous group of lymphomas, arising from the post thymic T lymphocytes. They account for 10-15% of all non-Hodgkin lymphomas (NHL), and 20% of aggressive NHL. The incidence of PTCL in all age groups is 2.1 per 100,000 (1). The recent world health organization (WHO) classification describes over 30 distinct subtypes of T cell lymphomas (2). In 2004, a letter to the editor identified the "large and unexplored problem of T cell lymphomas" (3) and outlined the poor outcome of T cell lymphomas compared to aggressive diffuse B cell lymphomas by publishing a survival curve that showed a 5-year survival of 30% in PTCL vs over 50% for B cell lymphomas with CHOP like therapies only with the exception of anaplastic large cell lymphoma. They pointed out that this was very

likely the result of the very aggressive nature of T cell lymphomas and lack of specific therapies for T cell lymphomas. There was no reason to believe that regimens used to treat aggressive B cell lymphomas should work in T cell lymphomas. Since then, there has been a vast increase in our understanding of T cell lymphomas and there have been treatments studied and approved that are specific to T cell lymphomas. These include pralatrexate, romidepsin, belinostat and Brentuximab Vedotin (4–7). Despite this, there has been little change in the overall prognosis of T cell lymphomas over the last 2 decades (8). Most subtypes carry an inferior prognosis when compared to the B cell lymphomas, with a 5-year OS of 30% (9, 10). This is in part due to lack of understanding of subtype specific pathogenesis as well as the challenges of performing prospective clinical trials evaluating therapies in this field of rare diseases.

In recent years, techniques of molecular biology have allowed the exploration of the genomic landscape of various subtypes of PTCL leading to a better understanding of the pathobiology of the diseases, as well as risk stratification and have paved a way for development of targeted molecules. Gene expression profiling (GEP) has been instrumental towards better identifying the subsets of PTCL and identifying prognostic markers. GEP and whole genome sequencing have also recognized distinct epigenetic regulators contributing to the lymphomagenesis (11). This is now leading to development of targeted therapies that are expected to change the treatment paradigms of PTCL. This article with review the current developments in the understanding of nodal PTCL from lymphomagenesis to therapeutic developments.

2 Updates in classification of PTCL

The most recent fifth edition of the world health organization (WHO) classification (2) and the international consensus classification of mature lymphoid neoplasms (ICC 2022) (12) now incorporates the recent advances in the understanding of T cell lymphomas. The mature T cell and NK-cell neoplasms are broadly grouped into 9 types, based on the cell of origin, cytomorphology, disease localization and clinical features (2). Broadly classified into nodal T cell lymphomas and extra-nodal leukemic T cell lymphomas, the major subtypes are summarized in Table 1 alongwith the comparison to the older 2017 classification. A more detailed description of the individual nodal subtypes are described below:

The various subtypes of peripheral T cells that form the complex immune system leave the thymus as mature T cells after undergoing T cell receptor (TCR) rearrangement, a process that is mediated by a complex array of cytokines and their receptors. The

TABLE 1 Outlines the changes made to the nomenclature/diagnostic criteria for nodal PTCL's.

2022 ICC Classification	2022 WHO classification	2017 updated WHO classification	Comments
Peripheral T-cell lymphoma, not otherwise specified	Peripheral T-cell lymphoma, not otherwise specified	Peripheral T-cell lymphoma, not otherwise specified	Remains a diagnosis of exclusion; Two biological variants - PTCL-TBX21 and PTCL-GATA3 are identified. Cytotoxic EBV-negative cases may represent a distinct subgroup
TFH lymphoma, angioimmunoblastic subtype	Nodal TFH cell lymphoma, angioimmunoblastic-type	Angioimmunoblastic T cell lymphoma	Diagnostic criteria is unchanged. Grouped under nodal T follicular helper cell lymphoma (TFH). New terminology introduced to group lymphomas with similar phenotype and gene expression profiling signatures.
TFH lymphoma, follicular type	Nodal TFH cell lymphoma, follicular-type	Follicular T cell lymphoma	New nomenclature. Diagnostic criteria is unchanged.
TFH lymphoma, NOS	Nodal TFH cell lymphoma, NOS	Nodal peripheral T-cell lymphoma with TFH phenotype	New nomenclature. Diagnostic criteria is unchanged.
Anaplastic large cell lymphoma, ALK positive	ALK positive, Anaplastic large cell lymphoma	Anaplastic large cell lymphoma, ALK-positive	No change in diagnostic criteria
Anaplastic large cell lymphoma, ALK negative	ALK negative, Anaplastic large cell lymphoma	Anaplastic large cell lymphoma, ALK-positive	No change in the diagnostic criteria. Recognized as a heterogenous entity. TP63 rearrangements,
Breast implant associated ALCL	Breast implant associated ALCL	Breast implant associated ALCL	Overexpression of PD-L1 in over half the cases Constitutive JAK-STAT activation by somatic mutations of STAT3, STAT5B, JAK1 and JAK2 and loss-of function mutations of SOCS1 and SOCS3
Primary nodal EBV+ NK/T cell lymphoma	EBV+ve nodal NK and T cell lymphoma	New entity	New distinct entity from PTCL -NOS. Now grouped under EBV+ve positive NK/T cell lymphomas

The table compares the nomenclature differences between the recent classifcations for nodal T cell lymphomas to the 2017 revised WHO classification. 2022 (ICC) International consensus classification of mature lymphoid neoplasms;. 2022 (WHO) World health organization classification of T cell lymphomas. TFH, T follicular helper cell; NOS, not otherwise specified; ALK, anaplastic lymphoma kinase; ALCL, anaplastic large cell lymphoma; EBV, Epstein barr virus.

differentiation of these cells is mediated by transcription factors such as FOXP3 for T_{regs}, Bcl 6 for T_{FH} Tbet for Th1 and GATA3 for Th2 transcription factors. Each of these cells secrete distinct cytokines, which play a role in the signaling pathways of the immune system and dysregulation of these pathways is thought to play a role in the tumorigenesis. The variability among several types of T cells and their differentiation forms the basis for the heterogeneity noted in PTCL. Gene expression profile (GEP) studies have identified subsets of PTCL's with molecular signatures similar to the non-neoplastic cells (11, 13–15).

2.1 PTCL-Nos

PTCL – NOS is a heterogenous category that does not correspond to any defined subgroup of PTCL and is typically a diagnosis of exclusion. PTCL- NOS is the most common subtype in the Western countries. It affects older patients, with a mean age of diagnosis of 60 years. There is a slight male predilection, with a majority of patients presenting with advanced stage disease. Patients typically present with nodal disease. They may also be extra-nodal involvement, with skin and gastrointestinal tract being the most common extra nodal sites of disease (16). Morphologically, PTCL-NOS demonstrates paracortical infiltrates, with effacement of the normal lymph node architecture. It is associated with expression of CD3, CD4, loss of CD7, CD5. CD30 is expressed in about 32% to 64% of cases with PTCL-NOS (17). Epstein barr virus has been reported to be present in 30% of all PTCL-NOS (16). However, in the recent 5th WHO classification, the EBV+ ve lymphomas are recognized as a distinct entity and are grouped under EBV+ve NK/T cell lymphomas. GEP studies identified two sub-groups within PTCL- NOS (11). One sub-group, designated as PTCL- TBX21 is characterized by expression of TBX21 and target genes such as CXCR3, CCL3 and IL2RB. TBX21 is the master regulator of Th1 cells. PTCL- TBX21 subtype is associated with dysregulated nuclear factor kappa B(NF-kB) pathway (18). The other group, PTCL- GATA3 is characterized by overexpression of GATA3 and its target genes such as CCR4, CXCR7, IL18RA an IK (19). and regulates Th2 differentiation, pointing towards a different cell of origin for these two subtypes. On the other hand, PTCL -GATA3 seen in 33% of the cases and is associated with poor prognosis, irrespective of the TpP53 status. Next generation sequencing shows PTCL- GATA3 to have high genomic complexity, with frequent loss of Tp53, PTEN and CDKNA (18). It is characterized by MYC over-expression and has more cytotoxic features. PTCL- GATA3 subtype is associated with altered PI3 Kinase – mTOR signaling as well (18). Although these are not recognized as distinct subtypes in the WHO classification, they are noted to have different morphological features. PTCL-TBX21 has a more polymorphic background, with reactive inflammatory cells when compared to PTCL -GATA3. PTCL -GATA3 has sheets of medium sized tumor cells with clear cytoplasm and is characterized by a lack of inflammatory background (19).

Epigenetic mutations (described in detail below) are less frequent than in AITL or PTCL with T_{FH} phenotype. TET2 mutations are seen on 38%-49%, DNMT3A (5%-36%) of PTCL-NOS. Mutations in epigenetic regulator genes such as KMT2C, KMT2D, KMT2A could have a possible association with a poor progression free survival and predict responsiveness to HDAC inhibitors, but more studies are warranted. Mutations in the TCT signaling/NF-kB genes have been reported in VAV1, RHOA, CARD11, CD23 and PTPRX (20). In-frame fusion transcripts involving TCR signaling genes have been identified [FYN: TRAF3IP2, KHDRBS1:LCK], which activate signaling pathways downstream of TCR complex and could be a potential target for tyrosine kinase inhibitors. Recurrent mutations in a tumor suppressor gene, FAT1 have been found in 39% of the cases and is associated with a poor prognosis (21).

2.2 Nodal T follicular helper cell lymphoma

This new nomenclature was introduced in the 2022 5th WHO classification to group together PTCLS that have a T_{FH} phenotype, overlapping molecular and clinical features. Specific diagnostic criteria have been described that include a primary nodal/systemic process expressing CD4 and have three of the five TFH markers, i.e CD10, BCL6, CD279, CXCL13, and ICOS. The cells also tend to express T cell surface antigens -CD2, CD3 and CD5, which can be detected on flow cytometry or on immunohistochemical staining. The expression of at least two TFH markers (CD10, BCL-6, CD279, CXCL13 and ICOS) are required only for nodal T-follicular helper lymphomas (nTFH), non-otherwise specified (nTFH-NOS); previously designated as Peripheral T-cell lymphoma with follicular helper phenotype (PTCL-TFH). The AITL group and follicular type do not require a threshold cut-off expression of those markers as the morphological architecture is characteristic.

There are three nodal lymphomas which have TFH cell origin that are sub grouped under this category - angioimmunoblastic subtype (previously known as AITL), Nodal TFH cell lymphoma, follicular-type (previously known as follicular T cell lymphoma) and Nodal TFH cell lymphoma, NOS (previously known as Nodal peripheral T cell lymphoma with a T follicular helper phenotype). GEP has been instrumental in differentiating among these subtypes (22).

2.2.1 TFH lymphoma, angioimmunoblastic subtype [TFH lymphoma - AITL subtype]

TFH lymphoma - AITL subtype is commonly seen in older individuals and is characterized by diffuse lymphadenopathy, along with diverse constitutional signs and symptoms. It is often associated with skin rashes, and autoimmune features, including cold agglutinin hemolytic anemia and immune mediated cytopenia's. Epstein-Barr virus (EBV) positive B immunoblasts are present, but the neoplastic cells do not have EBV (22). AITL exhibits various histological patterns and is often associated with a prominent microenvironment that can obscure the neoplastic cells, atypical B cell proliferations and clonal B cell expansion can be seen. This lymphoma is characterized by effacement of the normal architecture of the lymph node, with diffuse infiltrates of medium

sized, atypical lymphocytes with clear cytoplasm Proliferation of arborizing post capillary vessels, is considered one of the hallmarks of this disease. TFH -AITL subtype is most frequently found to have mutations resulting in epigenetic dysregulation and can explain the higher response rates that are seen with epigenetic therapies. TET2 is present in 50-90% of cases, DNMT3A and IDH2 are seen on 20% to 55% of cases (23) An inactivating RHOA mutation can be seen in 70% of TFH lymphoma -AITL subtype and usually co-occurs with TET2 mutations. $RHOA_{GI7V}$ mutated AITL has a higher incidence of B symptoms, splenomegaly, and increased expression of T_{FH} markers. IDH2 $_{R172}$ mutation is associated with a histologically distinct subgroup, with medium to large clear cells and CXCL13 expression (24). Other mutations that are present are CD28, VAV1, PLCG1, STAT3, JAK2 (25).

2.2.2 Nodal TFH cell lymphoma, follicular helper type

FTCL predominantly has a follicular growth pattern and lacks the classical morphologic features of AITL. Morphologically, two distinct growth patterns are recognized: a follicular growth pattern that mimics B-cell follicular lymphoma (FL-like) and a progressive transformation of a germinal centers-like pattern (PTGC-like) In cases with the FL-like pattern, the neoplastic T-cells are arranged in well-defined nodules that lack the morphological features of normal follicles. In PTGC-like cases, the neoplastic T-cells are seen in small aggregates surrounded by small mantle zone B-cells arranged in large, irregular nodules. FTCL shares similar clinical features and mutational profiles with AITL and nodal TFH lymphoma, NOS. phenotype. A t(5;9) (q33;q22) ITK : SYK translocation has been reported in 15% of FTCL, which results in constitutive SYK kinase activation.

2.2.3 Nodal TFH lymphoma, NOS

Nodal TFH NOS phenotype show an infiltrative growth pattern and lack vascular proliferation, without sufficient pathological features to be diagnosed as AITL. Their mutational profile is similar to that of AITL. TET2, DNMT3A, IDH2R172, and RHOAG17V are frequently identified. Mutations in TCR signaling genes, such as PLCG1, PIK3R1, CARD11, CTNNB1, and KRAS have also been reported (23).

2.3 Anaplastic large cell lymphoma

The 5th edition of the WHO classification defines ALCL as mature T cell lymphomas characterized by sheets of pleomorphic, large horse-shoe shaped lymphoma cells, with a uniform strong expression of CD30 and often a lack of expression of T lineage markers. There are three distinct subtypes recognized in this classification – ALK negative ALCL, ALK positive ALCL and breast implant associated ALCL (26, 27).

2.3.1 Systemic ALCL

They have a nodal presentation and is classified into ALK+ and ALK-ve ALCL based on the presence of ALK protein detected by IHC (26). The high expression of CD30 has led the path to CD30 directed therapies like Brentuximab Vedotin with high response rates in this subtype (27).

ALK+ve ALCL is seen in younger patients and is associated with good prognosis if the IPI is low at presentation (28, 29). Presence of ALK by IHC is associated with an improved 5-year OS of 79% when compared to 46% for ALK negative patients (29, 30). ALK has several fusion partners, with NPM 1 on chromosome 5 being the most common one (31). ALK gene fusions lead to activation of multiple intracellular transduction pathways, notably the JAK-STAT3, NOTCH and PI3Kinase pathways (32). TP53 mutation is seen in 11% of ALK +ve cases and more frequently seen in younger patients (33). LRP1B gene mutation is reported but does not play a role in the prognosis. NOTCH1 is seen in 20% of patients and has been associated with increased cellular proliferation (34).

ALK negative ALCL nodal lymphomas have all the other morphological and phenotypical features of a CD30 positive cytotoxic ALCL, except ALK expression. They occur in an older age group and in general have a prognosis that is intermediate between ALK+ ALCL and other PTCL histology's. Emerging molecular data shows substantial heterogeneity, with several subgroups emerging (35). About 70% of Alk-ve ALCLs also express activated STAT3 through tyrosine kinase activity mediated by TYK or ROS1 and mutations of JAK1 and STAT 3 itself. Two recurrent and mutually exclusive rearrangements – DUSP22 and TP63 are seen in ALK-ve ALCL (18). DUSP22-R can be seen in 30% of the cases, is thought to have tumor suppressor function, has several distinct clinicopathological and molecular features (36). Morphologically, ALK negative DUSP22- R is characterized by monomorphic medium sized cell population with increased doughnut shaped cells, lack of JAK-STAT3 pathway activation, absence of PD-L1 expression, characteristic GEP signature (37). Although in the 2022 ICC classification, ALK-negative ALCL with DUSP22-R is considered as a subtype of ALK-negative ALCL, the WHO classification does not recognize DUSP22 -R ALK negative ALCL. (12) DUSP-22 R in general is considered to have favorable prognosis, with outcomes comparable to ALK+ve ALCL, however recent studies have also reported poor outcomes in patients with DUSP22-R (38). Fluorescence *in situ* hybridization (FISH) for DUSP22-R is recommend in ALK negative ALCL to identify the subtype for prognostic purposes. Testing for JAK2- rearrangements may help not only in identification but may also have therapeutic implications (39). However, further studies are needed before FISH can be used in routine clinical practice.

TP63 mutations are associated with a poor prognosis, with a 5 year survival of 17%. GEP studies have shown enrichment of IRF4 and MYC signature as well as proliferation of mTOR gene signatures in ALK-ve ALCL (35, 35). There is low expression of genes involved in TCR signaling and a high expression of *CD30* and its associated genes (*TNFRSF8, BATF3*, and *TMOD1*). There are altered responses to proapoptotic signals as well as Treg and TAM mediated immune dysregulation. PD-1 overexpression is also present in some DUSP22-R negative ALCL. This may provide further therapeutic opportunities.

2.3.2 Breast implant associated ALCL

Breast implant associated ALCL is typically a non-invasive neoplasm occurring in association with textured surface breast implants (40). It is an entity considered distinct from ALK-ve ALCL and is associated with an excellent prognosis. GEP show consistent absence of ALK, DUSP22 and TP63 rearrangements (41). Invasion of adjuvant structures is less common and associated with a poor prognosis. Studies have shown the pathogenesis to be mediated by the TH2 inflammatory cells, subsequent immune evasion. Certain bacterial pathogens such as *Ralstonia* have also been implicated (42). PD-L 1 overexpression is seen in over 50% of the cases. Somatic mutations of STAT3, STAT5B, JAK1 and JAK 2 resulting in constitutive activation of the JAK-STAT pathways, loss of function mutations of SOCS1 and SOCS have also been reported (43). Treatment consists of removal of the implant. Systemic therapy is reserved only for extracapsular spread (40).

2.4 EBV positive T and NK cell lymphoma

EBV positive T and NK cell lymphoma was previously classified under the PTCL-NOS. It is now recognized as a distinct entity in both the WHO classification and the 2022 ICC classification (12). It is seen more commonly among East Asians, with patients presenting with advanced lymphadenopathy with or without extra nodal involvement, B symptoms. EBV positive T/NK cell lymphoma is an aggressive disease, with a median survival of 1.5 months (44). Morphologically, diffuse infiltrate of uniform appearing medium to large, EBV+ve cells are noted. Absence of angioinvasion and necrosis are the characteristic features and are used for identification of this lymphoma. The lymphoma shows increased expression of PD-L1, demonstrates frequent mutations of TET2 and PI3 Kinase (45).

TABLE 2 Novel agents for treatment of PTCL and their targeted pathways.

Target	Novel agents
Epigenomic mutations	HDAC inhibitors Hypomethylating agents EZH2 inhibitors
Signaling pathways/Kinase directed therapies	PI3Kinase inhibitors Janus kinase pathway inhibitors SYK pathway inhibitors Aurora kinase inhibitors ALK inhibitors
Cell surface receptors	CD 30 inhibitors CD 25 directed therapies
Tumor microenvironment	Immunomodulatory agents Check point inhibitors Anti-CD 47 agents
Non cell signaling pathways inhibition	Antiapoptotic therapies Farnesyl transferase inhibitors

3 Therapeutic advances

Improved understanding of the tumor biology has not only led to more accurate diagnosis, risk stratification and prognosis, but has also identified critical pathways, which can potentially be altered to improve treatment outcomes. Novel agents targeting epigenomic mutations, cell cycle signaling pathways, tumor microenvironment and cell surface receptors are being used with promising results. Table 2 summarizes some of the potential novel agents and their targets that are being used and are under investigation. These will be discussed in detail below

3.1 Epigenetic mutations and targeted therapies

Changes in epigenetic signaling and DNA methylation have been described in T cell lymphoma (46). Epigenetic modifiers such as TET2, DNMT1, DNMT3A, 3B and TDH2 are implicated in T cell lymphoma pathogenesis. TET2 mutations result in cytosine demethylation and repress gene transcription (47). IDH2 is a specific isoform of the krebs cycle isocitrate dehydrogenase and the mutated enzyme catalyzes the conversion of alpha ketoglutarate (TET2 co-factor) to 2- hydroxyglutarate. Although IDH2 is not an epigenetic modifier, its alterations affect epigenetic modifications downstream on the TET2 pathway. TET2 mutations are loss of function mutations and have been implicated in the pathogenesis of AITL and T_{FH} derived PTCL (70-80% of cases), ALK- negative ALCL (33% of cases). DNTM3A mutations are seen in about 11-33% of patients with PTCL, the mutations frequently co-exist with TET2 mutations, resulting in suppression of transcription (47). These mutations are more commonly seen in AITL. Both TET2 and DNMT3A mutations occur during early hematopoietic stem cell maturation. IDH2 mutations are more commonly seen in AITL and their occurrence correlates with T_{FH} GEP signatures (24). EZH2 (enhancer of zeste homolog 2) has found to be mutated and overexpressed in several subtypes of PTCL (48). In ATLL, NF-kB plays a critical role in expression of EZH2.

Mutations in EZH2 result in transcriptional silencing, and inhibition of EZH2 in PTCL results in cell growth arrest *via* gene upregulation (48). KMT2D (mixed lineage leukemia 2) encodes histone methyltransferase, with gene mutations seen in 25% of patients with AITL and 36% in PTCL- NOS. Valemetostat is a dual inhibitor of EZH2 and EZH1 and is thought to increase the gene expression of pro-apoptotic and tumor suppressor genes by altering histone methylation. In a phase I study, 45 patients with rel/ref PTCL were treated with single agent valemetostat, with an ORR of 48%. The side effects observed were mainly thrombocytopenia (59%) and dysgeusia (51%) (49). A phase II study evaluating the efficacy of valemetostat in rel/ref PTCL is ongoing, with early results suggesting it to be an effective agent in rel/ref PTCL (50).

Histone deacetylate inhibitors (HDACI) such as romidepsin and belinostat are approved for relapsed PTCL (51–53). Given that the epigenomic dysfunction is seen to a greater extent in AITL and PTCL – FH subtype, the HDACS seem to have a higher impact in these subtypes. The ORR of romidepsin in AITL is 30% when

compared to an ORR of 25% across all subtypes of PTCL. Studies have shown a response rate of 45% in AITL (52). Hypomethylating agents such as 5- azacytidine and decitabine, inhibit DNMT and are being evaluated in AITL (54). A study by Delarue et al. evaluated the efficacy of 5-azacitidine in 19 patients with relapsed/refractory PTCL. Twelve patients had AITL (54). The ORR was 75% in patients with AITL when compared to 15% on other subtypes. It is to be noted that responding patients had a TET2 mutation. Real world data showed an ORR of 40% with 5-azacitdine among patients with heavily pretreated rel/ref AITL (55).

A study evaluating romidepsin and 5 azacytidine in 25 patients with relapsed/refractory PTCL had response rates of 61% and a CR of 48% (56). A retrospective study evaluating real-world data of 26 patients reported an ORR 73% and a CR 53%. Most common mutations found were TET2, RHOA, IDH2 and DNMT3A (57). A phase II study treated patients with 5-azacitidine and CHOP as first line therapy for PTCL (58). This study prioritized patients with TFH subtype of PTCL and enrolled 17 patients with TFH nodal lymphoma, 3 patients with PTCL-NOS and 1 patient with ATLL. Upon completion of treatment, the CR was 76.5% for all patients and 86.7% for patients with TFH nodal lymphoma. The 1-year PFS was also superior at 61% for the TFH lymphoma subtype when compared to 57% for all patients. NGS showed that TET2 mutations were associated with a significantly higher CR rate, favorable PFS and OS (58). The final efficacy data from this study is pending, but the preliminary data appears promising for consideration of such biomarker guided therapies in the front-line setting.

Patients with T_{FH} subtype had response rates of 80%, median progression free survival of 20.6 months. The responders also had a higher average number of DNMT mutations. IDH 2 mutations with substitution at R172 noted in TFH -AITL subtype could be a potential target. A phase Enasidenib, a novel IDH 2 inhibitor is being evaluated in patients with IDH-2 mutated TFH- AITL (NCT02273739) (59).TET- selective small molecular inhibitors (TETi 76) (60), IDH2 specific inhibitors for R172 codon noted in TFH- AITL are also being explored (61).

3.2 Signaling pathways

Dysregulated signaling pathways are being evaluated to find novel therapeutic targets. RHOA gene belongs to the Rho family of small GTPases, a group of Ras- like proteins involved in intracellular signaling (62). Gain of function mutations of the RHOA gene drive the T_{FH} differentiation, NFAT signaling activation, augment the PI3 kinase- AKT- mTOR signaling pathway, resulting in increased cell proliferation and transformation. Gain of function mutations are seen in up to 70% of AITL, 15% ATLL and are associated with cell proliferation. Agents targeting the kinase pathways could potentially be more efficacious in patients who have the RHOAG12V mutation. PI3K inhibitors such as duvelisib, multi-kinase inhibitors such as dasatinib have been proposed as therapeutic options (63). Current studies targeting these pathways are described below.

3.3 Kinase targeted therapies

3.3.1 PI3kinase inhibitors

PI3kinases are intracellular signaling molecules and are critical for growth and differentiation of lymphocytes. They occur in 4 isoforms – alpha, beta, gamma, delta (64). The gamma and delta subtypes are involved in adaptive immune response and are preferentially expressed in leucocytes. Targeting PI3K – gamma delta pathway has emerged as a promising treatment option for PTCL. Tenalisib and Duvelisib have been evaluated in patients with relapsed/refractory PTCL. A phase I study evaluated duvelisib (oral gamma delta specific PI3Kinase inhibitor) in heavily pretreated 16 patients with relapsed/refractory PTCL and showed an ORR of 50%, with an OS of 8.4 months (65). Grade 2 or higher toxicities included elevated liver function tests (35%), pyrexia (37%), cough (34%), cytopenia's (24%). This prompted a larger phase II dose finding/expansion study [PRIMO NCT03372057] (66). The preliminary data was consistent with the phase I study, with an ORR of 49%, CR 34% and a median duration of response of 7.7 months (67). Tenalisib, an oral dual PI3Kinase gamma/delta inhibitor was evaluated in 28 patients with heavily treated relapsed/refractory PTCL. Preliminary data shows a response rate of 45%, with a median duration of 4.1 months (68). The toxicity profile is similar to duvelisib. Several combinations with PI3Kinase inhibitors are also being evaluated. Duvelisib and romidepsin have been studied in combination with an ORR of 50%. This combination had lower rates of grade 3/4 transaminitis (8%) when compared to single agent duvelisib (40%), suggesting a potential immunomodulatory effect of romidepsin on PI3Kinase associated transaminitis (69).

Based on the preliminary data and the relatively safe toxicity profile, PI3 Kinase inhibitors are being studied in a combination with immune checkpoint blockade (copanlisib plus pembrolizumab, NCT02535247), with chemotherapy (copanlisib + gemcitabine, NCT03052933. Upfront studies, combining chemotherapy with PI3K inhibitors are ongoing (A051902).

a) Janus Kinase Pathway

The Janus kinase family proteins (JAK1, JAK2, JAK3, TYK2) play a role in mediating immune responses. Downstream to the JAK proteins are STAT family of transcription factors, which regulate the effects of JAK. Activating mutations and gene fusion resulting in activated JAK-STAT pathway signaling have been reported in several subtypes of PTCL. A phase II study of ruxolitinib in 53 patients with relapsed/refractory PTCL reported an ORR of 29% among patients with activating JAK/STAT mutations (70). Response rates were higher in patients with PTCL-T_{FH} subtype and in AITL – 44%, large granular leukemia 75%. The commonly reported grade 3 or higher side effects were cytopenia's. Golidoctinib, is an oral JAK1 specific inhibitor, is being evaluated in patients with rel/ref PTCL. A phase I/II study (NCT04105010), study enrolled 51 patients with rel/ref PTCL and reports a response rate of 43%. The drug was well tolerated, with grade 3 or higher neutropenia was seen in 29% of the patients, thrombocytopenia seen in 10% (Kim, Yoon et al. 2022).

b) SYK pathway

SYK encodes a tyrosine kinase protein that regulates PI3K downstream signaling pathway (71). It is normally expressed on the B cells, but not on the T cells. Aberrant expression of SYK is noted in about 90% of the T cells. Cedulatinib, a JAK/SYK inhibitor was evaluated in 60 patients with PTCL, with an ORR of 35% (72). The response rates in patients with PTCL-T_{FH} type and AITL was 55%. Most common adverse events included elevated lipase, diarrhea, cytopenia's. TAK-659, a novel SKY inhibitor is currently in development, with preclinical studies showing promising results in non-Hodgkin lymphomas. [NCT-3357626, NCT02000934, NCT03742258]

c) Aurora A kinase pathway

Aurora Kinase is a protein kinase that mediates spindle formation during mitosis and is overexpressed in PTCL. Alisertib is an oral aurora kinase A inhibitor, which showed preclinical activity in PTCL (73). A phase III randomized study of alisertib in rel/ref PTCL showed an ORR 33% (74). However, there was lack of benefit in progression free survival when compared to the control arm and this trial was terminated early.

d) Anaplastic Lymphoma Kinase

As ALCL is divided based on the presence of translocation that fuses the anaplastic lymphoma kinase (ALK) gene to NPM, ALK inhibitors have been evaluated in patients with ALK + ALCL. Crizotinib, an ALK inhibitor is FDA approved for treatment of pediatric patients with rel/ref ALK+ALCL (75). The approval was based in a single arm trial of crizotinib in 26 patients, aged <20 years with relapsed/refractory ALCL. The ORR was 88%. A phase I study evaluated 9 adult patients with heavily pretreated rel/ref ALK+ ALCL (76). All patients experienced complete responses to therapy. There is an ongoing study evaluating ceritinib, a second generation ALK inhibitor, with promising preliminary data (77). Alectinib, a second generation ALK inhibitor has also been evaluated, with promising results. Crizotinib is being evaluated in combination with cytotoxic chemotherapy in patients as a part of front-line therapy in ALK+ALCL [NCT01979536]. *In vitro* studies have shown synergistic activity with crizotinib and brentuximab vedotin (78) and clinical studies are being designed evaluating this combination.

4 Specific targeted therapies

4.1 CD30 directed therapy

The tumor necrosis family receptor CD30 is universally expressed in ALCL irrespective of the ALK status. It is variably expressed in other subtypes and can be present in 58-64% of PTCL, 43-63% AITL, 55% ATLL. Brentuximab vedotin (BV) is an antibody drug conjugate, composed of an antiCD30 monoclonal antibody linked to monomethyl auristatin E, a microtubule inhibitor (79). BV was first evaluated in patients with rel/ref ALCL, with an ORR of 86% (27). BV also evaluated in patients CD30+ PTCL, with an ORR of 41% and a median PFS of 6.7 months (80). Based on the encouraging results from these studies, ECHELON -2, a randomized double blinded study randomized 226 patients with newly diagnosed PTLD, with at least 10% CD30 expression to receive either BV in combination with cyclophosphamide, vincristine and prednisone (A+CHP) or cyclophosphamide, vincristine, prednisone, doxorubicin (CHOP) (5). Combining BV to chemotherapy proved superior to chemotherapy alone, with a median PFS of 48 months v.s 21 months of CHOP. The hazard ratio was 0.71 indicating a 29% reduction in the risk of relapse. Five-year follow up shows the durability of the responses, with a PFS of 51% with the A+ CHP arm when compared to 43% with the CHOP arm (81). The OS was 70% with the A+CHP arm and 61% with the CHOP arm. The benefit in PFS and OS was seen across all subtypes of PTCL. This is a pivotal study that led to the approval of A+ CHP as front-line therapy for CD30+ PTCL. Other combinations are being evaluated including the combination of BV with CHEP s upfront therapy and other agents like gemcitabine (ASH abstract).

4.2 CCR4

CC chemokine receptor 4 (CCR4) promotes memory T cell homing to peripheral tissues and is preferentially expressed by Th2 and regulatory T cell subsets. CCR4 is predominantly expressed in ATLL. CCR4 is also expressed in about 30–65% of other PTCL cells, including a high expression (65%) in ALK -ve ALCL, variable expression (40%) in PTCL- NOS, AITL and transformed MF (82). Mogamulizumab is a humanized, IgG1 monoclonal anti CCR4 antibody. It has a direct cytotoxic effect on CCR-4 positive lymphoma cells *via* ADCC and depletes the regulatory T cell, resulting in an antitumor activity. It is approved for CCR4+ rel/ref ATLL in Japan based on a phase 2 study that reported a response rate of 50%, with median PFS and OS of 5.2 and 13.7 months (83). In the US, it is approved for patients with cutaneous T cell lymphoma. The main side effects are skin reactions seen in up to 63% of patients, infusion reactions and hematologic toxicity.

4.3 CD25 directed therapies

T-cell activation (CD 25) is the alpha subunit of the heterotrimeric IL-2 receptor. CD25 is seen in upto 40-50% of all PTCL's (84). As it is differentially expressed on malignant T cells, this has been evaluated for a potential target. Antibody drug conjugates towards CD25 have shown promising results. Camidanlumab tesitine, a CD 25 antibody conjugated to cytotoxic pyrrolobenzodiazepine dimer has shown promising results in relapsed PTCL. Responses correlate with CD25 expression (85). Camidanlumab tesitine is currently accruing patients in phase II study, with promising responses seen in patients with hodgkin lymphoma. Radiolabeled antibodies such as Y^{90} labeled basiliximab are being evaluated in combination with cytotoxic chemotherapy in PTCL directed transplant regimens (86).

5 Altering the tumor microenvironment

A three-signal pathway is thought to contribute to T cell lymphomagenesis – (i) antigenic stimulation through TCR engagement – signal 1 (ii) co-stimulatory signals from non- MHC

surface receptors -signal 2, (iii) cytokine signals through paracrine stimulation – signal 3 (14). Both type 2 and type 3 signals are mediated by non-neoplastic T cells, highlighting the role the tumor microenvironment plays in this disease (14). The tumor cells cause modulation of the microenvironment and decrease the tumor immunogenicity and in doing this, promote their survival (14). The neoplastic T cells produce several soluble cytokines such as IL-5, IL-13 and VEGF and other immune regulatory molecules. The IL-13 recruit eosinophilic granulocytes from the blood stream. These eosinophils secrete high amounts of IL-10, IL-4, which sustain macrophage differentiation into a tumor favoring subtype. These differentiated macrophages express PD-L1 and produce several angiogenetic and immune-modulatory soluble mediators which facilitate tumor immune escape from antitumor cytotoxic T cells. PTCL also recruits non-neoplastic Tregs lymphocytes which dampen the antitumor immune responses (87).

Therefore, targeting the tumor microenvironment may provide a therapeutic opportunity.

5.1 Immunomodulatory agents

Lenalidomide acts by modulation of substrate specificity of $CRL4^{CRBN}$ ubiquitin ligase complex and in doing so, induces degradation of protein targets that mediate immune responses such as interleukin production and NK/T cell proliferation (88). By doing so, lenalidomide decreases cell proliferation and enhances NK cell cytotoxicity. Lenalidomide has being investigated in PTCL, both as a single agent and in combination therapy. A phase 1/2 study reported an ORR of 22% in 54 patients with Rel/Ref PTCL (89). The median PFS was 2.5 months. Response rates were comparable across all subgroups. A multicenter trial (EXPECT) reported a similar response rate of 22% with 11% CR, mostly in patients with AITL (90). While single agent activity is low, lenalidomide is being studied in combination regimens in T–cell Lymphomas. The combination of CHOEP and lenalidomide as an upfront treatment resulted in an ORR of 48% in a phase 2 study in 40 patients; however, there was significant toxicity (91). Romidepsin and lenalidomide has been combined safely; the phase 1/2 study of 21 patients produced a response rate of 53% with acceptable hematologic toxicity (92). The combination is now being evaluated in an upfront study of elderly patients with PTCL. The combination of Romidepsin, lenalidomide and carfilzomib resulted in an ORR of 50% in a phase 1/2 trial of 20 patients (93). Lenalidomide is being studied in combination with BV and with immune check point inhibitors such as anti-PD1 antibody durvalumab (NCT03011814) in cutaneous T cell lymphomas.

5.2 Checkpoint inhibitors

Use of checkpoint inhibitors in T cell malignancies poses concerns as although there is an anti-tumor effect on the cytotoxic T cells, there could also be an unintended blocking of PD-1 driven tumor suppression maintained by the PD-L1 expressed on the antigen presenting cells. There have been reports of rapid progression in patients with ATLL when treated with nivolumab (94). PD-1 inhibitors have been used in ENKL, with response rates of 100% with pembrolizumab. The EBV viral proteins upregulate the PD-1 receptor levels, which is thought to be the reason for superior response rates with PD-1 inhibitors when compared to.

Pembrolizumab has been used in patients with relapsed/refractory cutaneous lymphomas, with response rates of 38% (95). However, these responses have been short-lived. The use of checkpoint inhibitors in PTCL is currently limited to clinical trials.

5.3 Anti CD47 antibodies

CD47 is a cell surface regulator of phagocytosis and is mediated by macrophages and dendritic cells. Increased expression of CD47 results in an inhibitory signal for phagocytosis. Inhibition of CD47 signaling can promote macrophage phagocytosis of tumor cells (96). Anti CD47 directed antibodies are being used in PTCL, with promising results.

6 Cellular therapy

Chimeric antigen receptor T cells, targeting CD19 have significantly improved outcomes in B cell lymphomas. CAR-T therapy has since been expanded into treatment of other hematological malignancies. Studies have been ongoing to determine if CAR-T cells could be used to treat T cell lymphomas.

Designing CAR-T cells to target T cell neoplasms has been challenging due to the following complications. CAR-T fratricide, due to endogenous expression of T cell antigens that CAR-T cells are designed against, limits T cell expansion (97). Preclinical studies are ongoing, investigating manipulating the CAR antigen target to bypass the fratricide. T cell aplasia is a potential complication. Unlike B cell aplasia that is encountered with CD 19 CAR's, T cell aplasia has a risk of permanent immunosuppression, resulting in life threatening infections. CAR-T cells targeting CD 30 are being developed, with pre-clinical studies showing good cell expansion and cytotoxic activity against PTCL xenograft tumors (98). CAR T cells targeting CD4, CD5, CD7 are also being developed. Allogenic T cells and use of alternate cell sources such as CAR-macrophages are also in early phase clinical trials.

7 Non- cell signaling Kinase/non epigenetic therapies

7.1 Anti-apoptotic therapies

Apoptosis is regulated by BCL2 family of proteins, which function by counter balancing pro and antiapoptotic members, MCL1 and BCL2. High levels of MCL 1 expression have been described in PTCL and inhibition of this pathway is associated with reduced cell survival (99). Koch et al. described MCL1 dependent PTCL xenograft model where MCL 1 inhibition was synergistic with multiagent chemotherapy and improved survival (100). Based on this data,

several studies, with agents targeting MCL1 and BCL2 in patients with rel/ref PTCL are ongoing (PRT1419 -NCT04543305; AMG 397 – NCT03465540, venetoclax – NCT03534180).

7.2 Farnesyltransferase inhibitors

CXCL12 expression has shown to have a prognostic significance in patients with PTCL. Tipifarnib, a farnesyltransferase/CXCR4 inhibitor has shown to downregulate CXCL12 secretion in stromal cultures (101). CXCL12 can be expressed in up to 50% of AITL. This drug is being selectively investigated in patients with CXCL 12+ PTCL- NOS or AITL. A phase 2 study evaluated the role of tipifanib heavily pretreated patients with AITL or PTCL- NOS (102). In the AITL cohort, there were 11 evaluable patients with an OR of 45%. Among the 3 evaluable CXCL12+ PTCL patients, one patient had a PR and 2 patients had SD to therapy. CXCL 12 expression was found to correlate with favorable outcome to treatment (102). Pre-treatment high CXCL12 expression had 90% sensitivity and 93% specificity to accurately predict a treatment response. The primary toxicities were thrombocytopenia and neutropenia.

8 Conclusions

How best to incorporate these novel agents into the existing treatment regimens is both exciting and challenging. Given the relatively low and short-lived response rates with single agent therapy, combination therapies are being explored. For frontline therapy, combination chemotherapy with addition of BV in CD30+ patients have become the current standard of care. Consolidation autologous stem cell transplant is recommended for eligible patients with PTCL, other than ALK+ve ALCL. Treatments combining novel agents such as romidepsin/azacytidine with cytotoxic chemotherapy are being explored. In the rel/ref setting, trials combining novel agents such as copanlisib+ romidepsin, durvalumab+ lenalidomide are being evaluated.

Clinical trials incorporating GEP to tailor treatments are warranted. An umbrella study in Japan is evaluating the efficacy and safety of targeted therapies guided by molecular subtypes in patients with rel/ref PTCL [NCT05559008]. This study is incorporating next generation sequencing to identify the molecular subtypes and interventions are being modified accordingly. The results of these study could potentially be practice changing. More such studies incorporating the molecular advances are warranted. The future for PTCL is promising. Recent advances in molecular and genomic profiling of PTCL has provided a unique insight into the molecular pathways and in the development of new therapeutic agents. Further studies are needed to tailor the treatments based on genomic markers in both front line and relapsed settings. Development of markers to define response to these agents and to predict patterns of treatment failure will help advance the field immensely.

Author contributions

JZ contributed to manuscript preparation, editing, submission. AK contributed to manuscript preparation. All authors contributed to the article and approved the submitted version.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Swerdlow SH. Who classification of tumours of haematopoietic and lymphoid tissues. *Int Agency For Res On Cancer* (2017).

2. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBDO, Berti E, et al. The 5th edition of the world health organization classification of hematolymphoid tumours: Lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

3. Armitage J, Vose J, Weisenburger D. Towards understanding the peripheral T-cell lymphomas. *Ann Of Oncol* (2004) 15(10):1447–9. doi: 10.1093/annonc/mdh409

4. O'connor OA, Pro B, Pinter-Brown L, Bartlett N, Popplewell L, Coiffier B, et al. Pralatrexate in patients with relapsed or refractory peripheral T-cell lymphoma: Results from the pivotal propel study. *J Of Clin Oncol Off J Of Am Soc Of Clin Oncol* (2011) 29 (9):1182–9. doi: 10.1200/JCO.2010.29.9024

5. Horwitz S, O'connor OA, Pro B, Illidge T, Fanale M, Advani R, et al. Brentuximab vedotin with chemotherapy for Cd30-positive peripheral T-cell lymphoma (Echelon-2): A global, double-blind, randomised, phase 3 trial. *Lancet* (2019) 393(10168):229–40. doi: 10.1016/S0140-6736(18)32984-2

6. Piekarz RL, Frye R, Prince HM, Kirschbaum MH, Zain J, Allen SL, et al. Phase 2 trial of romidepsin in patients with peripheral T-cell lymphoma. *Blood* (2011) 117 (22):5827–34. doi: 10.1182/blood-2010-10-312603

7. Foss F, Advani R, Duvic M, Hymes KB, Intragumtornchai T, Lekhakula A, et al. A phase ii trial of belinostat (Pxd 101) in patients with relapsed or refractory peripheral or cutaneous T-cell lymphoma. *Br J Of Haematology* (2015) 168(6):811–9. doi: 10.1111/bjh.13222

8. Chihara D, Fanale MA, Miranda RN, Noorani M, Westin JR, Nastoupil LJ, et al. Survival outcome of patients with relapsed/refractory peripheral T-cell lymphoma-not otherwise specified and angioimmunoblastic T-cell lymphoma. *Br J Haematol* (2017) 176:750–8. doi: 10.1111/bjh.14477

9. Vose J. International T-cell lymphoma project. international peripheral T-cell and natural Killer/T-cell lymphoma study: Pathology findings and clinical outcomes. *J Clin Oncck* (2008) 26:4124–30. doi: 10.1200/JCO.2008.16.4558

10. Wang SS, Vose JM. Epidemiology and prognosis of T-cell lymphoma. *T-Cell Lymphomas. Springer* (2013), 25–39. doi: 10.1007/978-1-62703-170-7_2

11. Iqbal J, Liu Z, Deffenbacher K, Chan WC. Gene expression profiling in lymphoma diagnosis and management. *Best Pract Res Clin Haematology* (2009) 22 (2):191–210. doi: 10.1016/j.beha.2009.05.001

12. Feldman AL, Laurent C, Narbaitz M, Nakamura S, Chan WC, De Leval L, et al. Classification and diagnostic evaluation of nodal T-and nk-cell lymphomas. *Virchows Archiv* (2022) 482(1):265–79. doi: 10.1007/s00428-022-03412-6

13. Wilcox RA. A three-signal model of T-cell lymphoma pathogenesis. *Am J Of Hematol* (2016) 91(1):113–22. doi: 10.1002/ajh.24203

14. Pizzi M, Margolskee E, Inghirami G. Pathogenesis of peripheral T cell lymphoma. *Annu Rev Of Pathology: Mech Of Dis* (2018) 13:293–320. doi: 10.1146/annurev-pathol-020117-043821

15. Fiore D, Cappelli LV, Broccoli A, Zinzani PL, Chan WC, Inghirami G. Peripheral T cell lymphomas: From the bench to the clinic. *Nat Rev Cancer* (2020) 20(6):323–42. doi: 10.1038/s41568-020-0247-0

16. Broccoli A, Zinzani PL. Peripheral T-cell lymphoma, not otherwise specified. *Blood J Of Am Soc Of Hematol* (2017) 129(9):1103–12. doi: 10.1182/blood-2016-08-692566

17. Karube K, Kakimoto Y, Tonozuka YO, Ohshima K. The expression of Cd30 and its clinic-pathologic significance in peripheral T cell lymphoma. *Expert Rev Of Hematol* (2021) 14(8):777–87. doi: 10.1080/17474086.2021.1955344

18. Heavican TB, Bouska A, Yu J, Lone W, Amador C, Gong Q, et al. Genetic drivers of oncogenic pathways in molecular subgroups of peripheral T-cell lymphoma. *Blood J Of Am Soc Of Hematol* (2019) 133(15):1664–76. doi: 10.1182/blood-2018-09-872549

19. Amador C, Greiner TC, Heavican TB, Smith LM, Galvis KT, Lone W, et al. Reproducing the molecular subclassification of peripheral T-cell lymphoma–nos by immunohistochemistry. *Blood* (2019) 134(24):2159–70. doi: 10.1182/blood.2019000779

20. Piccaluga PP, Fuligni F, De Leo A, Bertuzzi C, Rossi M, Bacci F, et al. Molecular profiling improves classification and prognostication of nodal peripheral T-cell lymphomas: Results of a phase iii diagnostic accuracy study. *J Of Clin Oncol Off J Of Am Soc Of Clin Oncol* (2013) 31(24):3019–25. doi: 10.1200/JCO.2012.42.5611

21. Laginestra MA, Cascione L, Motta G, Fuligni F, Agostinelli C, Rossi M, et al. Whole exome sequencing reveals mutations in Fat1 tumor suppressor gene clinically impacting on peripheral T-cell lymphoma not otherwise specified. *Modern Pathol* (2020) 33(2):179–87. doi: 10.1038/s41379-019-0279-8

22. Dobay MP, Lemonnier F, Missiaglia E, Bastard C, Vallois D, Jais J, et al. Integrative clinicopathological and molecular analyses of angioimmunoblastic T-cell lymphoma and other nodal lymphomas of follicular helper T-cell origin. *Haematologica* (2017) 102(4):E148–51. doi: 10.3324/haematol.2016.158428

23. Lemonnier F, Couronné L, Parrens M, Jaïs J, Travert M, Lamant L, et al. Recurrent Tet2 mutations in peripheral T-cell lymphomas correlate with tfh-like features and adverse clinical parameters. *Blood J Of Am Soc Of Hematol* (2012) 120 (7):1466–9. doi: 10.1182/blood-2012-02-408542

24. Wang C, Mckeithan TW, Gong Q, Zhang W, Bouska A, Rosenwald A, et al. Idh2 R172 mutations define a unique subgroup of patients with angioimmunoblastic T-cell lymphoma. *Blood J Of Am Soc Of Hematol* (2015) 126(15):1741–52. doi: 10.1182/blood-2015-05-644591

25. Willemsen M, Hamid A, M. W, B. And Zur Hausen A. Mutational heterogeneity of angioimmunoblastic T-cell lymphoma indicates distinct lymphomagenic pathways. *Blood Cancer J* (2018) 8(1):1–4. doi: 10.1038/s41408-017-0047-2

26. Hafeez S, Cushman-Vokoun AM. Molecular advances in nodal peripheral T-cell lymphoma. *Adv In Mol Pathol* (2022) 5(1):51–8. doi: 10.1016/j.yamp.2022.05.002

27. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, et al. Brentuximab vedotin (Sgn-35) in patients with relapsed or refractory systemic anaplastic Large-cell lymphoma: Results of a phase ii study. *J Of Clin Oncol Off J Of Am Soc Of Clin Oncol* (2012) 30(18):2190–6. doi: 10.1200/JCO.2011.38.0402

28. Gascoyne RD, Aoun P, Wu D, Chhanabhai M, Skinnider BF, Greiner TC, et al. Prognostic significance of anaplastic lymphoma kinase (Alk) protein expression in adults with anaplastic Large cell lymphoma. *Blood J Of Am Soc Of Hematol* (1999) 93(11):3913–21.

29. Maurer MJ, Ellin F, Srour L, Jerkeman M, Bennani NN, Connors JM, et al. International assessment of event-free survival At 24 months and subsequent survival in peripheral T-cell lymphoma. *J Of Clin Oncol Off J Of Am Soc Of Clin Oncol* (2017) 35 (36):4019–26. doi: 10.1200/JCO.2017.73.8195

30. Savage KJ, Harris NL, Vose JM, Ullrich F, Jaffe ES, Connors JM, et al. Alk–anaplastic Large-cell lymphoma is clinically and immunophenotypically different from both alk alcl and peripheral T-cell lymphoma, not otherwise specified: Report from the international peripheral T-cell lymphoma project. *Blood J Of Am Soc Of Hematol* (2008) 111(12):5496–504. doi: 10.1182/blood-2008-01-134270

31. Werner MT, Zhao C, Zhang Q, Wasik MA. Nucleophosmin-anaplastic lymphoma kinase: The ultimate oncogene and therapeutic target. *Blood J Of Am Soc Of Hematol* (2017) 129(7):823–31. doi: 10.1182/blood-2016-05-717793

32. Morris SW, Kirstein MN, Valentine MB, Dittmer KG, Shapiro DN, Saltman DL, et al. Fusion of a kinase gene, alk, to a nucleolar protein gene, npm, in non-hodgkin's lymphoma. *Sci (New York N.Y.)* (1994) 263(5151):1281–4. doi: 10.1126/science.8122112

33. Lobello C, Tichy B, Bystry V, Radova L, Filip D, Mraz M, et al. Stat3 and Tp53 mutations associate with poor prognosis in anaplastic Large cell lymphoma. *Leukemia* (2021) 35(5):1500–5. doi: 10.1038/s41375-020-01093-1

34. Larose H, Prokoph N, Matthews JD, Schlederer M, Hogler S, Alsulami AF, et al. Whole exome sequencing reveals Notch1 mutations in anaplastic Large cell lymphoma and points to notch both as a key pathway and a potential therapeutic target. *Haematologica* (2021) 106(6):1693–704. doi: 10.3324/haematol.2019.238766

35. Parrilla Castellar ER, Jaffe ES, Said JW, Swerdlow SH, Ketterling RP, Knudson RA, et al. Alk-negative anaplastic Large cell lymphoma is a genetically heterogeneous disease with widely disparate clinical outcomes. *Blood J Of Am Soc Of Hematol* (2014) 124(9):1473–80. doi: 10.1182/blood-2014-04-571091

36. Luchtel RA, Dasari S, Oishi N, Pedersen MB, Hu G, Rech KL, et al. Molecular profiling reveals immunogenic cues in anaplastic Large cell lymphomas with Dusp22 rearrangements. *Blood J Of Am Soc Of Hematol* (2018) 132(13):1386–98. doi: 10.1182/blood-2018-03-838524

37. King RL, Dao LN, Mcphail ED, Jaffe ES, Said J, Swerdlow SH, et al. Morphologic features of alk-negative anaplastic Large cell lymphomas with Dusp22 rearrangements. *Am J Of Surg Pathol* (2016) 40(1):36–43. doi: 10.1097/PAS.0000000000000500

38. Hapgood G, Ben-Neriah S, Mottok A, Lee DG, Robert K, Villa D, et al. Identification of high-risk Dusp22-rearranged alk-negative anaplastic Large cell lymphoma. *Br J Of Haematology* (2019) 186(3):E28–31. doi: 10.1111/bjh.15860

39. Fitzpatrick MJ, Massoth LR, Marcus C, Vergilio J, Severson E, Duncan D, et al. Jak2 rearrangements are a recurrent alteration in Cd30 systemic T-cell lymphomas with anaplastic morphology. *Am J Of Surg Pathol* (2021) 45(7):895–904. doi: 10.1097/PAS.0000000000001708

40. Miranda RN, Aladily TN, Prince HM, Kanagal-Shamanna R, De Jong D, Fayad LE, et al. Breast implant-associated anaplastic Large-cell lymphoma: Long-term follow-up of 60 patients. *J Of Clin Oncol Off J Of Am Soc Of Clin Oncol* (2014) 32(2):114–20. doi: 10.1200/JCO.2013.52.7911

41. Oishi N, Brody GS, Ketterling RP, Viswanatha DS, He R, Dasari S, et al. Genetic subtyping of breast implant–associated anaplastic Large cell lymphoma. *Blood J Of Am Soc Of Hematol* (2018) 132(5):544–7. doi: 10.1182/blood-2017-12-821868

42. Hu H, Johani K, Almatroudi A, Vickery K, Van Natta B, Kadin ME, et al. Bacterial biofilm infection detected in breast implant–associated anaplastic Large-cell lymphoma. *Plast And Reconstructive Surg* (2016) 137(6):1659–69. doi: 10.1097/PRS.0000000000002010

43. Di Napoli A, Jain P, Duranti E, Margolskee E, Arancio W, Facchetti F, et al. Targeted next generation sequencing of breast implant-associated anaplastic Large cell lymphoma reveals mutations in Jak/Stat signalling pathway genes, Tp53 and Dnmt3a. *Br J Of Haematology* (2018) 180(5):741–4. doi: 10.1111/bjh.14431

44. Jung KS, Cho S, Kim SJ, Ko YH, Kim WS. Clinical features and treatment outcome of Epstein–Barr virus-positive nodal T-cell lymphoma. *Int J Of Hematol* (2016) 104(5):591–5. doi: 10.1007/s12185-016-2068-1

45. Wai CMM, Chen S, Fan S, Leong SM, Zheng W, Low LCY, et al. Immune pathway upregulation and lower genomic instability distinguish ebv-positive nodal T/Nk-cell lymphoma from enktl and ptcl-nos. *Haematologica* (2022) 107(8):1864–79. doi: 10.3324/haematol.2021.280003

46. Epstein-Peterson ZD, Horwitz SM. Molecularly targeted therapies for relapsed and refractory peripheral T-cell lymphomas. In: *Seminars in hematology*. Elsevier (2021). p. 78–84.

47. Couronné L, Bastard C, Bernard OA. Tet2 and Dnmt3a mutations in human T-cell lymphoma. *New Engl J Of Med* (2012) 366(1):95–6. doi: 10.1056/NEJMc1111708

48. Dhiran K, Libien J, Brunson C, Jain P, Gupta R. Ezh2 expression is increased in neoplastic T cells as compared with reactive lymphocytes and differs in different subtypes of T-cell lymphoma. *Am J Of Clin Pathol* (2012) 138(Suppl_1):A161. doi: 10.1093/ajcp/138.suppl1.147

49. Ishitsuka K, Izutsu K, Maruyama D, Makita S, Jacobsen E, Horwitz S, et al. First-In-Human study of the Ezh1 and Ezh2 dual inhibitor valemetostat tosylate (Ds-3201b) in patients with relapsed or refractory non-Hodgkin lymphomas. *Hematological Oncol* (2021) 39. doi: 10.1002/hon.14_2879

50. Foss FM, Porcu P, Horwitz SM, Izutsu K, Ishitsuka K, Kato K, et al. A global phase 2 study of valemetostat tosylate (Valemetostat) in patients with relapsed or refractory (R/R) peripheral T-cell lymphoma (Ptcl), including R/R adult T-cell Leukemia/Lymphoma (Atl)-Valentine-Ptcl01. *Blood* (2021) 138:2533. doi: 10.1182/blood-2021-144676

51. Park J, Jong H, Kim SG, Jung Y, Lee K, Lee J, et al. Inhibitors of histone deacetylases induce tumor-selective cytotoxicity through modulating aurora-a kinase. *J Of Mol Med* (2008) 86(1):117–28. doi: 10.1007/s00109-007-0260-8

52. Coiffier B, Pro B, Prince HM, Foss F, Sokol L, Greenwood M, et al. Results from a pivotal, open-label, phase ii study of romidepsin in relapsed or refractory peripheral T-cell lymphoma after prior systemic therapy. *J Of Clin Oncol* (2012) 30(6):631–6. doi: 10.1200/JCO.2011.37.4223

53. O'connor OA, Horwitz S, Masszi T, Van Hoof A, Brown P, Doorduijn J, et al. Belinostat in patients with relapsed or refractory peripheral T-cell lymphoma: Results of the pivotal phase ii belief (Cln-19) study. *J Of Clin Oncol Off J Of Am Soc Of Clin Oncol* (2015) 33(23):2492–9. doi: 10.1200/JCO.2014.59.2782

54. Delarue R, Dupuis J, Sujobert P, Barbieux S, Marçais A, Tournilhac O, et al. Treatment with hypomethylating agent 5-azacytidine induces sustained response in angioimmunoblastic T cell lymphomas. *Blood* (2016) 128(22):4164. doi: 10.1182/blood.V128.22.4164.4164

55. Yoon SE, Cho J, Kim YJ, Kim SJ, Kim WS. Real-world efficacy of 5-azacytidine as salvage chemotherapy for angioimmunoblastic T-cell lymphoma. *Clin Lymphoma*

Myeloma And Leukemia (2022) 22(11):E972–80. Invalid Date. doi: 10.1016/j.clml.2022.07.009

56. Horwitz SM, Moskowitz AJ, Jacobsen ED, Mehta-Shah N, Khodadoust MS, Fisher DC, et al. The combination of duvelisib, a Pi3k-Δ, Γ inhibitor, and romidepsin is highly active in Relapsed/Refractory peripheral T-cell lymphoma with low rates of transaminitis: Results of parallel multicenter, phase 1 combination studies with expansion cohorts. *Blood* (2018) 132:683. doi: 10.1182/blood-2018-99-115241

57. Kalac M, Jain S, Tam CS, Goldfinger M, Xiao Z, Montanari F, et al. A real world experience of combined treatment with romidepsin and azacitidine in patients with peripheral T-cell lymphoma-a bridge to transplant and an effective salvage for the unfit. *Blood* (2022) 140(Supplement 1):9422–4. doi: 10.1182/blood-2022-165165

58. Ruan J, Moskowitz AJ, Mehta-Shah N, Sokol L, Chen Z, Rahim R, et al. Multicenter phase ii study of oral azacitidine (Cc-486) plus chop as initial treatment for peripheral T-cell lymphoma (Ptcl). *Blood* (2020) 136:33–4. doi: 10.1182/blood-2020-136023

59. Yamagishi M, Hori M, Fujikawa D, Ohsugi T, Honma D, Adachi N, et al. Targeting excessive Ezh1 and Ezh2 activities for abnormal histone methylation and transcription network in malignant lymphomas. *Cell Rep* (2019) 29(8):2321–37,e7. doi: 10.1016/j.celrep.2019.10.083

60. Hu L, Zhang X, Li H, Lin S, Zang S. Targeting Tet2 as a therapeutic approach for angioimmunoblastic T cell lymphoma. *Cancers* (2022) 14(22):5699. doi: 10.3390/cancers14225699

61. Guo J, Zhang R, Yang Z, Duan Z, Yin D, Zhou Y. Biological roles and therapeutic applications of Idh2 mutations in human cancer. *Front In Oncol* (2021) 11:644857. doi: 10.3389/fonc.2021.644857

62. Sakata-Yanagimoto M. Multistep tumorigenesis in peripheral T cell lymphoma. *Int J Of Hematol* (2015) 102(5):523–7. doi: 10.1007/s12185-015-1738-8

63. Fujisawa M, Sakata-Yanagimoto M, Nishizawa S, Komori D, Gershon P, Kiryu M, et al. Activation of rhoa–Vav1 signaling in angioimmunoblastic T-cell lymphoma. *Leukemia* (2018) 32(3):694–702. doi: 10.1038/leu.2017.273

64. Huang D, Song TL, Nairismägi M, Laurensia Y, Pang W, Zhe DCM, et al. Evaluation of the Pik3 pathway in peripheral T-cell lymphoma and Nk/T-cell lymphoma. *Br J Of Haematology* (2020) 189(4):731–44. doi: 10.1111/bjh.16435

65. Horwitz SM, Koch R, Porcu P, Oki Y, Moskowitz A, Perez M, et al. Activity of the Pi3k-Δ, Γ inhibitor duvelisib in a phase 1 trial and preclinical models of T-cell lymphoma. *Blood J Of Am Soc Of Hematol* (2018) 131(8):888–98.

66. Pro B, Brammer JE, Casulo C, Jacobsen E, Mead M, Mehta-Shah N, et al. Duvelisib in patients with Relapsed/Refractory peripheral T-cell lymphoma from the phase 2 primo trial: Dose optimization efficacy update and expansion phase initial results. *Blood* (2020) 136:38–9. doi: 10.1182/blood-2020-140412

67. Jacobsen ED, Zinzani PL, Zain J, Mead M, Casulo C, Gritti G, et al. Duvelisib in patients with Relapsed/Refractory peripheral T-cell lymphoma from the phase 2 primo trial expansion phase: Impact of prior treatment and expanded safety analysis. *Blood 140(Supplement 1)* (2022), 9387–9. doi: 10.1182/blood-2022-169432

68. Huen A, Haverkos BM, Zain J, Radhakrishnan R, Lechowicz MJ, Devata S, et al. Phase I/Ib study of tenalisib (Rp6530), a dual Pi3k Δ/Γ inhibitor in patients with Relapsed/Refractory T-cell lymphoma. *Cancers* (2020) 12(8):2293. doi: 10.3390/cancers12082293

69. Horwitz S, Moskowitz A, Mehta-Shah N, Jacobsen E, Khodadoust M, Ganesan N, et al. The combination of duvelisib and romidepsin (Dr) is highly active against Relapsed/Refractory peripheral T-cell lymphoma with low rates of transaminitis: Final results. *Hematological Oncol* (2021) 39. doi: 10.1002/hon.56_2879

70. Moskowitz AJ, Jacobsen E, Ruan J, Schatz JH, Obadi O, Motylinski K, et al. Durable responses observed with jak inhibition in T-cell lymphomas. *Blood* (2018) 132:2922. doi: 10.1182/blood-2018-99-112123

71. Leveille E, Chan LN, Mirza A, Kume K, Müschen M. Syk and Zap70 kinases in autoimmunity and lymphoid malignancies. *Cell Signalling* (2022), 110331. doi: 10.1016/j.cellsig.2022.110331

72. Horwitz SM, Feldman TA, Hess BT, Khodadoust MS, Kim YH, Munoz J, et al. The novel Syk/Jak inhibitor cerdulatinib demonstrates good tolerability and clinical response in a phase 2a study in Relapsed/Refractory peripheral T-cell lymphoma and cutaneous T-cell lymphoma. *Blood* (2018) 132:1001. doi: 10.1182/blood-2018-99-119944

73. Qi W, Spier C, Liu X, Agarwal A, Cooke LS, Persky DO, et al. Alisertib (Mln8237) an investigational agent suppresses aurora a and b activity, inhibits proliferation, promotes endo-reduplication and induces apoptosis in T-nhl cell lines supporting its importance in ptcl treatment. *Leukemia Res* (2013) 37(4):434–9. doi: 10.1016/j.leukres.2012.10.017

74. O'connor OA, Ozcan M, Jacobsen ED, Roncero JM, Trotman J, Demeter J, et al. Randomized phase iii study of alisertib or investigator's choice (Selected single agent) in patients with relapsed or refractory peripheral T-cell lymphoma. *J Of Clin Oncol* (2019). doi: 10.1200/JCO.18.00899

75. Merino M, Kasamon Y, Li H, Ma L, Leong R, Zhou J, et al. Fda approval summary: Crizotinib for pediatric and young adult patients with relapsed or refractory systemic anaplastic Large cell lymphoma. *Pediatr Blood Cancer* (2022):E29602. doi: 10.1002/pbc.29602

76. Gambacorti Passerini C, Farina F, Stasia A, Redaelli S, Ceccon M, Mologni L, et al. Crizotinib in advanced, chemoresistant anaplastic lymphoma kinase–positive lymphoma patients. *J Of Natl Cancer Institute* (2014) 106(2):Djt378. doi: 10.1093/jnci/djt378

77. Richly H, Kim TM, Schuler M, Kim D, Harrison SJ, Shaw AT, et al. Ceritinib in patients with advanced anaplastic lymphoma kinase–rearranged anaplastic Large-cell lymphoma. *Blood J Of Am Soc Of Hematol* (2015) 126(10):1257–8. doi: 10.1182/blood-2014-12-617779

78. Hudson S, Wang D, Middleton F, Nevaldine BH, Naous R, Hutchison RE. Crizotinib induces apoptosis and gene expression changes in alk anaplastic Large cell lymphoma cell lines; brentuximab synergizes and doxorubicin antagonizes. *Pediatr Blood Cancer* (2018) 65(8):E27094. doi: 10.1002/pbc.27094

79. Lunning MA, Horwitz S. Treatment of peripheral T-cell lymphoma: Are we data driven or driving the data? *Curr Treat Options In Oncol* (2013) 14(2):212–23. doi: 10.1007/s11864-013-0232-x

80. Fanale MA, Horwitz SM, Forero-Torres A, Bartlett NL, Advani RH, Pro B, et al. Brentuximab vedotin in the front-line treatment of patients with Cd30+ peripheral T-cell lymphomas: Results of a phase I study. *J Of Clin Oncol Off J Of Am Soc Of Clin Oncol* (2014) 32(28):3137–43. doi: 10.1200/JCO.2013.54.2456

81. Horwitz S, O'connor O, Pro B, Trümper L, Iyer S, Advani R, et al. The echelon-2 trial: 5-year results of a randomized, phase iii study of brentuximab vedotin with chemotherapy for Cd30-positive peripheral T-cell lymphoma. *Ann Of Oncol* (2022) 33(3):288–98. doi: 10.1016/j.annonc.2021.12.002

82. Murai K, Hiroyuki H, Sato A, Yasuro M, Mori Y, Sakuma T. Mogamulizumab monotherapy in the treatment of Relapsed/Refractory peripheral T-cell lymphoma or cutaneous T-cell lymphoma patients in single-institution experience. *Blood* (2017) 130:5179.

83. Ishida T, Jo T, Takemoto S, Suzushima H, Uozumi K, Yamamoto K, et al. Dose-intensified chemotherapy alone or in combination with mogamulizumab in newly diagnosed aggressive adult T-cell leukaemia-lymphoma: A randomized phase ii study. *Br J Of Haematology* (2015) 169(5):672–82. doi: 10.1111/bjh.13338

84. Karube K, Aoki R, Nomura Y, Yamamoto K, Shimizu K, Yoshida S, et al. Usefulness of flow cytometry for differential diagnosis of precursor and peripheral T-cell and nk-cell lymphomas: Analysis of 490 cases. *Pathol Int* (2008) 58(2):89–97. doi: 10.1111/j.1440-1827.2007.02195.x

85. Hamadani M, Collins GP, Caimi PF, Samaniego F, Spira A, Davies A, et al. Camidanlumab tesirine in patients with relapsed or refractory lymphoma: A phase 1, open-label, multicentre, dose-escalation, dose-expansion study. *Lancet Haematology* (2021) 8(6):E433–45. doi: 10.1016/S2352-3026(21)00103-4

86. Zain J, Simpson J, Palmer J, Wong J, Dandapani S, Colcher D, et al. Phase I study of yttrium-90 labeled anti-Cd25 (Atac) monoclonal antibody plus beam for autologous hematopoietic cell transplantation (Ahct) in patients with mature T-cell non-Hodgkin lymphoma, the "A-Tac-Beam regimen". *Blood* (2018) 132:611. doi: 10.1182/blood-2018-99-116907

87. Cai Q, Huang H, Bai B, Yan G, Li J, Lin S, et al. The serum spectrum of cytokines in patients with Nk/T-cell lymphoma and its cilincial significance in survival. *Blood* (2013) 122(21):1759. doi: 10.1182/blood.V122.21.1759.1759

88. Fink EC, Ebert BL. The novel mechanism of lenalidomide activity. *Blood J Of Am Soc Of Hematol* (2015) 126(21):2366–9. doi: 10.1182/blood-2015-07-567958

89. Toumishey E, Prasad A, Dueck G, Chua N, Finch D, Johnston J, et al. Final report of a phase 2 clinical trial of lenalidomide monotherapy for patients with T-cell lymphoma. *Cancer* (2015) 121(5):716–23. doi: 10.1002/cncr.29103

90. Morschhauser F, Fitoussi O, Haioun C, Thieblemont C, Quach H, Delarue R, et al. A phase 2, multicentre, single-arm, open-label study to evaluate the safety and efficacy of single-agent lenalidomide (Revlimid®) in subjects with relapsed or refractory peripheral T-cell non-Hodgkin lymphoma: The expect trial. *Eur J Of Cancer* (2013) 49(13):2869–76. doi: 10.1016/j.ejca.2013.04.029

91. Lunning MA, Horwitz SM, Advani R, Vose JM, Lee HJ, Mehta-Shah N, et al. Phase I/Ii study of choep plus lenalidomide as initial therapy for patients with stage ii-iv peripheral T-cell lymphoma: Phase ii results. *Blood* (2018) 132:2899. doi: 10.1182/blood-2018-99-114110

92. Mehta-Shah N, Lunning MA, Boruchov AM, Ruan J, Nair S, Lynch P, et al. No title. a phase I/Ii trial of the combination of romidepsin and lenalidomide in patients with Relapsed/Refractory lymphoma and myeloma: Activity in T-cell lymphoma. *Am J Hematol* (2015) 96(10):1211–22. doi: 10.1200/jco.2015.33.15_suppl.8521

93. Mehta-Shah N, Moskowitz AJ, Lunning MA, Lynch P, Scheuerman M, Minnal V, et al. A phase Ib/Iia trial of the combination of romidepsin, lenalidomide and carfilzomib in patients with Relapsed/Refractory lymphoma shows complete responses in relapsed and refractory b-and T-cell lymphomas. *Blood* (2017) 130:821. doi: 10.1182/blood.V130.Suppl_1.821.821

94. Ratner L, Waldmann TA, Janakiram M, Brammer JE. Rapid progression of adult T-cell leukemia–lymphoma after pd-1 inhibitor therapy. *New Engl J Of Med* (2018) 378(20):1947–8. doi: 10.1056/NEJMc1803181

95. Khodadoust M, Rook AH, Porcu P, Foss FM, Moskowitz AJ, Shustov AR, et al. Pembrolizumab for treatment of Relapsed/Refractory mycosis fungoides and sezary syndrome: Clinical efficacy in a citn multicenter phase 2 study. *Blood* (2016) 128(22):181. doi: 10.1182/blood.V128.22.181.181

96. Jain S, Van Scoyk A, Morgan EA, Matthews A, Stevenson K, Newton G, et al. Targeted inhibition of Cd47-sirpα requires fc-fcγr interactions to maximize activity in T-cell lymphomas. *Blood J Of Am Soc Of Hematol* (2019) 134(17):1430–40.

97. Safarzadeh Kozani P, Safarzadeh Kozani P, Rahbarizadeh F. Car-T cell therapy in T-cell malignancies: Is success a low-hanging fruit? *Stem Cell Res Ther* (2021) 12 (1):1–17. doi: 10.1186/s13287-021-02595-0

98. Wu Y, Chen D, Lu Y, Dong S, Ma R, Tang W, et al. A new immunotherapy strategy targeted Cd30 in peripheral T-cell lymphomas: Car-modified T-cell therapy based on Cd30 mab. *Cancer Gene Ther* (2022) 29(2):167–77. doi: 10.1038/s41417-021-00295-8

99. Spinner S, Crispatzu G, Yi J, Munkhbaatar E, Mayer P, Höckendorf U, et al. Re-activation of mitochondrial apoptosis inhibits T-cell lymphoma survival and treatment resistance. *Leukemia* (2016) 30(7):1520–30. doi: 10.1038/leu.2016.49

100. Koch R, Christie AL, Crombie JL, Palmer AC, Plana D, Shigemori K, et al. Biomarker-driven strategy for Mcl1 inhibition in T-cell lymphomas. *Blood J Of Am Soc Of Hematol* (2019) 133(6):566–75. doi: 10.1182/blood-2018-07-865527

101. Gualberto A, Scholz C, Mishra V, Janes MR, Kessler L. Ps1002 rhoe, Cxcl12 and Cxcr3 may identify complete responses in acute myeloid leukemia patients treated with tipifarnib. *Hemasphere* (2019) 3(S1):450–1. doi: 10.1097/01.HS9.0000562304.50732.0a

102. Witzig T, Sokol L, Kim W, Foss F, Jacobsen E, De La Cruz Vincente F, et al. Tipifarnib in relapsed or refractory angioimmunoblastic T-cell lymphoma (Aitl) and Cxcl12 peripheral T-cell lymphoma (Ptcl): Preliminary results from a phase 2 study. *Hematological Oncol* (2019) 37:64–5. doi: 10.1002/hon.32_2629

frontiers | Frontiers in Oncology

TYPE Original Research
PUBLISHED 26 May 2023
DOI 10.3389/fonc.2023.1177330

OPEN ACCESS

EDITED BY
Juliana Pereira,
University of São Paulo, Brazil

REVIEWED BY
Maria Christina Cox,
Sapienza University of Rome, Italy
Abrahao Hallack Neto,
Juiz de Fora Federal University, Brazil
Luis Alberto De Pádua Covas Lage,
University of São Paulo, Brazil

*CORRESPONDENCE
Daniel Heudobler
daniel.heudobler@ukr.de

RECEIVED 01 March 2023
ACCEPTED 24 April 2023
PUBLISHED 26 May 2023

CITATION
Fante MA, Harrer DC, Zartner B, Lüke F, Mayer S, Menhart K, Reichle A, Herr W, Vogelhuber M and Heudobler D (2023) All-oral low-dose chemotherapy TEPIP is effective and well-tolerated in patients with peripheral T-cell lymphoma.
Front. Oncol. 13:1177330.
doi: 10.3389/fonc.2023.1177330

All-oral low-dose chemotherapy TEPIP is effective and well-tolerated in patients with peripheral T-cell lymphoma

Matthias A. Fante[1], Dennis C. Harrer[1], Barbara Zartner[1], Florian Lüke[1,2], Stephanie Mayer[1], Karin Menhart[3], Albrecht Reichle[1], Wolfgang Herr[1], Martin Vogelhuber[1] and Daniel Heudobler[1,4]*

[1]Department of Internal Medicine III, Hematology and Internal Oncology, University Hospital Regensburg, Regensburg, Germany, [2]Division of Personalized Tumor Therapy, Fraunhofer Institute for Toxicology and Experimental Medicine, Regensburg, Germany, [3]Department of Nuclear Medicine, University Hospital Regensburg, Regensburg, Germany, [4]Bavarian Cancer Research Center (BZKF), University Hospital Regensburg, Regensburg, Germany

Purpose: Peripheral T-cell lymphoma (PTCL) is a rare and heterogenous hematologic malignancy with poor prognosis especially in elderly and frail patients who are not eligible for intensive treatment. The resulting palliative setting necessitates tolerable but effective schedules for outpatient treatment. TEPIP is a locally developed, all-oral low-dose regimen comprising trofosfamide, etoposide, procarbazine, idarubicin, and prednisolone.

Methods: In this observational retrospective, single-center study, the safety and efficacy of TEPIP was evaluated in 12 patients (pts.) with PTCL treated at the University Medical Center Regensburg between 2010 and 2022. The endpoints were overall response rate (ORR) and overall survival (OS), and adverse events were individually reported according to the Common Terminology Criteria for Adverse Events (CTCAE) criteria.

Results: The enrolled cohort was characterized by advanced age (median 70 years), extensive disease (100% Ann Arbor ≥stage 3), and poor prognosis (75% high/high-intermediate international prognostic index). The most common subtype was angioimmunoblastic T-cell lymphoma (8/12), and 11/12 patients had relapsed or refractory disease at TEPIP onset with a median of 1.5 prior treatment regimens. After a median of 2.5 TEPIP cycles (total of 83 cycles), the ORR was 42% (complete remission 25%), and the OS reached a median of 185 days. Any grade of adverse event (AE) occurred in 8/12 patients, with four patients showing AE ≥CTCAE grade 3 (33%), and the AEs were mainly non-hematological.

Conclusion: TEPIP demonstrated competitive efficacy with a tolerable safety profile in a highly palliative cohort of patients with difficult-to-treat PTCL. The all-oral application, which makes outpatient treatment possible, is particularly noteworthy.

KEYWORDS

TEPIP, relapsed/refractory PTCL, PTCL, metronomic chemotherapy, all-oral treatment, relapsed lymphoma, palliative treatment

Introduction

We recently reported promising safety and efficacy data of the locally developed all-oral, low-dose chemotherapy regimen TEPIP (trofosfamide, etoposide, procarbazine, idarubicin, prednisolone) in relapsed/refractory (R/R) high-grade B cell lymphoma contributing to quality of life by enabling outpatient treatment in a palliative setting (1). Peripheral T-cell lymphomas (PTCL) represent a further rare and heterogenous group of non-Hodgkin lymphoma (2) which is also lacking in effective treatment options in the palliation of patients (pt.) in a R/R state. PTCL with its most common subtypes PTCL not otherwise specified (PTCL-NOS), angioimmunoblastic T-cell lymphoma (AITL), and anaplastic large cell lymphoma (ALCL) (3) exhibit a more aggressive pathobiology as compared with B cell lymphoma showing a dismal overall survival (OS) (4–6).

The established first-line treatment consists of an anthracycline-based multi-agent chemotherapy CHOP backbone (cyclophosphamide, doxorubicin, vincristine, and prednisolone) followed by consolidative high-dose chemotherapy and autologous stem cell transplantation (ASCT) in responding and transplant-eligible patients if considered appropriate. In younger (<60 years) patients, CHOP might be complemented by etoposide to improve event-free survival and progression-free survival (PFS) (7, 8). A recent therapy adjustment was made under the impact of the phase III ECHELON-2 study, which demonstrated a significant improvement of overall response rate (ORR), PFS, and OS in combination with the anti-CD30 antibody brentuximab-vedotin (*i.e.*, BV-CHP) and resulted in the approval for $CD30^+$ PTCL (US Food and Drug Administration) and ALCL (European Medicines Agency (EMA)), respectively (9). Despite all efforts to optimize upfront therapy, the rates of primary refractory and relapsed patients with poor prognosis remain high at up to 70% (10–12), and salvage chemotherapy with consecutive allogeneic stem cell transplantation might be the only option for durable disease control in this high-risk constellation.

In frail and transplant-ineligible patients receiving dose-attenuated CHOP, the 2-year PFS (37%) and OS (47%) are disappointing (13), and the treatment of R/R PTCL in this vulnerable cohort remains challenging. A retrospective analysis from the COMPLETE registry has demonstrated the superiority of a single agent to combination chemotherapy in R/R PTCL (14). The FDA-approved [but not EMA-approved] single-agent therapy antifolate pralatrexate and the histone-deacetylase inhibitors romidepsin and belinostat have shown moderate ORR of approximately 30% and tolerable safety profiles (15–17). Furthermore, special attention is paid to recent studies investigating the efficacy of hypomethylating agents in PTCL with mutations in epigenetic regulators (*e.g.*, TET2), which have yielded promising results (18, 19) and justified the initiation of further studies.

While conventional chemotherapies aim for a maximally tolerated dose, metronomic regimens combine low-dose agents to overcome therapy resistance and reduce toxicity while targeting both the tumor cells and the tumor-promoting microenvironment (20–23). To the best of our knowledge, only a few groups have focused on suchlike regimens in PTCL, however with partially encouraging results (24–27).

In aggregate, we would like to raise awareness for the all-oral, prolonged low-dose chemotherapy regimen TEPIP (trofosfamide, etoposide, procarbazine, idarubicin, prednisolone) as an effective therapeutic option in patients suffering from R/R PTCL with special emphasis on the use in outpatient setting.

Methods

In this retrospective single-center study, we analyzed the efficacy and safety of the all-oral, low-dose chemotherapy TEPIP administered in 12 patients at the University Medical Center Regensburg (UKR) between 2010/01/01 and 2022/12/31. All patients were treated on a compassionate-use basis. We identified the cohort by an in-clinic, medical file database query using the term "TEPIP" and considered only T-cell lymphomas, excluding mixed histopathologies (*e.g.*, NK/T-cell lymphoma). Due to the retrospectivity and the outpatient drug administration, clinical parameters and histologic diagnoses were obtained from medical reports, resulting in a partially limited data set. The analysis was approved by the local Ethics Committee (reference number: 23-3250-104) and performed in compliance with the current Declaration of Helsinki. All cases analyzed were pseudonymized, and patients who were alive gave written informed consent for publication.

Chemotherapy regimen

TEPIP was administered as an all-oral chemotherapy regimen (Figure 1), allowing a fully outpatient treatment, and comprised of trophosphamide at 50 mg (1-1-1, abs. 150 mg), etoposide at 50 mg (1–0–0), procarbazine at 100 mg (1-0-0), and prednisolone at 100 mg (1-0-0) on days 1 to 10, which was shortened to 7 days in case of numerous pre-treatments (equal to or more than two lines of therapy) or advanced age (biological age ≥65 years) as necessary in the majority of our cohort. On days 8 to 10 and 5 to 7, respectively, a daily single dose at 10 mg was added. The course was repeated every 28 days provided that the leukocyte count exceeded 3,000/µl and continued until disease progression or the occurrence of adverse events. All patients treated with TEPIP received sulfamethoxazole-trimethoprim and acyclovir as prophylactic therapies for infections. Antifungal agents or additional antibiotics, such as quinolones, were just initiated in case of suspected infections. Apart from an appropriate antiemesis (*e.g.*, metoclopramide), no specific supportive therapy (*e.g.*, granulocyte-colony-stimulating factor) was administered.

Disease classification and response assessment

At first diagnosis and TEPIP onset, lymphoma disease was staged according to the Ann Arbor criteria. All patients underwent bone marrow biopsy. Prognosis as assessed by the use of the International Prognostic Index (IPI) score is reported. Response assessment was conducted as clinically indicated without fixed schedules, as most patients were treated in an outpatient, palliative setting. Therefore, response was reported as the best response documented by reports of CT or PET-CT imaging according to the 2014 Lugano criteria (28). For patients treated in recent years, staging was performed with PET-CT imaging (patients 3, 4, 10, 11, and 12). Prior to 2020, CT imaging was the standard for response assessment.

Adverse event assessment

Toxicities are listed as reported by outpatient reports and graded according to the Common Terminology Criteria for Adverse Events (CTCAE) version 5.0.

Statistical analysis

Analyses were performed using PRISM 5 (GraphPad, San Diego, CA, USA) and SPSS 28 (IBM, Armonk, NY, USA). The ORR was defined as the sum of patients acquiring complete (CR) or partial remission (PR). Survival was analyzed as OS covering the period between the onset of TEPIP treatment and the patient's death or the end of the observation period (2022/12/31), respectively. PFS was calculated as the difference between the onset of TEPIP treatment and the diagnosis of relapse or patient's death, whereas duration of response (DOR) was defined as the period between a primarily recorded response and the subsequent progression of disease in responding patients. The endpoints OS and PFS (median and estimated OS/PFS at 6 and 12 months ± standard error) were depicted as Kaplan–Meier curves.

Results

Patient characteristics and response to TEPIP therapy

In total, 12 adult patients (five female and seven male) with histologically confirmed PTCL were treated at our medical center

A

10-day protocol			days	1	2	3	4	5	6	7	8	9	10	11-28
TRO	50 mg	1-1-1		x	x	x	x	x	x	x	x	x	x	-
ETO	50 mg	1-0-0		x	x	x	x	x	x	x	x	x	x	-
PRC	100 mg	1-0-0		x	x	x	x	x	x	x	x	x	x	-
IDA	10 mg	1-0-0		-	-	-	-	-	-	-	x	x	x	-
PDN or DEX	100 mg or 8 mg	1-0-0 / 1-0-1		x	x	x	x	x	x	x	x	x	x	-

B

7-day protocol			days	1	2	3	4	5	6	7	8-28
TRO	50 mg	1-1-1		x	x	x	x	x	x	x	-
ETO	50 mg	1-0-0		x	x	x	x	x	x	x	-
PRC	100 mg	1-0-0		x	x	x	x	x	x	x	-
IDA	10 mg	1-0-0		-	-	-	-	x	x	x	-
PDN or DEX	100 mg or 8 mg	1-0-0 / 1-0-1		x	x	x	x	x	x	x	-

FIGURE 1
Trofosfamide, etoposide, procarbazine, idarubicin, and prednisolone schedules. **(A)** A full-dose, 10-day or **(B)** a dose-reduced, 7-day protocol was applied depending on the patient's performance status and expected toxicity tolerance. TRO, trofosfamide; ETO, etoposide; PRC, procarbazine; IDA, idarubicin; PDN, prednisolone; DEX, dexamethasone; x, administration; hyphen, no administration.

on a compassionate-use basis with the all-oral low-dose chemotherapy regimen TEPIP based on the daily application of low-dose trofosfamide, etoposide, procarbazine, idarubicin, and prednisolone (Figure 1). The baseline patient characteristics, including previous lines of therapy, are depicted in Table 1. The median age was 70 years (range, 43–84). AITL represented the most common T-cell lymphoma subtype (8 pt.). Prior to TEPIP initiation, all patients presented with stage 3 or higher according to the Ann Arbor classification accompanied by IPI scores predominantly at the high-intermediary level. A total of 11 patients were subjected to TEPIP treatment after having failed up to four different lines of therapy, including ASCT (1 pt.) and experimental checkpoint blockade with nivolumab (1 pt.). Only one patient did not receive any treatment before starting TEPIP. The clinical responses to TEPIP are documented in Table 2, and individual clinical courses are shown in Figure 2. Each patient underwent on average (median) of 2.5 cycles (range, 1 to 24) of TEPIP treatment, and a total of 83 cycles was applied. The median OS was 185 days (± 64.4 days, n = 11), and the median PFS amounted to 114 days (± 45.0 days, n = 12) (Figure 3). The overall response rate (CR + PR) was 42%, and the median duration of response was 10 months (range, 1–35 months). Three patients, including a heavily pretreated patient relapsing from high-dose chemotherapy and subsequent autologous stem cell transplantation (pt. 11), achieved complete remission (25%) in response to TEPIP treatment. At the time of this writing (February 2023), one patient (pt. 3) is still in complete remission despite the discontinuation of TEPIP treatment, while relapse was observed in the other complete responders after 10 months (patient 10) and 25 months (pt. 11), respectively. Remarkably, re-initiation of TEPIP treatment could re-induce complete remission in patient 11. Thereafter, TEPIP had to be permanently stopped due to pancytopenia originating from arising secondary acute myeloid leukemia, which necessitated switching to 5-azacitidine treatment. Moreover, two patients displayed transient partial remissions of their T-cell lymphoma lesions, and stable disease could be observed in two additional patients. Approximately half of all patients were primary refractory to TEPIP treatment. Those patients failing TEPIP treatment were

TABLE 1 Characteristics of T-cell lymphoma patients.

Patient ID	Sex	Age[a] (years)	Subtype	Ann Arbor staging at 1st dx/TEPIP start	IPI[a]	Pre-treatment
1	Male	70	AITL	IIA/IIIA	Low	6 × CHOP 4 × DHAC
2	Female	76	AITL	IIIB/IIIA	High-intermediate	6 × CHOP
3	Female	72	AITL	IIIA/IVA	Low-intermediate	12 × CHOP
4	Female	70	AITL	IIIA/IIIA	Low-intermediate	6 × CHOP
5	Male	45	AITL	IVA/IVA	High-intermediate	5 × R-CHOEP 1 × DHAP
6	Male	79	PTCL-NOS	IVAE/IVAE	High-intermediate	Prednisolone Vincristine 3 × CHOP
7	Male	84	AITL	-/IVA	High	None
8	Male	43	HSTL	IVB/IVB	High-intermediate	3 × CHOEP 1 × DHAP Fludarabine Alemtuzumab
9	Female	71	AITL	IVB/IIIA	High-intermediate	6 × CHOP 2 × DHAC 8 × BV
10	Female	67	FTCL	IVAE/IVAE	High-intermediate	6 × CHOEP 6 × N/Gem/Ox MTX weekly
11	Male	66	AITL	IIIA/IIIA	High-intermediate	5 × CHOP 1 × DHAP/1 × DHAC BEAM + ASCT 2 × Gem/Ox
12	Male	66	ALCL	IVA/IVA	High-intermediate	6 × BV-CHP

ATIL, angioimmunoblastic T-cell lymphoma; PTCL-NOS, peripheral T-cell lymphoma not otherwise specified; HSTL, hepatosplenic T-cell lymphoma; FTCL, follicular T-cell lymphoma; ALCL, anaplastic large cell lymphoma; CHOEP, cyclophosphamide, doxorubicin, vincristine, etoposide, and prednisolone; DHAP/C, dexamethasone, cytarabine, cisplatin/carboplatin; BV, brentuximab vedotin; N/Gem/Ox, nivolumab, gemcitabine, oxaliplatin; BEAM, BCNU, etoposide, cytarabine, and melphalan; ASCT, autologous stem cell transplantation; CHP, cyclophosphamide, doxorubicin, and prednisolone.
[a]At first diagnosis.

10.3389/fonc.2023.1177330

TABLE 2 Response to trofosfamide, etoposide, procarbazine, idarubicin, and prednisolone (TEPIP) therapy.

Patient ID	TEPIP cycles (n)	Best response[a] (DOR, months)[c]	OS[b] (days)	Subsequent therapy
1	2	PD	77	BSC
2	16	SD (18)	704	BSC
3	15	CR (35+)	1193*	No treatment
4	2	PD	185	Gemcitabine/carboplatin/dexamethasone
5	2	PR (2)	82	BSC
6	1	PD	NA[d]	Cyclophosphamide/etoposid/procarbazine/prednisolone
7	3	PD	148	BSC
8	1	PD	42	BSC
9	4	SD (1)	134	Lomustin
10	11	CR (10)	558	Belinostat
11	24	CR (25)	218	5-Azacitidine
12	2	PR (1)	251	BSC
median	2.5	10 (DOR)	185	

DOR, duration of response; PD, progressive disease; SD, stable disease; PR, partial remission; CR, complete remission; BSC, best supportive care; NA, not available.
[a]Response to TEPIP therapy.
[b]Overall survival of patients, (the symbol "*" signifies being still alive).
[c]Duration of response in the case of SD/PR/CR (the symbol "+" signifies an ongoing response).
[d]Overall survival not available because of loss to follow-up.

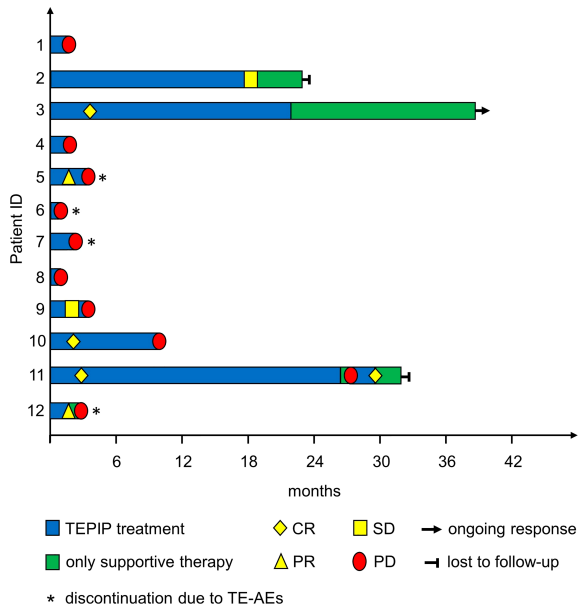


FIGURE 2
Treatment and response of the included patients during the treatment course as depicted by a swimmer plot.

either switched to best supportive care treatment or were continued on individual protocols as depicted in Table 2. In summary, the all-oral TEPIP treatment conducted in an outpatient setting harbors the potential for durable remissions in heavily pretreated patients with advanced T-cell lymphoma.

Analysis of the versatile therapeutic potential of TEPIP in patients with PTCL based on five clinical courses

Out of the 12 patients with PTCL treated at our center, the clinical courses of five patients (patient ID numbers 3, 5, 10, 11, and 12 according to Tables 1 and 2) particularly emphasize the versatile therapeutic potential of TEPIP in PTCL patients.

Case 1 (pt. 3)

A 72-year-old female patient presented with progressive localized axillary lymphadenopathy to our department. After a diagnosis of AITL, treatment with 6x CHOP-14 helped her achieve complete remission, but it had to be repeated due to relapse at 27 months later. After a further six cycles, a long-lasting (6.5 years) CR was observed. Thereafter, the patient suffered again from stage IVA [disseminated lymph nodes (LN), 1% to 2% bone marrow (BM) infiltration] relapse of the known AITL, which was molecularly characterized as ALK1-negative, TP53/17p del-negative, and TET2-positive. Due to the advanced patient age, a palliative treatment with all-oral TEPIP without idarubicin (q4w, d1-7) was initiated. After 4 months, a CR was observed despite dose reductions (trofosfamide, prednisolone, and procarbazine) and a stretched cycle duration from 4 to 6 weeks due to limited tolerability (fatigue). TEPIP was discontinued after 23 months due to multiple cutaneous squamous cell carcinomas with persistent complete remission. Until the end of the observational period (31-Dec-2022), no relapse has occurred.

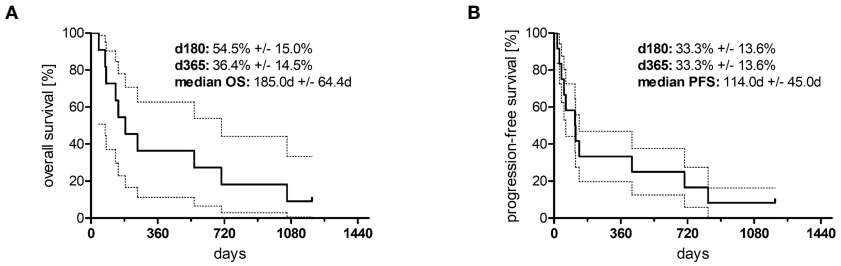


FIGURE 3
Kaplan–Meier curves of **(A)** overall survival (*n* = 10) and **(B)** progression-free survival (*n* = 11). The area between the dotted lines represents the standard error.

Conclusion: Patient case 1 demonstrates a durable response to TEPIP treatment in an elderly and frail patient, which allowed dose adjustment to achieve individual treatment tolerability.

Case 2 (pt. 5)

A 45-year-old male patient presented with a liver-infiltrating stage IVB AITL (additional BM infiltration) and simultaneous EBV-positive DLBCL. During the first-line treatment with 5x R-CHOEP, the patient experienced several severe adverse events, and a CT scan and bone marrow puncture before the planned sixth course showed a massive progression of AITL. However, no persisting evidence of DLBCL was found. Therapy was rotated to the salvage regimen DHAP in the intention of later high-dose (HD) chemotherapy and ASCT. A pneumogenic sepsis interrupted the treatment course, resulting in another rapid progression. As a consequence, TEPIP treatment (without idarubicin) was initiated, which achieved prompt and impressive PR (resolved liver and minimal pulmonary manifestation). After a second course, the patient developed pancytopenia that necessitated a protracted recovery period, which led to progression and death due to failure of lymphoma-infiltrated organs.

Conclusion: Patient case 2 identifies TEPIP as a treatment option in refractory AITL, however, at the expense of potentially serious adverse events such as life-threatening cytopenia.

Case 3 (pt. 10)

A 67-year-old female patient was referred to the Department of Dermatology at the University Hospital Regensburg with pruritus, eczema, and subcutaneous nodules turning out to be a stage IVAE follicular T-cell lymphoma (aberrant co-expression of CD79a) with cutaneous and disseminated LN manifestation coupled with simultaneous EBV and CD30+ B cell proliferation. After 6x (R-) CHOEP, a CR was achieved; however, within 3 months, a very early relapse occurred. The patient was enrolled and randomized into the experimental arm (nivolumab, gemcitabine, and oxaliplatin) of the NIVEAU trial (29) but suffered from distinct PD after an initial mixed response. As a third-line treatment, MTX weekly was administered, however without any response. Finally, treatment was rotated to TEPIP, and the patient developed CR which lasted for 10 months. During this period, no relevant adverse events occurred. Upon progression, the patient underwent involved site radiation and belinostat treatment followed by short episodes of bendamustine and brentuximab-vedotin treatment, but no further response was achieved.

Conclusion: Patient case 3 demonstrates a long-lasting response without adverse events by TEPIP in a heavily pre-treated and refractory patient with follicular T-cell lymphoma in nodal and extranodal lesions.

Case 4 (pt. 11)

A 66-year-old male patient with a history of prostatectomy due to prostate carcinoma and with a persistent single osteoblastic osseous metastasis (left os ilium) presented to our department with a first diagnosis of stage III AITL with exclusive lymph node manifestation. After five courses of CHOP, peripheral blood stem cell (PBSC) apheresis between each course of DHAP and DHAC was performed. At the timepoint of HD chemotherapy (BEAM) and ASCT, the patient was in complete remission. Within 6 months upon ASCT, early relapse occurred, and despite two courses of salvage treatment with gemcitabine and oxaliplatin, the disease progressed. The PSMA-PET result confirmed a stable disease (M1 oss.) of prostate carcinoma. Due to AITL progression, treatment was rotated to all-oral TEPIP (q4w, d1-7), and complete and durable remission (24 months) was achieved within two courses (Figure 4). Finally, treatment was terminated due to long-lasting CR after 2 years. Within the following 3 months, the patient relapsed with lymphoma infiltration of the skin, lymph nodes, and bone marrow (stage IV). At the same timepoint, a myelodysplastic neoplasm (MDS-RS-MLD) was incidentally diagnosed. TEPIP was re-initiated in five-weekly courses, resulting in a renewed very good PR within 3 months (Figure 4). Due to MDS-related cytopenia, TEPIP was replaced with 5-azacitidine. However, the patient died due to progression to AML at 5 months later.

Conclusion: Patient case 4 illustrates TEPIP as a safe and potent treatment option in AITL even after ASCT and with concomitant metastasized prostate carcinoma. In addition, it underpins its ability to reinduce remission after relapse.

Case 5 (pt. 12)

A 66-year-old pre-diseased (CVD, s/p esophagogastric junctional adenocarcinoma) male patient presented with stage IV A ALK-negative, CD30-positive ALCL with lymph node, cutaneous, and pulmonary manifestations, and treatment was initiated with BV-CHP. Between courses 2 and 3, PBSC apheresis

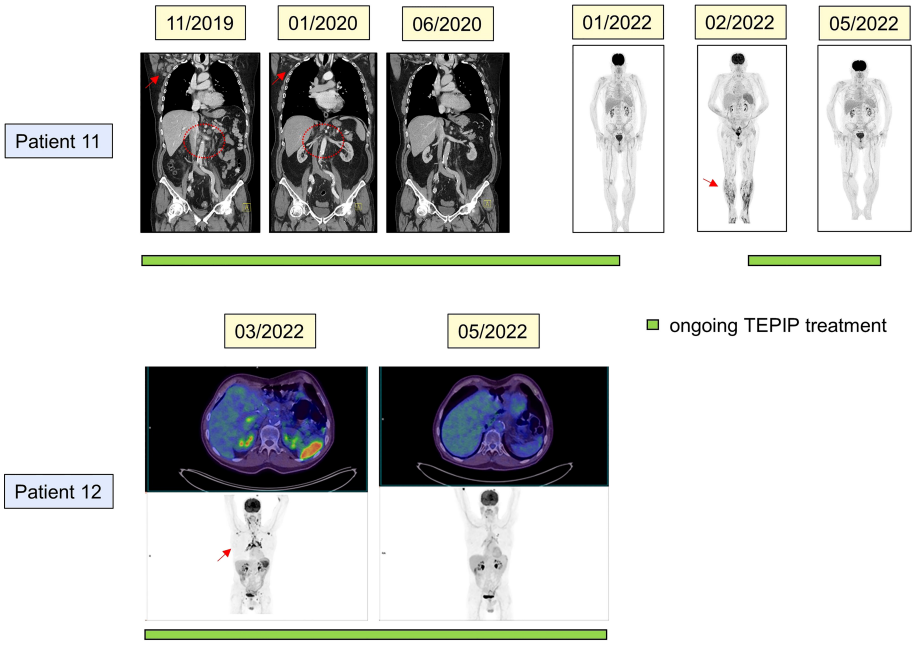


FIGURE 4
Clinical course of two exemplary patients (pt. 11: upper panels; pt. 12 lower panels) as assessed by radiological imaging. Depicted are the treatment responses at the indicated timepoints. Computed tomography and positron emission tomography were utilized. Green bars denote time on trofosfamide, etoposide, procarbazine, idarubicin, and prednisolone treatment. Red arrows and dotted circles indicate lymphoma manifestations.

was performed. After six courses, only PR was achieved, and within a further 2 months of watchful waiting, the disease distinctly progressed with new liver, spleen, and testicular manifestations. Due to a reduced general condition and his medical history, an intensive salvage or high-dose regimen was discarded, and low-dose TEPIP was administered (q4w, d1-7). After two courses, a PR was observed (Figure 4). However, the patient developed neurologic symptoms (*e.g.*, amnesic aphasia) which later turned out to be a symptom of transient ischemic attack/stroke. After the interruption of treatment, the patient rapidly relapsed within 3 months and passed away.

Conclusion: Patient case 5 demonstrates the therapeutic option of TEPIP in ALCL when treatment with ALK inhibitors is not reasonable or possible.

Safety and toxicity profile of TEPIP therapy

Adverse events emergent to TEPIP were observed in eight patients (4/8 CTCAE ≥grade 3) as documented in Table 3. Treatment-related deaths did not occur. One patient (pt. 11) died from concomitant secondary acute myeloid leukemia arising from myelodysplastic syndrome. The development of MDS/secondary acute myeloid leukemia was rated to be most likely attributed to multiple previous therapies including high-dose chemotherapy; nevertheless, a causal relation between MDS/AML and TEPIP is also possible. Grade 4 toxicity was only observed in one patient developing severe leukopenia, which improved after the discontinuation of TEPIP therapy. The most frequent toxicities overall were fatigue, thrombopenia, and elevated levels of liver transaminases, with the latter two additionally representing the most frequent grade 3 toxicities. In four patients, TEPIP was permanently ceased: the reasons were hepatotoxicity (pt. 5 and 6), cytopenia (pt. 5 and 6), and renal toxicity (pt. 7) coupled with insufficient responses to therapy, whereas one patient (pt. 12) developed neurological toxicity manifesting itself as amnesic aphasia. Nevertheless, this patient suffered from long-standing atrial fibrillation and pronounced atherosclerosis, which could have crucially contributed to the occurrence of neurological toxicity. Furthermore, dose reductions of trofosfamide owing to intolerable fatigue (pt. 3 and 11) or dysuria (pt. 2) were required in three patients, while dose reduction of etoposide and idarubicin due to fatigue was necessary in one patient (pt. 11). After the dose modifications, increase of treatment intervals, or discontinuation of TEPIP treatment, almost all toxicities appeared to be at least partially reversible. One patient (pt. 3) developed cutaneous squamous cell carcinoma (cSCC) lesions on both arms, prompting the cessation of TEPIP therapy to boost wound healing after the surgical removal of cSCC. In summary, TEPIP treatment is associated with a tolerable safety profile qualifying for use in an outpatient setting.

Discussion

Treatment of T-cell lymphoma in elderly and frail patients poses a challenge to clinicians. While first-line treatment with age-adjusted, dose-attenuated multi-agent chemotherapy (*e.g.*, miniCHOP) yields disappointing therapeutic success with only low PFS and OS rates (13), the next-line options are limited as

TABLE 3 Treatment-emergent adverse events.

Patient ID	Toxicity	Grade[a]	Response	Outcome
1	Fatigue	1	Temporary discontinuation of TEPIP	Improvement
2	Thrombopenia	2	Temporary discontinuation of TEPIP	Improvement
	Dysuria	2	Dose reduction of trofosfamide and prednisolone	Improvement
3	Fatigue	2	Dose reduction of trofosfamide and longer treatment intervals of 6 weeks	Improvement
	Cutaneous squamous cell carcinoma	Not applicable	Discontinuation of TEPIP	Improvement after surgical excision
4	None	–	–	–
5	Elevated liver transaminases	3	Discontinuation of TEPIP	Resolution
	Leukopenia	4	Discontinuation of TEPIP	Improvement
	Anemia	3	Discontinuation of TEPIP	Improvement
	Thrombopenia	3	Discontinuation of TEPIP	Improvement
6	Elevated liver transaminases	3	Discontinuation of TEPIP	Improvement
	Thrombopenia	3	Discontinuation of TEPIP	Persistence
7	Infection (urinary)	3	Discontinuation of TEPIP	Improvement
	Elevated creatinine	3	Discontinuation of TEPIP	Improvement
8	None	–	–	–
9	None	–	–	–
10	None	–	–	–
11	Fatigue MDS-RS-MLD	2 Not applicable	Discontinuation of Idarubicin and dose reduction of trofosfamide and etoposide -	Improvement Progression to AML
12	Neurological symptoms (word finding disorder) originating from stroke	3	Discontinuation of TEPIP	Improvement

[a]According to Common Terminology Criteria of Adverse Events Version 5 (2017).

the respective patients are not eligible for intensive salvage treatment, HD chemotherapy, or stem cell transplantation. As a result, there is medical need for palliative regimens considering the necessity of effective low-dose approaches while meeting many patients' request for outpatient concepts.

In the study presented here, we show data from an elderly (median, 70 years) cohort of 12 patients with PTCL being under treatment with an all-oral, low-dose chemotherapy regimen TEPIP at the Department of Hematology of the University Medical Center Regensburg over the past decade. TEPIP comprises four oral chemotherapeutic drugs (trofosfamide, etoposide, procarbazine, and idarubicin) plus steroids (prednisolone or dexamethasone), each of which has been proven effective in the treatment of non-Hodgkin lymphoma disease (7, 27, 30–33). The majority of the cohort is characterized by poor prognosis with high-intermediate or high IPI scores and extensive disease (Ann Arbor stage III or IV) at TEPIP onset. Treatment with the TEPIP regimen followed a median of 1.5 prior treatment lines (range, 0–4), and a median of 2.5 TEPIP courses was applied. An ORR of 42% was achieved; however, an equally large group of patients experienced primary treatment failure. Overall survival reached a median of 6.2 months (185 days) with one patient still being alive in sustained complete remission. Anecdotal observations of responding patients (as demonstrated in the brief patient cases) highlight the potential benefit of TEPIP treatment in complex everyday settings, such as frail patients with relapse, refractory disease in various PTCL subtypes, and patients with a concurrent solid malignancy. Remarkably, several patient cases have shown rapid relapse after the interruption or discontinuation of therapy, but partial remission was achieved at least in one case by resuming TEPIP therapy.

All but one patient received TEPIP at a relapsed or refractory stage, which historically has a dismal prognosis. Mak et al. reported a median OS in R/R PTCL of 5.5 months (11), which was still confirmed 5 years later and more favorable only in a selected patient cohort with unimpaired performance status who were receiving salvage chemotherapy (12). This is particularly important for elderly patients who are not eligible for intensive first-line (and next-line) treatment due to their increased susceptibility to chemo-associated toxicities and, as a result, are more often likely to have primary refractory or relapsed diseases as reflected by the reported 2-year PFS of 37% (13). FDA-approved single-agent R/R treatment strategies like anti-folate pralatrexate or HDAC inhibitors romidepsin and belinostat have

shown ORR of 25% to 29% and DOR of 10 to 17 months (15–17), not exceeding the ORR observed with TEPIP treatment. Better ORR could be achieved with the antibody–toxin conjugate brentuximab–vedotin (BV). However, studies required CD30 positivity or an ALCL phenotype (34, 35), which, in turn, limits the availability to a selected patient population. Additionally, most novel drugs are administered intravenously and require close monitoring of side effects or through in-patient treatment. Furthermore, promising results demonstrating the efficacy of hypomethylating agents (HMA) in PTCL with mutations in epigenetic regulators (*e.g.*, DNTM3A, TET2, and IDH2) have been obtained in smaller studies (18, 36), raising hope for precise, molecularly tailored treatment regimens. In this context, oral HMA preparations, in particular, represent a promising approach for future outpatient strategies and are the subject of current trials (NCT04747236 and NCT03161223). However, robust data are still pending, and the potential role in a therapeutic sequence remains unclear. As of now, neither HDAC inhibitors nor HMA has received approval from the EMA for use in T-cell lymphoma. Thus, the second-line treatment of elderly/frail patients with R/R PTCL requires a broad therapeutic repertoire to enable individual treatment. Cox and colleagues have recently shown that the all-oral chemotherapy regimen DEVEC (prednisolone, etoposide, vinorelbine, and cyclophosphamide) achieves impressive ORR/CR (66%/25%) and OS (13 months) with a tolerable rate of adverse events in an elderly cohort of R/R PTCL with poor prognosis (IPI ≥3: 75%) (24). The outperformance in terms of ORR and OS despite a comparable cohort might be at least partially explained by a higher median number of therapy courses in the DEVEC (median, 8.5 courses) compared with our study (median, 2.5 courses), underscoring the highly palliative intention to treat in the TEPIP cohort. Furthermore, DEVEC was administered in a metronomic manner, potentially leading to differential, pleiotropic pharmacodynamics (37, 38). In recognition of the outstanding DEVEC results, a metronomic application of TEPIP with lower doses, more regular administration, and reduced drug-free brakes (39, 40) should be evaluated in future studies. There are few other reports of low-dose, all-oral treatment regimens in PTCL (25–27), emphasizing the need for more outpatient options in advanced palliative settings that provide a decentralized treatment approach, particularly for rural areas. Importantly, oral HMA and TEPIP may even pose an appropriate palliative first-line therapy for PTCL in elderly/frail patients in case of rejection of intravenous chemotherapy, which requires regular intravenous access and presentation to specialized oncological facilities. For patients declining systemic therapy in favor of best supportive care, all-oral therapy with TEPIP might be discussed in the first line.

In our study, treatment-emergent adverse events (TE-AE) CTCAE ≥ grade 3 were observed in 33% of patients, with one grade 4 (leukopenia) but no fatal TE-AE. Relevant infectious disease only occurred in one patient, which is most likely due to the recommended dose adjustments in case of cytopenia. As this was performed according to the physician's choice, reliable numeric statements unfortunately are not available. One patient with long-term TEPIP treatment (pt. 3) developed multiple cSCC, which was classified as possibly related to treatment. However, a retrospective analysis of the patient's previous circumstances (*e.g.*, sun exposure) was not possible. Another patient (pt. 11) died from MDS/AML 5 months after re-discontinuation of TEPIP, which occurred concurrently with PTCL relapse after an initial complete response and prolonged 2 years of TEPIP treatment (Figure 1). TEPIP includes alkylating agents (procarbazine and trofosfamid) and topoisomerase II inhibitors (etoposide and idarubicin), which are known to cause secondary MDS and AML in a group of patients after the treatment of hematologic malignancies (41, 42). Thus, we recognize a potentially therapy-related event. However, one must consider the four prior treatment regimens, including high-dose BEAM, in this patient. Furthermore, in the recently reported cohort of patients with DLBCL treated with TEPIP, no patient suffered from secondary malignancies (1). Hypothetically, an additional potentially contributing factor to the occurrence of MDS and AML in the cohort of patients with PTCL treated with TEPIP might derive from potential clones with pre-lymphomatous TET2 and DNMT3A mutations. Those genetical alterations are frequently present in patients diagnosed with AITL, and AITL *per se* is associated with a certain co-occurrence of myeloid neoplasias (43). Nonetheless, we recommend that the maintenance of therapy be regularly and carefully reviewed. Notably, the same patient started TEPIP while suffering from an osseous metastasized prostate carcinoma which has not been progressing despite a long-term treatment. In summary, compared with previous palliative treatment regimens (15–17, 34), the safety profile of TEPIP is tolerable and manageable. Due to retrospectivity and the outpatient setting, additional unreported events cannot be completely excluded.

The rapid relapse after discontinuation in patient 4 with a subsequent re-induction of remission stirs the question for TEPIP maintenance therapy. A limiting factor for continuous therapy is set by the cumulative dose of idarubicin to prevent anthracycline-associated cardiomyopathy. Thereafter, TEPP without idarubicin could pose a backbone for long-lasting maintenance. In clinical practice, we advocate the continuation of TEPIP/TEPP therapy past remission induction until disease progression or the occurrence of unacceptable toxicity.

We are aware of the limitations of the study derived from the small, heterogenous cohort, a partially incomplete data set due to the palliative outpatient setting, retrospectivity without a control group as well as the short median duration of TEPIP treatment, which negatively affect the explanatory power of our results. R/R PTCL is a rare disease, and the clinical courses of the reported cohort span more than a decade (2010–2022), partially explaining the incomplete histopathologic reports (examination of molecular profiles or antigen expression) that may impact treatment decisions today. Additionally, the currently available treatment guidelines were published after the first patients were treated. All patients included in this study were treated individually and independently on a compassionate-use basis. Hence, individual treatment regimens differ, compromising the direct comparability of patients as well as the generation of pooled analyses for common outcome parameters, such as overall survival and progression-free survival. Moreover, no fixed and pre-established parameters for the evaluation of safety and efficacy are present. Thus, the direct comparability of patients is compromised.

In conclusion, the TEPIP regimen presented is a treatment option for patients with R/R PTCL that is both effective and well tolerated, with a focus on the quality of life in advanced palliative settings. We encourage the initiation of controlled clinical trials to prospectively evaluate TEPIP and gain further experience.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors without undue reservation.

Ethics statement

The analysis was approved by the University of Regensburg (reference number: 23-3250-104) and performed in compliance with the current Declaration of Helsinki. All cases analyzed were pseudonymized, and patients who were alive gave written informed consent for publication.

Author contributions

MF, DCH, BZ, FL, SM, WH, MV, AR and DH treated the patients. MF, DCH and DH analyzed the data and wrote the manuscript. All authors contributed to the article and approved the submitted version.

Funding

This study incorporated funding by the Deutsche Forschungsgemeinschaft (German Research Foundation) to DCH (grant 324392634-TRR221) and by the Else-Kröner Fresenius Foundation to DCH.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Fante MA, Felsenstein M, Mayer S, Gerken M, Klinkhammer-Schalke M, Herr W, et al. All-oral low-dose chemotherapy TEPIP is effective and well-tolerated in Relapsed/Refractory patients with aggressive b-cell lymphoma. *Front Oncol* (2022) 12:852987. doi: 10.3389/fonc.2022.852987

2. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, et al. The 2016 revision of the world health organization classification of lymphoid neoplasms. *Blood* (2016) 127:2375–90. doi: 10.1182/blood-2016-01-643569

3. Vose J, Armitage J, Weisenburger D. International peripheral T-cell and natural killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26:4124–30. doi: 10.1200/JCO.2008.16.4558

4. Teras LR, DeSantis CE, Cerhan JR, Morton LM, Jemal A, Flowers CR. 2016 US Lymphoid malignancy statistics by world health organization subtypes. *CA Cancer J Clin* (2016) 66:443–59. doi: 10.3322/caac.21357

5. Abouyabis AN, Shenoy PJ, Sinha R, Flowers CR, Lechowicz MJ. A systematic review and meta-analysis of front-line anthracycline-based chemotherapy regimens for peripheral T-cell lymphoma. *ISRN Hematol* (2011) 2011:623924. doi: 10.5402/2011/623924

6. Gisselbrecht C, Gaulard P, Lepage E, Coiffier B, Brière J, Haioun C, et al. Prognostic significance of T-cell phenotype in aggressive non-hodgkin's lymphomas. groupe d'Etudes des lymphomes de l'Adulte (GELA). *Blood* (1998) 92:76–82.

7. Schmitz N, Trümper L, Ziepert M, Nickelsen M, Ho AD, Metzner B, et al. Treatment and prognosis of mature T-cell and NK-cell lymphoma: an analysis of patients with T-cell lymphoma treated in studies of the German high-grade non-Hodgkin lymphoma study group. *Blood* (2010) 116:3418–25. doi: 10.1182/blood-2010-02-270785

8. Ellin F, Landström J, Jerkeman M, Relander T. Real-world data on prognostic factors and treatment in peripheral T-cell lymphomas: a study from the Swedish lymphoma registry. *Blood* (2014) 124:1570–7. doi: 10.1182/blood-2014-04-573089

9. Horwitz S, O'Connor OA, Pro B, Illidge T, Fanale M, Advani R, et al. Brentuximab vedotin with chemotherapy for CD30-positive peripheral T-cell lymphoma (ECHELON-2): a global, double-blind, randomised, phase 3 trial. *Lancet* (2019) 393:229–40. doi: 10.1016/S0140-6736(18)32984-2

10. Lansigan F, Horwitz SM, Pinter-Brown LC, Rosen ST, Pro B, Hsi ED, et al. Outcomes for relapsed and refractory peripheral T-cell lymphoma patients after front-line therapy from the COMPLETE registry. *Acta Haematol* (2020) 143:40–50. doi: 10.1159/000500666

11. Mak V, Hamm J, Chhanabhai M, Shenkier T, Klasa R, Sehn LH, et al. Survival of patients with peripheral T-cell lymphoma after first relapse or progression: spectrum of disease and rare long-term survivors. *J Clin Oncol* (2013) 31:1970–6. doi: 10.1200/JCO.2012.44.7524

12. Bellei M, Foss FM, Shustov AR, Horwitz SM, Marcheselli L, Kim WS, et al. The outcome of peripheral T-cell lymphoma patients failing first-line therapy: a report from the prospective, international T-cell project. *Haematologica* (2018) 103:1191–7. doi: 10.3324/haematol.2017.186577

13. Choi E-J, Hong JY, Yoon DH, Kang J, Park C-S, Huh J, et al. Treatment outcomes of dose-attenuated CHOP chemotherapy in elderly patients with peripheral T cell lymphoma. *Blood Res* (2017) 52:270–5. doi: 10.5045/br.2017.52.4.270

14. Stuver RN, Khan N, Schwartz M, Acosta M, Federico M, Gisselbrecht C, et al. Single agents vs combination chemotherapy in relapsed and refractory peripheral T-cell lymphoma: results from the comprehensive oncology measures for peripheral T-cell lymphoma treatment (COMPLETE) registry. *Am J Hematol* (2019) 94:641–9. doi: 10.1002/ajh.25463

15. O'Connor OA, Pro B, Pinter-Brown L, Bartlett N, Popplewell L, Coiffier B, et al. Pralatrexate in patients with relapsed or refractory peripheral T-cell lymphoma: results from the pivotal PROPEL study. *J Clin Oncol* (2011) 29:1182–9. doi: 10.1200/JCO.2010.29.9024

16. O'Connor OA, Horwitz S, Masszi T, van Hoof A, Brown P, Doorduijn J, et al. Belinostat in patients with relapsed or refractory peripheral T-cell lymphoma: results of the pivotal phase II BELIEF (CLN-19) study. *J Clin Oncol* (2015) 33:2492–9. doi: 10.1200/JCO.2014.59.2782

17. Coiffier B, Pro B, Prince HM, Foss F, Sokol L, Greenwood M, et al. Results from a pivotal, open-label, phase II study of romidepsin in relapsed or refractory peripheral T-cell lymphoma after prior systemic therapy. *J Clin Oncol* (2012) 30:631–6. doi: 10.1200/JCO.2011.37.4223

18. Lemonnier F, Dupuis J, Sujobert P, Tournillhac O, Cheminant M, Sarkozy C, et al. Treatment with 5-azacytidine induces a sustained response in patients with angioimmunoblastic T-cell lymphoma. *Blood* (2018) 132:2305–9. doi: 10.1182/blood-2018-04-840538

19. O'Connor OA, Falchi L, Lue JK, Marchi E, Kinahan C, Sawas A, et al. Oral 5-azacytidine and romidepsin exhibit marked activity in patients with PTCL: a multicenter phase 1 study. *Blood* (2019) 134:1395–405. doi: 10.1182/blood.2019001285

20. Kareva I, Waxman DJ, Lakka Klement G. Metronomic chemotherapy: an attractive alternative to maximum tolerated dose therapy that can activate anti-tumor immunity and minimize therapeutic resistance. *Cancer Lett* (2015) 358:100–6. doi: 10.1016/j.canlet.2014.12.039

21. Kerbel RS. Inhibition of tumor angiogenesis as a strategy to circumvent acquired resistance to anti-cancer therapeutic agents. *Bioessays* (1991) 13:31–6. doi: 10.1002/bies.950130106

22. Klement G, Baruchel S, Rak J, Man S, Clark K, Hicklin DJ, et al. Continuous low-dose therapy with vinblastine and VEGF receptor-2 antibody induces sustained tumor regression without overt toxicity. *J Clin Invest* (2000) 105:R15–24. doi: 10.1172/JCI8829

23. Klement G, Huang P, Mayer B, Green SK, Man S, Bohlen P, et al. Differences in therapeutic indexes of combination metronomic chemotherapy and an anti-VEGFR-2 antibody in multidrug-resistant human breast cancer xenografts. *Clin Cancer Res* (2002) 8:221–32.

24. Cox MC, Banchi M, Pelliccia S, Di Napoli A, Marcheselli L, Patti C, et al. All-oral metronomic DEVEC schedule in elderly patients with peripheral T cell lymphoma. *Cancer Chemother Pharmacol* (2020) 86:841–6. doi: 10.1007/s00280-020-04172-3

25. Tucci A, Cerqui E, Ungari M, Ferrari S, Baushi L, Facchetti F, et al. Continuous oral cyclophosphamide and prednisolone as a valuable treatment option for peripheral T cell lymphoma. *Br J Haematol* (2011) 152:113–6. doi: 10.1111/j.1365-2141.2010.08367.x

26. Yuan S, Yu J, Haverkos B, Su H. Remission of an extensively pre-treated relapsing/refractory ALK-positive anaplastic large cell lymphoma following metronomic therapy. *Leuk Lymphoma* (2016) 57:1194–6. doi: 10.3109/10428194.2015.1055484

27. Coleman M, Martin P, Ruan J, Furman R, Niesvizky R, Elstrom R, et al. Prednisone, etoposide, procarbazine, and cyclophosphamide (PEP-c) oral combination chemotherapy regimen for recurring/refractory lymphoma: low-dose metronomic, multidrug therapy. *Cancer* (2008) 112:2228–32. doi: 10.1002/cncr.23422

28. Cheson BD, Fisher RI, Barrington SF, Cavalli F, Schwartz LH, Zucca E, et al. Recommendations for initial evaluation, staging, and response assessment of Hodgkin and non-Hodgkin lymphoma: the lugano classification. *JCO* (2014) 32:3059–67. doi: 10.1200/JCO.2013.54.8800

29. Houot R, Poeschel V, Altmann B, Angel S, Thurner L, Illmer T, et al. Prolonged remissions after nivolumab plus Gemcitabine/Oxaliplatin in Relapsed/Refractory T-cell lymphoma. *Hemasphere* (2022) 6:e672. doi: 10.1097/HS9.0000000000000672

30. Wist E, Risberg T. Trofosfamide in non-hodgkin's lymphoma. a phase II study. *Acta Oncol* (1991) 30:819–21. doi: 10.3109/02841869109091828

31. Andersson PO, Braide I, Nilsson-Ehle H. Trofosfamide as salvage therapy for anaplastic large cell lymphoma relapsing after high-dose chemotherapy. *Leuk Lymphoma* (2002) 43:2351–3. doi: 10.1080/1042819021000040053

32. Pasmantier MW, Coleman M, Silver RT. Chemotherapy of the non-hodgkin's lymphomas. *Med Clin North Am* (1976) 60:1043–52. doi: 10.1016/s0025-7125(16)31847-8

33. Xiong Y-Y, Wang J, Wang L, Chen J-B, Liu L, Tang X-Q, et al. Comparison of CEAC, BEAM and IEAC conditioning regimens followed by autologous stem cell transplantation in peripheral T-cell lymphoma patients. *Sci Rep* (2022) 12:14369. doi: 10.1038/s41598-022-18540-x

34. Pro B, Advani R, Brice P, Bartlett NL, Rosenblatt JD, Illidge T, et al. Brentuximab vedotin (SGN-35) in patients with relapsed or refractory systemic anaplastic large-cell lymphoma: results of a phase II study. *J Clin Oncol* (2012) 30:2190–6. doi: 10.1200/JCO.2011.38.0402

35. Younes A, Bartlett NL, Leonard JP, Kennedy DA, Lynch CM, Sievers EL, et al. Brentuximab vedotin (SGN-35) for relapsed CD30-positive lymphomas. *N Engl J Med* (2010) 363:1812–21. doi: 10.1056/NEJMoa1002965

36. Wong J, Gruber E, Maher B, Waltham M, Sabouri-Thompson Z, Jong I, et al. Integrated clinical and genomic evaluation of guadecitabine (SGI-110) in peripheral T-cell lymphoma. *Leukemia* (2022) 36:1654–65. doi: 10.1038/s41375-022-01571-8

37. André N, Carré M, Pasquier E. Metronomics: towards personalized chemotherapy? *Nat Rev Clin Oncol* (2014) 11:413–31. doi: 10.1038/nrclinonc.2014.89

38. André N, Tsai K, Carré M, Pasquier E. Metronomic chemotherapy: direct targeting of cancer cells after all? *Trends Cancer* (2017) 3:319–25. doi: 10.1016/j.trecan.2017.03.011

39. Bocci G, Kerbel RS. Pharmacokinetics of metronomic chemotherapy: a neglected but crucial aspect. *Nat Rev Clin Oncol* (2016) 13:659–73. doi: 10.1038/nrclinonc.2016.64

40. Kerbel RS, Kamen BA. The anti-angiogenic basis of metronomic chemotherapy. *Nat Rev Cancer* (2004) 4:423–36. doi: 10.1038/nrc1369

41. Leone G, Pagano L, Ben-Yehuda D, Voso MT. Therapy-related leukemia and myelodysplasia: susceptibility and incidence. *Haematologica* (2007) 92:1389–98. doi: 10.3324/haematol.11034

42. Eichenauer DA, Thielen I, Haverkamp H, Franklin J, Behringer K, Halbsguth T, et al. Therapy-related acute myeloid leukemia and myelodysplastic syndromes in patients with Hodgkin lymphoma: a report from the German Hodgkin study group. *Blood* (2014) 123:1658–64. doi: 10.1182/blood-2013-07-512657

43. Odejide O, Weigert O, Lane AA, Toscano D, Lunning MA, Kopp N, et al. A targeted mutational landscape of angioimmunoblastic T-cell lymphoma. *Blood* (2014) 123:1293–6. doi: 10.1182/blood-2013-10-531509

frontiers | Frontiers in Oncology

TYPE Original Research
PUBLISHED 15 June 2023
DOI 10.3389/fonc.2023.1171048

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Xun Sun,
Huazhong University of Science and Technology, China
Rui Guo,
Shanghai Jiao Tong University, China

*CORRESPONDENCE
Dasheng Qiu
hbpetct@163.com
Xiaoyan Hu
hxyhelen0702@163.com

RECEIVED 21 February 2023
ACCEPTED 05 June 2023
PUBLISHED 15 June 2023

CITATION
Hu L, Luo N, Li L, Qiu D and Hu X (2023) A preliminary investigation of the relationship between ^{18}F-FDG PET/CT metabolic parameters and prognosis in angioimmunoblastic T-cell lymphoma. *Front. Oncol.* 13:1171048. doi: 10.3389/fonc.2023.1171048

A preliminary investigation of the relationship between ^{18}F-FDG PET/CT metabolic parameters and prognosis in angioimmunoblastic T-cell lymphoma

Lanping Hu, Nana Luo, Lei Li, Dasheng Qiu* and Xiaoyan Hu*

Department of Nuclear medicine, Hubei Cancer Hospital, The Seventh Clinical School Affiliated of Tongji Medical College, Huazhong University of Science and Technology, Wuhan, Hubei, China

Purpose: The goal of the study was to determine the prognostic significance of metabolic parameters in baseline ^{18}F-FDG PET/CT images obtained from patients with angioimmunoblastic T-cell lymphoma (AITL).

Methods: Forty patients with pathologically diagnosed AITL who had baseline ^{18}F-FDG PET/CT between May 2014 and May 2021 were assessed as part of this study. Maximum standardized uptake value (SUVmax), total lesion glycolysis (TLG), and total metabolic tumor volume (TMTV) were obtained and analyzed. In addition, many relevant features were evaluated, including sex, age, staging, International Prognostic Index (IPI), prediction index for T-cell lymphoma (PIT), Ki-67, and so on. Estimates of progression-free survival (PFS) and overall survival (OS) were determined using the log-rank test and Kaplan-Meier.

Results: The median follow-up was 30.2 months (interquartile range 9.82-43.03). Throughout the follow-up period, 29 (72.5%) deaths occurred and 22 (55.0%) patients made progress. The rates for 2- and 3-year PFS were 43.6% and 26.4%, respectively. The 3- and 5-year OS were 42.6% and 21.5%. For TMTV, TLG, and SUVmax, the cut-off values were 87.0 cm3, 711.1, and 15.8, respectively. Poorer PFS and OS were substantially correlated with high SUVmax and TLG. An increased TMTV suggested a shorter OS. TLG performed independently as OS predictors in multivariate analysis. The risk score for predicting the prognosis of AITL includes the TMTV, TLG, SUVmax, and IPI scores, with 4.5 for TMTV, 2 for TLG, 1.5 for IPI scores, and 1 for SUVmax. Three risk categories of patients with AITL had 3-year OS rates of 100.0%, 43.3%, and 25.0%, respectively.

Conclusion: Baseline TLG was a strong predictor of OS. Here a new prognostic scoring system for AITL based on the clinical indicators and PET/CT metabolic parameters was constructed, which might make stratification of prognosis easy and also help to individualize treatment.

KEYWORDS

Angioimmunoblastic T-cell lymphoma, PET/CT, metabolic parameters, prognosis, ^{18}F-FDG

Introduction

Angioimmunoblastic T-cell lymphomas (AITL), a distinct subtype of peripheral T-cell lymphomas (PTCL), account for between 18 and 36% of PTCL (1). Uncertainty surrounds its pathophysiology. Common T-cell antigens including CD2, CD3, CD5, and CD4 were also strongly expressed in AITL, pointing to the presence of T follicular helper (TFH) markers. TFH cells were also demonstrated to be the counterpart cell type to AITL (2–4). Many studies have suggested that immunological abnormalities brought on by viral infections (e.g. Epstein-barr virus) may be related to the pathophysiology (5, 6). The majority of AITL patients are elderly; at the time of diagnosis, most are already in clinical stages III or IV and don't present any symptoms in the early stages. Previous studies found OS was approximately 32% at 5 years (7). Previous studies have shown that AITL with poorer prognosis may be associated with overexpression of some specific genes such as IDH2, TET2, FYN, and CD2 (8, 9). For the purpose of directing treatment plans, it is essential to identify which patients would relapse earlier at the time of diagnosis. The prognosis of lymphoma is closely related to multiple factors such as age, stage, symptoms, and physical status. According to these prognostic influencing factors, different prognostic scoring systems have been established for different pathological types of lymphoma. The basis for determining the prognosis of PTCL in clinical and research settings is the International Prognostic Index (IPI), which includes five markers of age, clinical stage, physical condition, lactate dehydrogenase (LDH), and extra-nodal involvement. Based on the IPI score, the prediction index for T-cell lymphoma (PIT), which takes into account bone marrow involvement, age, physical condition, and LDH, was created. However, due to the heterogeneity of AITL, these two indices remain insufficient for early risk stratification (10). Only a few studies have particularly focused on prognostic prediction models for patients with AITL up to this point, such as the AITL prognostic index and the AITLI model, although it is uncertain how applicable these new AITL prognostic models will be in the future (11–14). Therefore, an effective prediction model for AITL prognosis is yet to be researched and developed.

Functional imaging and anatomical imaging are combined in PET/CT with ^{18}F-FDG as the tracer, which is currently often utilized in clinical practice for lymphoma staging, determining effectiveness, and prognostic evaluation. It has been shown that tumor load before initial treatment is of significant value in predicting prognosis. To assess the tumor load, PET/CT can quantify the volume of all positive lesions and determine the intensity of ^{18}F-FDG uptake within the lesion. The tumor metabolic volume (MTV) and total focal glycolysis (TLG) of high-uptake lymphomas such as diffuse large B-cell lymphoma (DLBCL), peripheral T-cell lymphoma (PTCL), and Hodgkin lymphoma (HL) can help predict patient survival (15, 16). Since there are currently few studies on the use of PET/CT metabolic parameters to assess AITL prognosis, the goal of this study was to determine the predictive value of PET/CT multiparametric indices for patient prognosis prior to initial treatment.

Materials and methods

Patients

Between May 2014 and May 2021, a total of 60 patients with AITL had baseline ^{18}F-FDG PET/CT evaluation; 40 patients were subsequently recruited after receiving full clinical follow-up and imaging data. 18 years of age or older, lymph node biopsy performed in accordance with 2008 WHO criteria to determine the diagnosis of AITL, patients who underwent ^{18} F-FDG PET/CT prior to receiving any treatment, refraining from using a colony-stimulating factor, glucocorticoids, or any other drugs that promote extramedullary hematopoiesis one week before imaging, and positive lesions on PET scans were the inclusion criteria. The following were listed as exclusion criteria: (1) prior or concurrent malignancies; (2) insufficient clinical, follow-up, and imaging data; (3) poor image quality; (4) severe cardiovascular disease; and (5) infection or chronic inflammation in the acute phase. Sex, age, staging, initial symptoms, ECOG-PS, bone marrow biopsy, IPI, PIT, LDH, beta 2-microglobulin (β2-MG), and Ki-67 labeling index were among the clinical and pathological parameters that were gathered and evaluated. 80% of patients were treated with CHOP or ECHOP chemotherapy regimens. The hospital ethics committee gave its approval for the trial, and patients gave their informed consent.

Instruments and methods

A GE Discovery STE PET/CT scanner was used to generate ^{18}F-FDG PET/CT pictures, and an American-made GE cyclotron was used to autonomously synthesize ^{18}F-FDG. All had radiochemical purity levels above 95%. Prior to receiving FDG injections, patients fasted for over 6 hours. Before the test, a tri-directional tube was used to inject ^{18}F-FDG (5.5 MBq/kg) intravenously, and the patient was given an hour to rest. Whole-body PET/CT imaging consisted of a PET scan of the entire body (2 minutes for each bed, totaling 6–8 beds) and a CT scan (200mA and 120 kV). PET scan adopted 3D acquisition. The body was scanned from the top of the skull to the middle of the thigh bone, and if necessary, the extremities. The ordered subsets expectation maximization (OSEM) approach was used to reconstruct the PET image, and the CT scanning data was used to adjust for image attenuation. The conventional approach was used to reconstruct the CT picture, and the layer thickness was 3.75mm. For frame-to-frame image parallel fusion presentation on Xeleris and AW workstations, PET and CT images were sent.

Image analysis

All PET/CT images were outlined and analyzed using Advantage Workstation 4.6 by two experienced nuclear medicine diagnosticians who were blinded to the patient's clinical outcome, with reference to a third nuclear medicine physician (associate director and above) for decision in case of disagreement. Semi-automated measurements were used to depict the volume of the

area of interest (VOI) and determine lesion boundaries, and the lesion's extent was then carefully adjusted to rule out inflammatory or physiological uptake in the brain, urinary system, and intestine. All involved lymph nodes and extra-nodal lesions were included. Thymus, nasopharynx, pharyngeal lymphatic ring, and spleen were intra-nodal lesions. Spleen SUVmax to liver background ratio greater than 1.5 or spleen longitudinal diameter greater than 13 cm was considered as splenic infiltration. Using the 41% SUVmax threshold approach, MTV was computed in accordance with the guidelines provided by the European Society of Nuclear Medicine. Only when bone marrow involvement was verified by BMB were volumetric measures taken. TLG was calculated by adding the products of the MTV and SUV averages of all lesions, and TMTV was calculated by adding the volumes of all hyper-metabolic lesions.

Statistical analysis

SPSS 23. 0 software (IBM, Chicago, IL, USA) and GraphPad Prism version 9.0 software (San Diego, CA, USA) were used to conduct the statistical analysis and drawing. Continuous variables were represented by their median or mean ± standard deviation, whereas categorical variables were portrayed as counts. The Pearson chi-squared test or Fisher's exact test was used to examine differences between subgroups. Receiver-operating characteristic (ROC) analysis was used to determine the ideal critical values for PET/CT metabolic parameters. PFS and OS were calculated using the log-rank test and Kaplan-Meier survival curves. The Cox proportional hazards regression model was utilized for multivariate survival analysis. Each relevant prognostic factor in the univariate analysis was given a N score, based on its HR value, to calculate the AITL prognostic score. Each factor's HR value was first rounded to obtain an integer and then divided by two to obtain the N score. For example, if HR is equal to 5.6, then N is equal to half of 6, which is equal to 3. The sum of N for each factor was defined as the AITL risk score. $p<0.05$ was considered to be statistically significant.

Results

Patients

This study involved forty patients. The average age of the patients was 60.3 years old, and the ratio of males to females was 3 to 1. At initial diagnosis, 36 (90.0%) patients were in stage III or stage IV, and 4 (10.0%) patients were in stage II. Twenty-five (62.5%) patients had extranodal involvement: skin (16 cases), parotid glands (14 cases), lungs (6 cases), bone marrow (5 cases), liver (2 cases), and gastrointestinal tract (1 case). In addition, splenic infiltration was present in 18 cases (45.0%). LDH greater than 400u/L and β2-MG greater than 3mg/L were considered elevated. Table 1 displays the clinical features of the patients.

ROC curves analysis of PET/CT metabolic multiparameter and Ki67

The median values of TMTV, TLG, SUVmax, and Ki67 were 249.3 (interquartile range 96.4-527.0) cm^3, 1300.1 (interquartile range 552.8-2925.0), 14.5 (interquartile range 9.9-23.8), and 55% (interquartile range 40.0-67.5%), respectively. ROC curve analysis was used to determine the cutoff values for SUVmax, TMTV, TLG, and Ki67. If the area under the curve (AUC) < 0.6, the grouping used the median as the cutoff value.

The ROC curve for each factor was outlined by assigning the deceased patients to the positive event group and the remaining patients to the negative event group as of the follow-up cutoff time. The SUVmax, TMTV, and TLG AUCs were, respectively, 0.770, 0.818, and 0.815. The cut-off values for TMTV, TLG, and SUVmax were 87.0 cm^3 (sensitivity 96.6%, specificity 63.6%, p = 0.002, Youden index of 0.602), 711.1 (sensitivity 79.3%, specificity 72.7%, p = 0.002, Youden index of 0.521), and 15.8 (sensitivity 55.2%, specificity 90.9%, p = 0.009, Youden index of 0.461). Due to Ki67's AUC being less than 0.6, the median expression of Ki67 (55%) was utilized as the dividing line for grouping.

Intergroup comparison of clinical characteristics

Patients were divided into groups with high TMTV (>87.0 cm3) and low TMTV (87.0 cm3) values, as well as groups with high TLG (>711.1) and low TLG (711.1) values. Patients with progressive disease or death at the follow-up cutoff were included in the poor prognosis group. High TLG was usually associated with higher Ann Arbor staging (p = 0.011), IPI score (p = 0.002), PIT score (p = 0.005), Ki67 index (p = 0.048), and elevated LDH (p = 0.012). The frequency of a bad prognosis was statistically substantially greater in the high-risk TMTV group than in the low-risk group (p=0.037), while the difference in TLG between patients with a poor and good prognosis was not statistically significant (p=0.082) (Table 2).

Survival analysis for clinical and PET/CT metrics

The median follow-up time was 30.2 (interquartile range 9.82 to 43.03) months. PFS and OS times were respectively 10.5 (95% CI 8.4-12.6) and 32.8 (95% CI 24.10-41.51) months in the median. Of these patients, 22 (55.0%) patients experienced disease progression (excluding death) during the course of treatment and 29 (72.5%) died. The rates for PFS at 2 and 3 years were 43.6% and 26.4%, respectively, while the rates for OS at 3 and 5 years were 42.6% and 21.5%, respectively.

The 2-year PFS rates for the high and low TMTV groups were 37.5% and 66.7%, respectively, according to Kaplan-Meier curves and log-rank testing (p = 0.090). The 3-year OS rates for the high and low TMTV groups were, respectively, 30.1% and 83.3% (p = 0.009). Those with higher TMTV had a median OS of 25.5 months.

10.3389/fonc.2023.1171048

TABLE 1 Characteristics of patients.

Characteristics	No. (%)
Sex/male	30 (75.0)
Age, mean (range, year)	60.25 (33-80)
B symptoms/yes	22 (55.0)
Ann Arbor stage	
II	4 (10.0)
III	20 (50.0)
IV	16 (40.0)
ECOG-PS >1	13 (32.5)
Elevated LDH	30 (75.0)
Elevated β2-MG	28 (70.0)
BMB positive	5 (12.5)
Extanodal involvement/yes	25 (62.5)
Extranodal sites >1/yes	14 (35.0)
IPI	
0-1	5 (12.5)
2-3	22 (55.0)
4-5	13 (32.5)
PIT	
0	4 (10.0)
1	15 (37.5)
2	11 (27.5)
3	9 (22.5)
4	1 (2.5)

ECOG-PS, Eastern Cooperative Oncology Group Performance Status; LDH, lactate dehydrogenase; β2-MG, β2-microglobulin; BMB, bone marrow biopsy; IPI, international prognostic index; PIT, prognostic index for T-cell lymphoma.

In the high and low TLG groups, the 2-year PFS rates were 34.6% and 60.0%, respectively (p = 0.042), and the 3-year OS rates were 33.5% and 64.2%, respectively (p = 0.003). The median OS for patients with higher TLG was 11.4 months. In the high and low SUVmax groups, the 2-year PFS rates were, respectively, 29.4% and 54.7% (p = 0.040), and the 3-year OS rates were 23.5% and 57.6%, respectively (p = 0.018). Those with higher SUVmax had a median OS of 16.7 months. Univariate survival analysis revealed that extranodal involvement sites greater than 1, TLG greater than 711.1, and SUVmax greater than 15.8 were risk factors for both PFS and OS, but that high TMTV was only a risk factor for OS and not PFS (Table 3; Figure 1).

There was a strong correlation between TMTV and TLG (r = 0.868, p < 0.001) by Spearman's rank correlation test; therefore, TMTV or TLG were included in the multivariate analysis with other clinical characteristics at p < 0.05 in the univariate analysis, respectively. TLG was a predictive factor for OS independently (HR 3.32, 95% CI 1.080-9.582, p=0.036), while TMTV tended to be an independent OS predictor (p=0.055) (Table 4).

Construction of AITL risk score and prognostic predictive efficacy

According to univariate analysis, TMTV, TLG, SUVmax, IPI score, and more than one extra-nodal involvement site were predictive variables for OS, and extra-nodal involvement sites >1 was an indicator of IPI score, so it was not included in the risk score. N points were allocated to each element in accordance with the AITL risk score methodology, with 4.5 scores for TMTV (HR: 8.960); 2 scores for TLG (HR: 4.007); 1.5 scores for IPI scores (HR: 2.542); and 1 score for SUVmax (HR: 2.413). Therefore, the AITL risk score for this group of patients ranged from 0 to 9. The AUC of the AITL risk score obtained by ROC curve analysis was 0.903 (Figure 2).

Next, patients were classified into three groups depending on their AITL risk scores: low-risk group (score 0–3), medium-risk group (score 4.5–7.5), and high-risk group (score 8–9). In the three groups, the 3-year OS rates were 100.0%, 43.3%, and 25.0%, respectively (p=0.001) (Figure 3). Kaplan-Meier survival analysis of IPI scores versus PIT scores showed a significant difference between the low-risk and high-risk groups in the patient groups by IPI (P < 0.05), while AUC = 0.746 was obtained by ROC curve analysis (Figure 4A). In the group of patients by PIT, there was no significant difference between the low-risk and high-risk groups (P > 0.05) (Figure 4B). These analyses suggest that neither IPI nor PIT showed good prognostic performance in patients with AITL.

Discussion

Current status of clinical assessment of prognosis in AITL

A rare form of PTCL, AITL is marked by quick disease progression and a dismal prognosis. A large-scale population-based analysis using the SEER database was carried out by Xu and Liu et al. to elucidate the temporal survival trends and prognostic variables of AITL patients (17). A total of 1207 AITL patients were enrolled in this study, and their respective OS rates at 2, 5, and 10 years were 46.8%, 32.9%, and 21.9%. The 3-year OS rate and 5-year OS rate in our retrospective analysis, respectively, were 42.6% and 21.5%. (95% CI: 24.092-41.508; median OS: 32.8 months). The 5-year survival rate for patients with AITL was less than 40%, according to studies conducted so far (18), and the current investigation supports this conclusion. Patients with AITL have poorer outcomes than patients with aggressive B-cell lymphoma and may relapse early, but a minority of patients are able to survive long-term or even be cured. Hence, it is crucial to accurately determine the prognosis of patients with AITL and to more accurately distinguish between those who are in danger and those who may be able to receive a cure.

Recently, some studies have analyzed the relationship between clinical features and the prognosis of AITL. In a retrospective investigation of 207 AITL patients, Tokunaga et al. discovered that age over 60 years, increased leukocyte and IgA levels, anemia

10.3389/fonc.2023.1171048

TABLE 2 Comparisons of clinicopathologic characteristics according to TMTV and TLG.

		TMTV			TLG		
	No.	Low	High	P	Low	High	P
Age, >60/≤60	21/19	5/3	16/16	0.408	6/8	15/11	0.286
Sex, male/female	30/10	4/4	26/6	0.089	10/4	10/6	0.492
B symptoms, yes/no	22/18	4/4	18/14	0.528	7/7	15/11	0.446
Ann Arbor stage, II/III-IV	4/36	3/5	1/31	0.020*	4/10	0/26	0.011*
ECGO-PS, 0-1/2-5	27/13	6/2	21/11	0.479	12/2	15/11	0.071
LDH, elevated/normal	30/10	4/4	26/6	0.089	7/7	23/3	0.012*
β2-MG, elevated/normal	28/12	4/4	24/8	0.170	7/7	21/5	0.049*
BMB, negative/positive	5/35	1/7	4/28	0.694	1/13	4/22	0.418
Extranodal sites>1, yes/no	13/27	1/7	12/20	0.179	4/10	9/17	0.491
IPI, 0-2/3-5	15/25	5/3	10/22	0.112	10/4	5/21	0.002*
PIT, 0-1/2-4	19/21	5/3	14/18	0.290	11/3	8/18	0.005*
Ki67, >55%/≤55%	20/20	4/4	16/16	0.653	4/10	16/10	0.048*
Poor prognosis, yes/no	32/8	4/4	28/4	0.037	9/5	23/3	0.082

*Statistically significant. ECOG-PS, Eastern Cooperative Oncology Group Performance Status; BMB, bone marrow biopsy; IPI, International Prognostic Index; PIT, prognostic index for T-cell lymphoma.

and thrombocytopenia, and more than one site of extra-nodal involvement were important predictive factors for OS, and for PFS, mediastinal lymph node metastasis, increased IgA, and anemia were significant prognostic factors (13). It is still debatable if these clinical signs have any real impact on the prognosis of AITL. The IPI score is widely used as a predictive model for various non-Hodgkin's lymphomas, and the recently proposed PIT score can also stratify or predict the risk of PTCL. But

TABLE 3 Univariate analysis for survivals.

	PFS			OS		
	HR	95%CI	P	HR	95%CI	P
Sex (male)	1.72	0.702-4.219	0.235	1.436	0.579-3.562	0.436
Age>60	1.45	0.707-2.961	0.313	1.224	0.581-2.576	0.595
B symptoms	0.67	0.327-1.355	0.262	0.738	0.347-1.567	0.429
Ann Arbor stag (III/IV)	0.04	0.000-2.995	0.143	0.040	0.000-5.222	0.195
ECGO>1	1.51	0.728-3.116	0.270	1.286	0.590-2.804	0.527
Elevated LDH	1.064	0.453-2.498	0.887	1.589	0.588-4.294	0.362
Elevated β2-MG	1.84	0.789-4.264	0.158	1.942	0.784-4.810	0.151
IPI>2	2.20	0.981-4.926	0.056	2.542	1.077-6.003	0.033
PIT>1	1.728	0.836-3.570	0.140	0.195	0.773-3.544	0.195
BMB/positive	1.44	0.707-2.910	0.317	1.222	0.417-3.581	0.715
Extranodal sites >1	2.586	1.224-5.463	0.013	2.502	1.118-5.599	0.026
TMTV>87.0	2.414	0.842-6.921	0.101	8.960	1.216-65.988	0.031
TLG>711.1	2.263	1.005-5.093	0.049	4.007	1.516-10.587	0.005
SUVmax>15.8	2.068	1.017-4.208	0.045	2.413	1.134-5.133	0.022
Ki-67>55%	1.029	0.507-2.091	0.936	0.986	0.468-2.076	0.970

IPI, international prognostic index; PIT, prognostic index for T-cell lymphoma, ECOG-PS, Eastern Cooperative Oncology Group Performance Status; LDH, lactate dehydrogenase; β2-MG, β2-microglobulin, BMB, bone marrow biopsy, SUVmax, maximum standard uptake value; TMTV, total metabolic tumour volume, TLG, total lesion glycolysis.

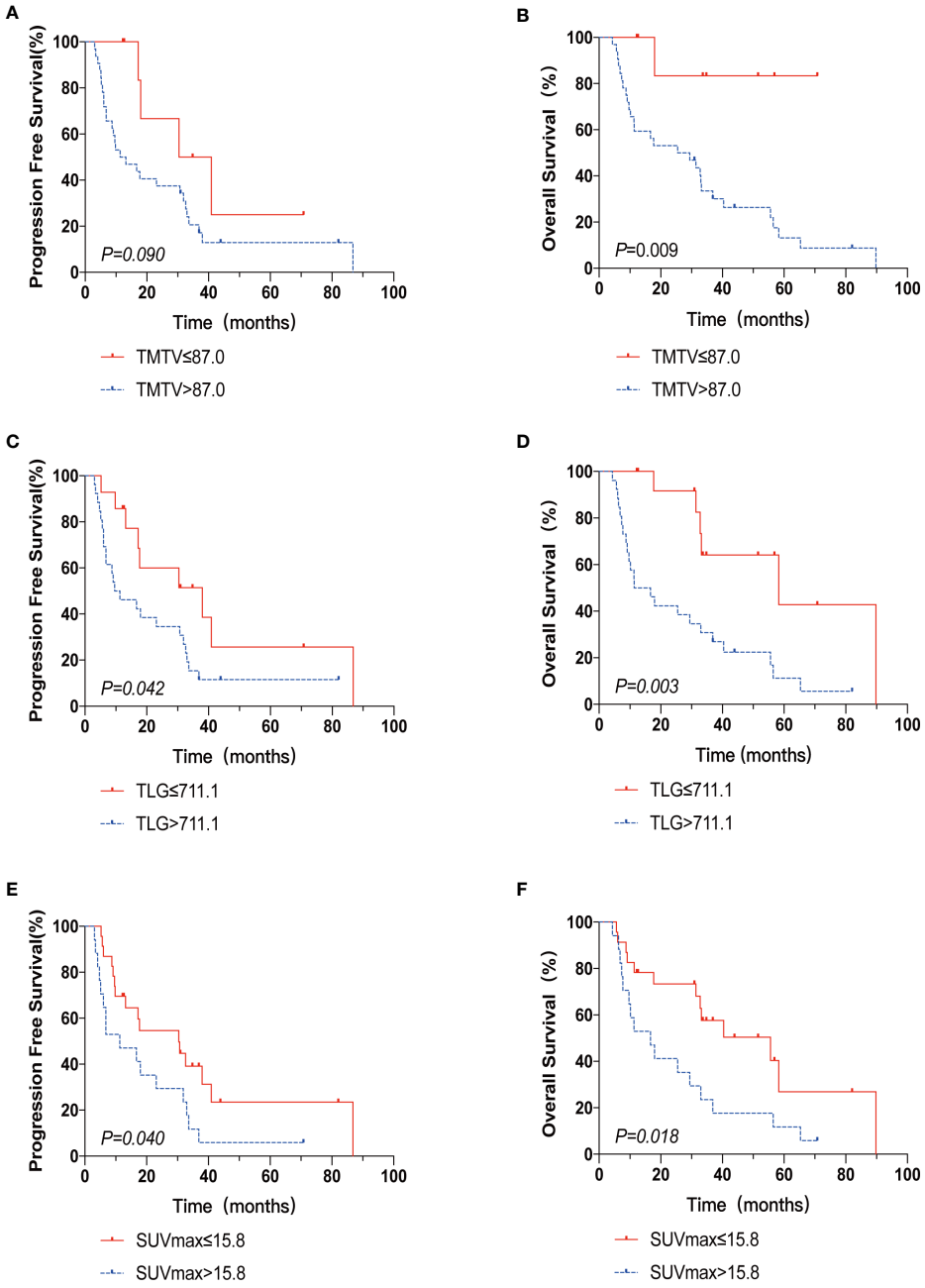


FIGURE 1
The Kaplan-Meier survival analysis and curves of OS and PFS according to TMTV **(A, B)**, TLG **(C, D)**, and SUVmax **(E, F)**. p value was acquired by the log-rank.

several investigations have demonstrated that the IPI and PIT were insufficient as indicators of prognosis in individuals with AITL (10, 12, 19, 20).

The value of ^{18}FDG-PET/CT in lymphoma prognosis prediction

^{18}F-FDG PET/CT plays an important role in the diagnosis and treatment of malignant lymphoma, and the calculation of TMTV as well as TLG allows the quantification of the total tumor metabolic activity volume. The International Society for Malignant Lymphoma stated that metabolic parameters of ^{18}F-FDG PET/CT could be used in the prognostic analysis of lymphoma (15). Several large retrospective or prospective studies have shown that multiple metabolic parameters of PET/CT (including TMTV and TLG) affect the survival prognosis of HL, DLBCL, and T-cell lymphoma subtypes (21, 22). As a result, there is an increasing interest in TMTV and TLG, sometimes in combination with clinical parameters to assess prognosis (23). In order to assess the prognosis of AITL or PTCL, Gong, Cottereau, and Jiang et al. used TMTV and PIT scores. They demonstrated that the two

TABLE 4 Multivariate analysis for survivals.

	PFS				OS		
	HR	95%CI	P		HR	95%CI	p
TMTV*							
TMTV	2.08	0.687-5.955	0.201	TMTV	7.19	0.957-54.00	0.055
SUVmax	1.82	0.881-3.796	0.105	SUVmax	1.70	0.808-4.985	0.133
Extranodal sites >1	2.08	0.996-4.500	0.061	IPI>2	1.30	0.442-3.673	0.655
				Extranodal sites >1	1.93	0.874-4.663	0.100
TLG*							
TLG	1.74	0.714-4.245	0.223	TLG	3.22	1.080-9.582	0.036
SUVmax	1.51	0.689-3.306	0.303	SUVmax	1.44	0.581-3.547	0.434
Extranodal sites >1	2.23	1.045-4.743	0.038	IPI>2	1.12	0.376-3.316	0.843
				Extranodal sites >1	2.35	0.989-5.598	0.053

PFS, progression-free survival; OS, overall survival; HR, hazard ratio; CI, confidence interval; TMTV, total metabolic tumour volume; TLG, total lesion glycolysis.
*TMTV and TLG were separately incorporated into multivariate survival analysis due to the correlation between TMTV and TLG.

factors together may more effectively predict the prognosis of patients (24–26).

We looked into the prognostic value of a variety of metabolic parameters of pretreatment PET/CT. In multivariate survival analysis, we found that TLG was an independent prognostic factor for OS (HR=3.22, 95% CI%:1.080-9.582, p=0.036), and TMTV showed a trend as an independent predictor of OS (HR=7.19, 95% CI%:0.957-54.00, p=0.55). According to the analysis of PET/CT metabolic characteristics of 56 patients before treatment by Gong et al., TMTV was a single factor affecting PFS and OS in AITL patients (24). In a multicentre retrospective study of 140 patients with PTCL in the lymph nodes, Cottereau et al. found that baseline TMTV was the only independent variable considered significant in PFS and OS (25). Zhou et al. believed that baseline TMTV and TLG were independent predictors of PFS and OS (27). Pak et al. found that TLG elevation was most significant in a multicenter trial involving 36 patients with extranodal NK/T cell lymphoma (28). We hypothesized that this contradiction was due to the high correlation between TLG and TMTV and that including both in a multivariate analysis would lead to an incorrect assessment. Furthermore, it was found that the optimal cut-off values for classifying patients into high- and low-risk populations differed depending on studies, which correlated with the clinical features of the study population (volume range, treatment effect, etc.). This implies that the ideal cut-off values for risk prediction using TMTV and TLG may be unique to certain patient traits, lymphoma subtypes, and treatments.

The most common index for determining the amount of ^{18}F-FDG uptake, SUVmax, indicates the tumor's most aggressive

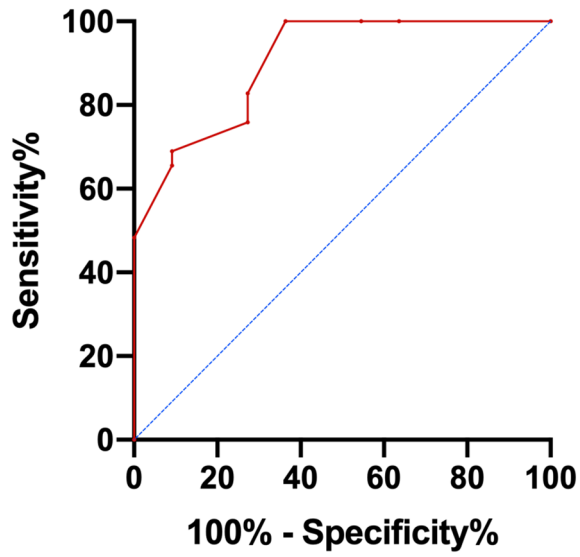


FIGURE 2
ROC curve analysis of AITL risk score.

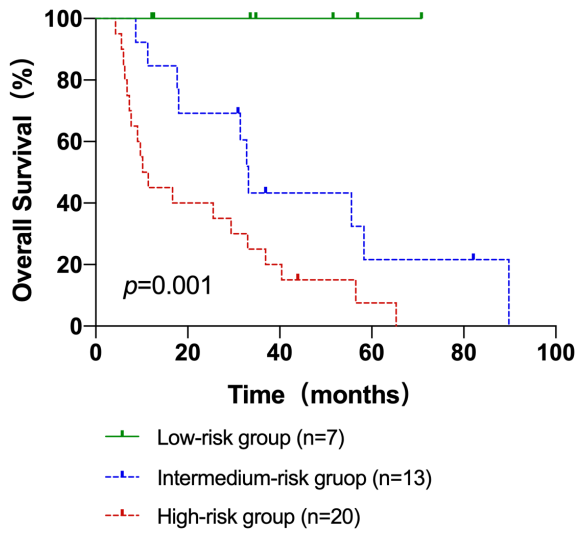


FIGURE 3
The Kaplan–Meier survival analyses and curves of OS according to the AITL risk scores. p value was acquired by the log-rank test.

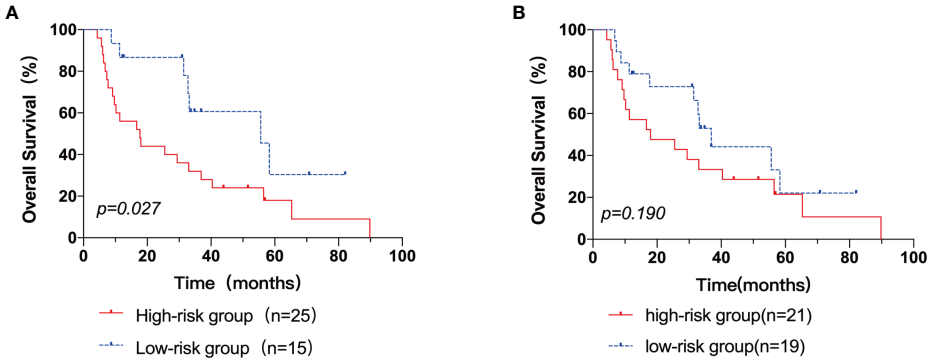


FIGURE 4
The Kaplan–Meier survival analyses and curves of OS according to the IPI score **(A)** and PIT score **(B)**. p value was acquired by the log-rank test.

cellular component's glycolytic metabolism, and studies have linked SUVmax to tumor aggressiveness (29). The predictive significance of SUVmax before therapy is still debatable, though. By using univariate analysis, the current study demonstrated a substantial correlation between SUVmax and prognosis, but it did not serve as an independent predictor of AITL (p>0.05). The findings of Gong and Wang were similar to ours, but there were also differences, which showed no significant prognostic value of SUVmax in both univariate and multivariate analyses (23, 30). The study by Jiang et al. did not find that SUVmax correlated with the prognosis of PTCL (25). The possible reasons we considered were as follow: firstly, SUVmax is susceptible to injection time, blood glucose levels, and partial volume effects. Focal FDG affinity is variable at baseline. A recent study used dynamic changes in SUVmax over the course of treatment to assess the predicted prognosis of lymphoma (31). Second, SUVmax represents only a portion of the volume of FDG uptake in a single lesion, whereas patients with AITL usually have multiple lesions. In addition, due to the large heterogeneity and prognostic differences between the different pathological subtypes of PTCL, the prognostic predictive role of SUVmax in PTCL did not lend itself to direct application to AITL.

Evaluation value of clinical features for prognosis in AITL

In this study, patients' clinical characteristics including age, sex, B symptoms, Ann Arbor stage, ECGO score, PIT score, and laboratory clinical indicators (LDH, β2-MG) were not linked with OS. IPI score correlated with OS (HR=2.542, 95%CI: 1.077-6.003, p=0.033), and extranodal involvement>1 was associated with OS and PFS in AITL (PFS: HR=2.586, 95%CI: 1.224-5.463, p=0.013; OS: HR=2.502, 95%CI: 1.118-5.599, p=0.026). Moreover, it served as a standalone predictive factor for PFS (HR = 2.230, 95% CI = 1.045–4.743, p = 0.038). However, the present study did not find the value of the PIT score in AITL prognosis (p>0.05). This contradicts some of the previous studies. On the one hand, it may be because the PIT includes 4 parameters, which has a confounding bias. On the other hand, the number of patients enrolled was small and there was selection bias. Previous studies have also shown that IPI and PIT scores for patients with AITL could not be used to predict survival, and even when they were taken into account in multivariate analyses, they had no appreciable impact on survival rates. The present study hypothesized that the clinical outcome of AITL is not so much a direct complication of tumor proliferation as a result of a severe regulatory disorder of the immune system.

Construction of an AITL risk score based on clinical indicators and PET/CT metabolic parameters

Based on clinical indices and PET/CT metabolic parameters, we innovatively constructed a risk score for prognosis prediction of AITL, including TMTV, TLG, SUVmax, and IPI score, and the prognosis of AITL patients was successfully stratified. Three groups of patients with AITL were created: a low-risk group, an intermediate-risk group, and a high-risk group. Patients in the low-risk group had considerably greater OS rates than those in the intermediate and high-risk groups, according to the Kaplan-Meier survival analysis. The three groups' respective 3-year OS rates were 100.0%, 43.3%, and 25.0% (2 = 14.639, p0.001). The novel prognosis scoring system has the ability to more accurately predict the prognosis of AITL, even if the IPI score and SUVmax were not independent prognostic markers for AITL and TMTV only showed a trend as an independent predictor of OS in this study (p>0.05). This study was unable to properly explore the effect of altering and shifting treatment patterns on the prognosis of patients with AILT due to the small sample size. This prognostic prediction score should be validated in future large sample and prospective studies, providing valuable information to optimize treatment decisions to benefit more AITL patients.

The limitation of the previous studies and differences of this study

Most previous studies have only explored the prognostic value of PET/CT metabolic parameters by retrospective analysis or

combined with clinical prognostic scores to construct prognostic models (24, 30). Due to the rarity of AITL, most studies currently only explored the prognostic value of PET/CT in PTCL as a whole, with AITL as only a subset of cases (27, 31, 32). Most studies on the prognosis of FDG-PET/CT in AITL were small sample studies, which means that unrecognized bias and the presence of overfitting cannot be avoided. The long time span of patients enrolled in the study and the inconsistency of first-line chemotherapy may cause bias, which is the limitation of this study. Here our study developed a new prognostic scoring system specifically designed for AITL based on clinical indicators and PET/CT parameters, which clearly defined risk groups in AITL patients and identified patients with relatively better prognosis, as compared to the existing prognostic models. Hence this novel prognostic model specially designed for AITL may facilitate risk-based stratification and therapy.

Future prospects

External data validation would be needed to verify the effectiveness of the new scoring system in the future. Several studies have verified the efficacy of intermediate FDG-PET as a prognostic indicator in lymphoma. 95 patients with PTCL participated in a recent study by Casulo et al. that examined intermediate FDG-PET/CT. This study showed that clinical outcomes could be predicted by measuring metabolic activity using intermediate FDG-PET/CT in PTCL (32). The value of interim FDG-PET for assessing the prognosis of AITL can be further explored in the future. Moreover, to combined predictive model of ^{18}F-FDG and clinicopathological characteristics is a promising research direction. More biomarkers need to be investigated in future studies and applied to the new AITL prognosis prediction model.

Conclusion

We concluded that baseline TMTV, TLG, and SUVmax were independent predictors of worse outcomes in AITL, while baseline TLG was an independent predictor of OS. We have developed a new prognostic scoring system specifically designed for AITL based on clinical indicators and PET/CT parameters, which may assist in clinical decision-making for AITL patients in clinical practice and also provide a basis for future research.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

The studies involving human participants were reviewed and approved by Hubei Cancer Hospital. The patients/participants provided their written informed consent to participate in this study.

Author contributions

LH participated in the design of the study, carried out analysis and interpretation of data, and drafted the manuscript; final approval of the version to be published and agree to be accountable for all aspects of the work. LH, NL, and LL involved in image analysis, participate in the discussion of the result of the part and final approval of the version to be published. DQ and XH gave conception and design of the study, participated in the image analysis, participate in the discussion of the results analysis, and approved the final submission. All authors read and approved the final manuscript. All authors contributed to the article and approved the submitted version.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

The reviewer XS declared a shared affiliation with the authors to the handling editor at the time of review.

Publisher's note

References

1. Armitage JO. The aggressive peripheral T-cell lymphomas: 2017. *Am J Hematol* (2017) 92(7):706–15. doi: 10.1002/ajh.24791

2. Matutes E. The 2017 WHO update on mature T- and natural killer (NK) cell neoplasms. *Int J Lab Hematol* (2018) 40:97–103. doi: 10.1111/ijlh.12817

3. Iqbal J, Weisenburger DD, Greiner TC, Vose JM, McKeithan T, Kucuk C, et al. Molecular signatures to improve diagnosis in peripheral T-cell lymphoma and prognostication in angioimmunoblastic T-cell lymphoma. *Blood* (2010) 115(5):1026–36. doi: 10.1182/blood-2009-06-227579

4. De Leval L, Rickman DS, Thielen C, Ad R, YL H, Delsol G, et al. The gene expression profile of nodal peripheral T-cell lymphoma demonstrates a molecular link between angioimmunoblastic T-cell lymphoma (AITL) and follicular helper T (TFH) cells. *Blood* (2007) 109(11):4952–63. doi: 10.1182/blood-2006-10-055145

5. Chiba S, Sakata-Yanagimoto M. Advances in understanding of angioimmunoblastic T-cell lymphoma. *Leukemia* (2020) 34(10):2592–606. doi: 10.1038/s41375-020-0990-y

6. Lunning MA, Vose JM. Angioimmunoblastic T-cell lymphoma: the many-faced lymphoma. *Blood* (2017) 129(9):1095–102. doi: 10.1182/blood-2016-09-692541

7. International T-Cell Lymphoma Project. International peripheral T-cell and natural Killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26(25):4124–30. doi: 10.1200/JCO.2008.16.4558

8. Odejide O, Weigert O, Lane AA, Toscano D, Lunning MA, Kopp N, et al. A targeted mutational landscape of angioimmunoblastic T-cell lymphoma. *Blood* (2014) 123(9):1293–6. doi: 10.1182/blood-2013-10-531509

9. Wang C, McKeithan TW, Gong Q, Zhang W, Bouska A, Rosenwald A, et al. IDH2 R172 mutations define a unique subgroup of patients with angioimmunoblastic T-cell lymphoma. *Blood* (2015) 126(15):1741–52. doi: 10.1182/blood-2015-05-644591

10. Mourad N, Mounier N, Brière J, Raffoux E, Delmer A, Feller A, et al. Clinical, biologic, and pathologic features in 157 patients with angioimmunoblastic T-cell lymphoma treated within the groupe d'Etude des lymphomes de l'Adulte (GELA) trials. *Blood* (2008) 111(9):4463–70. doi: 10.1182/blood-2007-08-105759

11. Advani RH, Skrypets T, Civallero M, Spinner MA, Manni M, Kim WS, et al. Outcomes and prognostic factors in angioimmunoblastic T-cell lymphoma: final report from the international T-cell project. *Blood* (2021) 138(3):213–20. doi: 10.1182/blood.2020010387

12. Federico M, Rudiger T, Bellei M, Nathwani BN, Luminari S, Coiffier B, et al. Clinicopathologic characteristics of angioimmunoblastic T-cell lymphoma: analysis of the international peripheral T-cell lymphoma project. *J Clin Oncol* (2013) 31(2):240–6. doi: 10.1200/JCO.2011.37.3647

13. Tokunaga T, Shimada K, Yamamoto K, Chihara D, Ichihashi T, Oshima R, et al. Retrospective analysis of prognostic factors for angioimmunoblastic T-cell lymphoma: a multicenter cooperative study in Japan. *Blood* (2012) 119(12):2837–43. doi: 10.1182/blood-2011-08-374371

14. Huang C, Zhang H, Gao Y, Diao L, Liu L. Development of a novel clinical prognostic model for patients with angioimmunoblastic T-cell lymphoma. *Technol Cancer Res Treat* (2020) 19:153303382096423. doi: 10.1177/1533033820964231

15. Barrington SF, Mikhaeel NG, Kostakoglu L, Meignan M, Hutchings M, Müeller SP, et al. Role of imaging in the staging and response assessment of lymphoma: consensus of the international conference on malignant lymphomas imaging working group. *J Clin Oncol* (2014) 32(27):3048–58. doi: 10.1200/JCO.2013.53.5229

16. Barrington SF, Kluge R. FDG PET for therapy monitoring in Hodgkin and non-Hodgkin lymphomas. *Eur J Nucl Med Mol Imaging* (2017) 44(S1):97–110. doi: 10.1007/s00259-017-3690-8

17. Xu B, Liu P. No survival improvement for patients with angioimmunoblastic T-cell lymphoma over the past two decades: a population-based study of 1207 cases. *PloS One* (2014) 9(3):e92585. doi: 10.1371/journal.pone.0092585

18. Li Y, Yang C, Mao L, Wang J, Li C, Qian W. Clinical characteristics of angioimmunoblastic T-cell lymphoma in China and c-reactive protein as an independent prognostic factor. *Medicine* (2017) 96(39):e8091. doi: 10.1097/MD.0000000000008091

19. Lin HN, Liu CY, Hong YC, Pai JT, Yang CF, Yu YB, et al. Clinical features and prognostic factors of angioimmunoblastic T-cell lymphoma in Taiwan: a single-institution experience. *Leuk Lymphoma* (2010) 51(12):2208–14. doi: 10.3109/10428194.2010.525270

20. Lee SS, Rüdiger T, Odenwald T, Roth S, Starostik P, Müller-Hermelink HK. Angioimmunoblastic T cell lymphoma is derived from mature T-helper cells with varying expression and loss of detectable CD4. *Int J Cancer* (2003) 103(1):12–20. doi: 10.1002/ijc.10758

21. Ceriani L, Milan L, Johnson PWM, Martelli M, Presilla S, Giovanella L, et al. Baseline PET features to predict prognosis in primary mediastinal b cell lymphoma: a comparative analysis of different methods for measuring baseline metabolic tumour volume. *Eur J Nucl Med Mol Imaging* (2019) 46(6):1334–44. doi: 10.1007/s00259-019-04286-8

22. Barrington SF, Meignan M. Time to prepare for risk adaptation in lymphoma by standardizing measurement of metabolic tumor burden. *J Nucl Med* (2019) 60(8):1096–102. doi: 10.2967/jnumed.119.227249

23. Shagera QA, Cheon GJ, Koh Y, Yoo MY, Kang KW, Lee DS, et al. Prognostic value of metabolic tumour volume on baseline 18F-FDG PET/CT in addition to NCCN-IPI in patients with diffuse large b-cell lymphoma: further stratification of the group with a high-risk NCCN-IPI. *Eur J Nucl Med Mol Imaging* (2019) 46(7):1417–27. doi: 10.1007/s00259-019-04309-4

24. Gong H, Li T, Li J, Tang L, Ding C. Prognostic value of baseline total metabolic tumour volume of 18F-FDG PET/CT imaging in patients with angioimmunoblastic T-cell lymphoma. *EJNMMI Res* (2021) 11(1):64. doi: 10.1186/s13550-021-00807-5

25. Cottereau AS, Becker S, Broussais F, Casasnovas O, Kanoun S, Roques M, et al. Prognostic value of baseline total metabolic tumor volume (TMTV0) measured on FDG-PET/CT in patients with peripheral T-cell lymphoma (PTCL). *Ann Oncol* (2016) 27(4):719–24. doi: 10.1093/annonc/mdw011

26. Jiang C, Teng Y, Chen J, Wang Z, Zhou Z, Ding C, et al. Baseline total metabolic tumor volume combined with international peripheral T-cell lymphoma project may improve prognostic stratification for patients with peripheral T-cell lymphoma (PTCL). *EJNMMI Res* (2020) 10(1):110. doi: 10.1186/s13550-020-00698-y

27. Zhou Y, Zhang X, Qin H, Zhao Z, Li J, Zhang B, et al. Prognostic values of baseline ^{18} f-FDG PET/CT in patients with peripheral T-cell lymphoma. *BioMed Res Int* (2020) 2020:1–10. doi: 10.1155/2020/9746716

28. Pak K, Kim BS, Kim K, Kim IJ, Jun S, Jeong YJ, et al. Prognostic significance of standardized uptake value on F18-FDG PET/CT in patients with extranodal nasal type NK/T cell lymphoma: a multicenter, retrospective analysis. *Am J Otolaryngol* (2018) 39(1):1–5. doi: 10.1016/j.amjoto.2017.10.009

29. Hutchings M, Loft A, Hansen M, Ralfkiaer E, Specht L. Different histopathological subtypes of Hodgkin lymphoma show significantly different levels of FDG uptake. *Hematol Oncol* (2006) 24(3):146–50. doi: 10.1002/hon.782

30. Wang H, Yu W, Wu T, Xue Y, Zhang D, Xu H. The incremental prognostic value of baseline ^{18} f-FDG PET/CT imaging in angioimmunoblastic T-cell lymphoma. *BioMed Res Int* (2020) 2020:1–7. doi: 10.1155/2020/4502489

31. Schmitz C, Rekowski J, Müller SP, Hertenstein B, Franzius C, Ganser A, et al. Baseline and interim PET-based outcome prediction in peripheral T-cell lymphoma: a subgroup analysis of the PETAL trial. *Hematol Oncol* (2020) 38(3):244–56. doi: 10.1002/hon.2697

32. Casulo C, Schöder H, Feeney J, Lim R, Maragulia J, Zelenetz AD, et al. 18F-fluorodeoxyglucose positron emission tomography in the staging and prognosis of T cell lymphoma. *Leuk Lymphoma* (2013) 54(10):2163–7. doi: 10.3109/10428194.2013.767901

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 23 June 2023
DOI 10.3389/fonc.2023.1202964

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Carlos Murga-Zamalloa,
University of Illinois Chicago, United States
Shih-Sung Chuang,
Chi Mei Medical Center, Taiwan
Kedar Inamdar,
Henry Ford Hospital, United States

*CORRESPONDENCE
Fanny Drieux
fanny.drieux@chb.unicancer.fr

RECEIVED 09 April 2023
ACCEPTED 22 May 2023
PUBLISHED 23 June 2023

CITATION
Drieux F, Lemonnier F and Gaulard P (2023) How molecular advances may improve the diagnosis and management of PTCL patients.
Front. Oncol. 13:1202964.
doi: 10.3389/fonc.2023.1202964

How molecular advances may improve the diagnosis and management of PTCL patients

Fanny Drieux[1]*, François Lemonnier[2,3] and Philippe Gaulard[3,4]

[1]Service d'Anatomie et de Cytologie Pathologiques, INSERM U1245, Centre Henri Becquerel, Rouen, France, [2]Unité hémopathies Lymphoïdes, Hôpitaux Universitaires Henri Mondor, Assistance Publique des Hôpitaux de Paris, Créteil, France, [3]Institut Mondor de Recherche Biomédicale, INSERM U955, Université Paris Est Créteil, Créteil, France, [4]Département de Pathologie, Hôpitaux Universitaires Henri Mondor, Assistance Publique des Hôpitaux de Paris, Créteil, France

Peripheral T-cell lymphomas (PTCL) comprised more than 30 rare heterogeneous entities, representing 10 to 15% of adult non-Hodgkin lymphomas. Although their diagnosis is still mainly based on clinical, pathological, and phenotypic features, molecular studies have allowed for a better understanding of the oncogenic mechanisms involved and the refinement of many PTCL entities in the recently updated classifications. The prognosis remains poor for most entities (5-year overall survival < 30%), with current conventional therapies based on anthracyclin-based polychemotherapy regimen, despite many years of clinical trials. The recent use of new targeted therapies appears to be promising for relapsed/refractory patients, such as demethylating agents in T-follicular helper (TFH) PTCL. However further studies are needed to evaluate the proper combination of these drugs in the setting of front-line therapy. In this review, we will summarize the oncogenic events for the main PTCL entities and report the molecular targets that have led to the development of new therapies. We will also discuss the development of innovative high throughput technologies that aid the routine workflow for the histopathological diagnosis and management of PTCL patients.

KEYWORDS

peripheral T-cell lymphoma, molecular diagnosis, oncogenesis, diagnosis, targeted therapy

1 Introduction

Peripheral T-cell lymphomas (PTCL) represent 10 to 15% of adult non-Hodgkin lymphomas. In the latest revised WHO and ICC classifications (1, 2), more than 30 entities are described, mostly defined by their clinical and pathological and phenotypic features, with a growing element of molecular data. Indeed, molecular studies based on high-throughput technologies have allowed for a better understanding of the oncogenic mechanisms involved and have improved the characterization of several entities.

Although only a few specific genomic alterations define a given entity, the use of molecular data, such as clonality assays and targeted next-generation sequencing (NGS), is now integrated into the routine diagnostic workflow of expert centers, in combination with clinical and pathological clues. However, the translation of high-throughput genomic studies to clinical practice is still limited due to the high cost of high-throughput technologies and little clinical relevance for most findings. In this review, we will detail the oncogenic mechanisms of the main non-cutaneous PTCL entities, the molecular targets that have an impact on their diagnosis or treatment, and the assays that are useful for the detection of these clinically relevant molecular alterations (3). Entities with a leukemic presentation (notably T-cell large granular lymphocytic leukemia and T-prolymphocytic leukemia) will not be detailed.

2 Biology of PTCLs

2.1 Oncogenic mechanisms

T-cell lymphomagenesis is a multistep process resulting from the accumulation of oncogenic events, such as genomic and epigenetic alterations and dysregulation of cellular signaling pathways, cell cycle, and immune surveillance (Figure 1). The microenvironment also plays a role in the initiation and maintenance of neoplastic transformation, best highlighted in angioimmunoblastic T-cell lymphoma (AITL), a disease characterized by a prominent tumor microenvironment (TME). However, the impact of the TME in other entities is still poorly understood.

Different types of genomic alterations can modify a biological function. Chromosomal translocations, detected by cytogenetic methods (karyotype, FISH, CGH), may produce fusion transcripts, detected by various technologies such as RT-PCR, RNAseq, or ld-RTPCR. They can result in aberrant expression, detectable by immunohistochemistry (for *ALK* fusions), or constitutive activation of oncogenes (such as *JAK2*, *VAV1*, *CD28*, etc.). Mutations in coding regions (single nucleotide variations or indels), detected by targeted exome or genomic sequencing, result in the gain of function of oncogenes or the loss of function of tumor suppressor genes. Mutations in noncoding regions have also been described, but their functional consequences are unclear. Disruption of the 3'UTR of PDL1 leads to its aberrant expression in extra-nodal NK/T-cell lymphomas and nasal-type (ENKTCL) and adult T-cell leukemia/lymphoma (ATLL), thus participating in immune escape (4, 5).

Epigenetic alterations appear to be a founding event in many PTCLs, mutations of genes involved in epigenetic regulation being frequently reported among different PTCL entities. Alterations of *TET2* and *DNMT3A*, reflecting clonal hematopoiesis (6), were initially described in tumoral and reactive cells of TFH lymphomas (7, 8), but have also been reported in other entities,

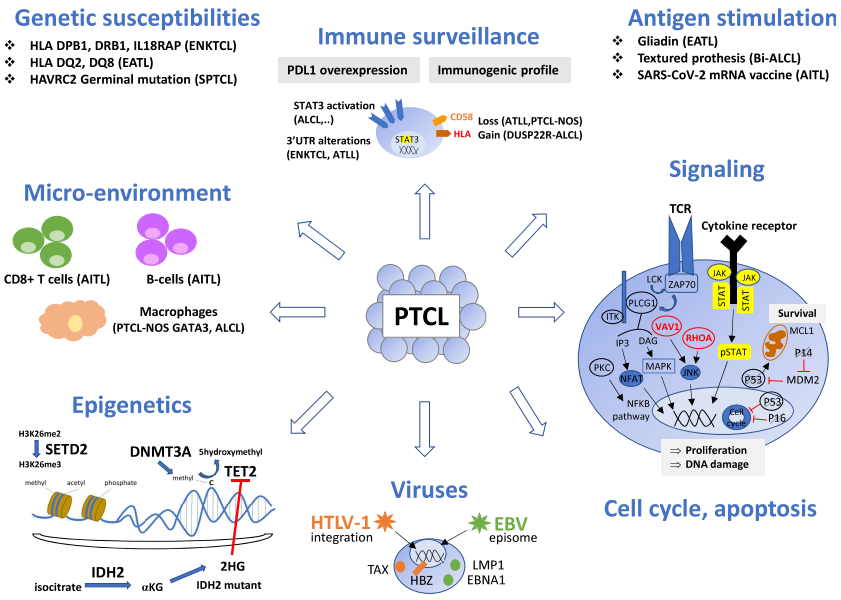


FIGURE 1
Oncogenic mechanisms of the main non-cutaneous PTCL entities. PTCL oncogenesis is a multistep process resulting from the accumulation of oncogenic events targeting epigenetics, signaling pathways (alterations of the TCR pathway is a common feature of TFH-PTCL, ATLL and certain PTCL-NOS, whereas alterations of the JAK/STAT pathway is shared by PTCL entities with a cytotoxic immunophenotype), cell cycle or apoptosis. Oncogenic viruses (HTLV1, EBV) are involved in a few specific entities. Chronic antigen stimulation may play a role as initiating event in several extranodal T or NK-cell lymphomas. Immune surveillance and crosstalk between neoplastic cells and reactive cells of the microenvironment is important, especially in AITL, where reactive cytotoxic CD8 T-cells and B-cells are associated with a poor and favorable outcome respectively. Genetic susceptibility is recognized in SPTCL, EATL and ENKTL. This figure depicts these events and their involvement for specific PTCL entities. Genes are crossed out when the alterations result in a loss of function. TFH, T follicular helper; ALCL, anaplastic large cell lymphoma; PTCL-NOS, peripheral T-cell lymphoma, not otherwise specified; ATLL, adult T-cell leukemia/lymphoma; ENKTCL, extra-nodal NK/T-cell lymphoma; HSTL, hepatosplenic T-cell lymphoma; EATL, enteropathy associated T-cell lymphoma; MEITL, monomorphic epitheliotropic intestinal T-cell lymphoma; SPTCL, subcutaneous panniculitis-like T cell lymphomas.

such as peripheral T-cell lymphoma not otherwise specified (PTCL-NOS), especially with a cytotoxic immunophenotype (9), or chronic lymphoproliferative disorders of NK cells (10). Although mutations of these two genes are not sufficient to induce lymphomas (11, 12), the loss of *TET2* is often required *in vitro* and *in vivo* prior to the occurrence of other genomic alterations (such as *RHOA* G17V mutation, less frequently VAV1 alterations or *FYN_TRAF3IP2* fusion) as a "second-hit" in the development of TFH-lymphoma (13–15). Recurrent mutations of *IDH2* R172, responsible for the production of the oncometabolite D-2 hydroxyglutarate, measurable in the serum of patients, are confined to tumoral T-cells in AITL (16). Mutations of *TET2*, *DNMT3A* and/or *IDH2* may explain the common loss of 5-hydroxymethylcytosine observed by immunohistochemistry in most PTCL entities, with the exception of hepatosplenic T-cell lymphoma (HSTL) (17), although it has been reported independently of the mutational status. Alterations of *SETD2* that inactivate histone methyltransferase function are almost ubiquitous in monomorphic epitheliotropic T-cell lymphoma (MEITL) and less frequent in HSTL (18, 19). Mutations of several other epigenetic modifiers (*KMT2C*, *KMT2D*, *CREBBP*, *EP300*) have also been reported among the main PTCL entities (20–22).

T-cell lymphomagenesis also implies the deregulation of signaling pathways, which occurs in many PTCL entities. Dysregulation of the TCR pathway is a common feature of TFH-lymphoma, ATLL, and PTCL-NOS (23, 24), whereas the JAK/STAT pathway is frequently altered in PTCL with a cytotoxic immunophenotype (ALK-positive or negative anaplastic large cell lymphoma (ALCL), breast implant associated-ALCL (Bi-ALCL), cytotoxic PTCL-NOS, extra-nodal NK/T-cell lymphoma, nasal-type (ENKTCL), enteropathy-associated T-cell lymphoma (EATL) and MEITL (9, 18, 22, 25, 26).

Dysregulation of the cell cycle in cancer is mostly due to inactivation of the tumor suppressor gene *TP53*, which is associated with a poor prognosis. In PTCL, alterations of *TP53* and *CDKN2A*/*PTEN* have been reported in GATA3-positive PTCL-NOS, associated with complex chromosomal rearrangements and genomic instability (27–29), as well as in ENKTCL (30) and EATL (31). On the contrary, these alterations appear to be infrequent in TFH-lymphoma and ATLL (24, 29). *TP63* rearrangements, described in a small subset of ALK-negative ALCL, appear to correlate with a poor prognosis (32, 33).

Another mechanism involved in T-cell lymphomagenesis is immune escape. Overexpression of PD-L1, due to alterations in the 3'-UTR region, lead to the anergy of reactive intra-tumoral lymphocytes in ENKTCL and ATLL (4, 5). PD-L1 expression has also been described in ALK-positive and ALK-negative ALCL, regulated by STAT3 activation, with a debated impact on the prognosis (34–36). The loss of CD58, HLA molecules, or β2-microglobulin, observed in ATLL and PTCL NOS, impairs recognition of the tumor cells by the immune system (24, 28). By contrast, DUSP22-rearranged ALK-negative ALCL shows immunogenic cues, with overexpression of the genes of T-cell co-stimulation *CD58* and *CD70* and HLA class II and decreasing PDL1 expression (37).

The role of reactive immune cells and stromal cells has been highlighted in AITL, a disease in which tumor cells are commonly scarce within a prominent microenvironment, thus influencing the results of gene expression studies (38). Microenvironmental molecular signatures may have prognostic relevance: a B-cell signature is associated with a favorable outcome, whereas macrophage and $CD8^+$ cytotoxic signatures correlate with an adverse prognosis (38–40). The presence of tumor-associated macrophages has also been reported to be associated with a poor prognosis in other PTCL entities, such as GATA3 PTCL-NOS (41), and ALK-positive anaplastic large cell lymphoma (ALCL) (42).

Viral infection (EBV and HTLV-1) is also recognized as a driver of PTCL oncogenesis.

a. HTLV-1 infection is required for the development of ATLL. This retrovirus is randomly integrated into the host DNA (43, 44), with a predilection for specific transcription factor binding sites, such as STAT1, HDAC6, and TP53 (45). While most HTLV-1 carriers are asymptomatic, with multiple clones, a dominant clone is detected in ATLL patients (46, 47). Viral replication is permitted by clonal expansion of infected $CD4^+$ T-cells (48). Expression of the oncogenic viral proteins TAX and HBZ leads to the disruption of homeostasis of infected cells, with the modification of epigenetic processes, genetic instability, and the accumulation of mutations (49). The TAX protein is highly immunogenic and responsible for the initiation of oncogenesis through NFKB and AP-1, while HBZ is involved in tumoral maintenance (50, 51).
b. EBV infection is a pre-requisite for the development of ENKTCL and other NK/T-cell neoplasms, such as aggressive NK-cell leukemia or the rare EBV^+ T/NK lymphoproliferative disorders of childhood. The mechanism for acquisition of the EBV receptor CD21 by NK and T-cells is still debated between trogocytosis and viral episome transfer (52, 53). The survival of infected cells is permitted by the type II latency pattern, with the expression of LMP1 and EBNA1 but not EBNA2. LMP1 promotes the proliferation of EBV-infected cells through deregulation of the p53, CMYC, and NF-κB pathways, in synergy with the production of cytokines (IL-2, IL-9, IL-10 et IL-15), by infected neoplastic cells and cells of the microenvironment (54).

Antigenic stimulation may also play a role in the initiation or progression of T/NK cell lymphomagenesis, as established for gliadin in EATL (55), textured breast-implants in Bi-ALCL (56), or recently suggested for the SARS-CoV-2 mRNA vaccine in AITL (57).

Finally, genetic susceptibility has been identified in several entities, notably association between the haplotypes HLA-DPB1, HLA-DRB1, and IL18RAP and ENKTCL (58, 59), HLA DQ2/DQ8 and EATL (60), and germline mutations of *HAVRC2* in subcutaneous panniculitis-like T-cell lymphoma (61).

2.2 Oncogenic events of the main non-cutaneous PTCL entities

PTCL can be derived from cells of the innate or adaptative immune system. Neoplasms likely deriving from the innate immune system comprise mostly extra-nodal lymphomas (ENKTCL, EATL, MEITL, HSTL, γδ-lymphomas, and probably cases among PTCL-NOS). They share a cytotoxic phenotype, alterations of the JAK/STAT pathway, and a context suggestive of chronic antigen stimulation. PTCL derived from cells of the adaptative immune system include most lymphomas with a nodal presentation with a T helper phenotype, such as TFH-lymphomas, ATLL, and PTCL-NOS. These lymphomas often show dysregulation of the TCR signaling pathway, in addition to alterations of epigenetic modifiers. The molecular characteristics of the main non cutaneous PTCL entities are summarized in Table 1.

2.2.1 Nodal TFH lymphomas

In the revised 2022 WHO and ICC classifications, the family of lymphomas derived from TFH cells is regarded as a single disease encompassing three morphological subtypes, commonly designated angio-immunoblastic T-cell lymphoma (AITL), follicular-type, and not otherwise specified. They have distinct morphological features but share a common TFH phenotype and signature, as well as a similar molecular pattern. In routine practice, the TFH phenotype is defined by the expression of CD4, with at least two TFH markers among PD1, ICOS, CD10, CXCL13, and BCL6, although none of them, in particular PD1 and ICOS, are fully specific, as they can be expressed by non-TFH reactive cells or other non-TFH PTCLs (62–65). TFH-lymphomas show a unique mutational landscape, characterized by the accumulation of alterations in genes involved in epigenetic regulation (*TET2*, *DNMT3A*, *IDH2*) (7, 11, 66) and the TCR pathway (*RHOA*, *VAV1*, *CD28*, *PLCG1*, *FYN*, *LCK*) (23, 67–73). Fusion transcripts involving genes of the TCR signaling (*ICOS_CD28*, *CTLA4_CD28*, *ITK_SYK* or involving *VAV1* with multiple partners) and NFKB (*FYN_TRAF3IP2*) pathways can be observed. Although mutations of *TET2* and *DNMT3A* may be observed in tumoral and reactive cells, hotspot mutations in *RHOA* G17V and *IDH2* R172 are thought to be restricted to the TFH tumor cells (74, 75). The recurrent *RHOA G17V* mutation, detected in 50 to 70% of AITL (23, 67–69, 75–78), impairs the GTPase domain, showing dominant negative activity and thus abolishing GTP binding and downstream signaling. This mutation is also responsible for VAV1 phosphorylation and TCR pathway activation (71). *RHOA G17V* drives TFH polarization and promotes lymphomagenesis *in vivo* through ICOS-PI3K-mTOR signaling (14, 15). The *IDH2* R172K mutation combined with *TET2* alterations modulate the tumoral microenvironment, promoting B-cell proliferation, the accumulation of plasma cells, and angiogenesis (79). Mutations in *CD28*, observed in 10% of TFH-PTCL, are reported to be mutually exclusive from fusion transcripts involving *CD28* and other genes of the TCR pathway (23, 72, 73, 76, 80, 81). Alterations in *VAV1* result in oncogenic activation of the NFAT pathway (70, 71, 82). Alterations of many other genes of the TCR pathway (*FYN*, *PLCG1*, *PIK3R1*, *PDPK1*, *AKT*, *LCK*, *TRAF6*) contribute to T-cell proliferation (23). The rare *ITK_SYK* fusion transcript has been described in follicular-type and in rare cases of AITL (83, 84). TFH lymphomas illustrate multistep oncogenesis, as shown by the development of « AITL » tumors *in vivo* in *TET2* knock-out mice transfected with a *RHOA* mutated gene (13, 14), or in double-mutant mice TET2/IDH2R172K (79).

Overall, although there is no pathognomonic genomic alteration that defines the TFH category, the detection of *RHOA G17V* and/or *IDH2 R172* mutations and, to a lesser extent, fusion transcripts involving *CD28* or *TRAF3IP2* constitute a supplemental clue to the diagnosis for pathologists.

2.2.2 - Anaplastic large-cell lymphomas

This category, defined by large “hallmark” cells showing strong and homogenous CD30 expression by immunohistochemistry, includes several entities based on the association of ALK-rearrangement and the clinical presentation as systemic, cutaneous, or breast implant-associated disease. Cutaneous ALCL are not considered here.

A) ALK-positive ALCL is the only entity defined by recurrent genomic translocations involving the *ALK* gene on chromosome 2p23 with various partners, the most frequent (~80%) being *NPM1*. The translocation produces an oncogenic fusion protein consisting of the association of the N-region of a partner gene with the catalytic tyrosine kinase domain of ALK, resulting in constitutive activation by dimerization. The chimeric NPM1_ALK protein triggers several oncogenic pathways (JAK/STAT, PI3K, MAPK, PLCG), leading to neoplastic transformation (85, 86), whereas *TRAF1_ALK* activates the NFKB pathway (87, 88). Recently, mutations of *NOTCH1* and genes of the TCR pathway have also been reported (89). The diagnosis is based on the detection of aberrant ALK expression by immunohistochemistry using anti-ALK antibodies. The pattern of staining may be nuclear +/- nucleolar and/or cytoplasmic, depending on the partner gene involved in the translocation (Table 2). The disease, which mainly occurs in children and young adults, follows a generally favorable prognosis (5-year OS around 90%) after chemotherapy with CHOEP (100–102) or BV-CHP (103). The prognosis may be less favorable in cases with secondary MYC overexpression or rearrangement, in certain histologic variants (small-cell or lymphohistiocytic) occurring in children (88, 104, 105).

B) Systemic ALK-negative ALCL is still heterogeneous in the current classifications, gathering cases with different oncogenic pathways:

- Rearrangement of the 6p25.3 locus involving *DUSP22* and *IRF4* (106) defines a peculiar subgroup (approximately 25-30% of ALK-negative ALCL), characterized by a non-cytotoxic phenotype, silencing of the tumor suppressor gene *DUSP22* while showing normal *IRF4* expression, absence of STAT3 activation, global DNA hypomethylation, an immunogenic molecular profile (overexpression of CD58, CTA, HLA class II), and expression of LEF1 (37, 107–110). Recurrent *MSC E116K* mutations are responsible for activation of the CD30-IRF4-CMYC axis and the

TABLE 1 Molecular characterization of the main non-cutaneous PTCL entities.

Entity	Differentiation	Molecular features
TFH-lymphomas	TFH	- DNA methylation: *TET2*, *DNMT3A*, *IDH2* R172 mutations - TCR pathway: *RHOA* G17V, *CD28*, *VAV1*, *PLCG1* mutations - Fusion transcripts: *ICOS_CD28*, *CTLA4_CD28*, *ITK_SYK*, *ITK_FER*, fusion transcripts involving *VAV1*
ALK-positive ALCL	Activated cytotoxic T cell	- Fusion transcripts involving *ALK* - Mutations in genes of the NOTCH1 pathway
ALK-negative ALCL	Activated cytotoxic T cell	STAT3 activation: *JAK1* and/or *STAT3* mutations Fusion transcripts involving *ROS*, *TYK2*, *FRK, CAPRIN2* Absence of STAT3 activation: *DUSP22*/*IRF4* (locus 6p25.3) rearrangement *MSC*E116K mutation Others: *TP63* rearrangements
Breast-implant ALCL	Activated cytotoxic T cell	- JAK/STAT pathway: *STAT3*, *JAK1*, *SOCS3*, *STAT5B*, *SOCS1*, *PTPN1* mutations - Epigenetics: *KMT2D*, *KMT2C*, *CREBBP*, *CHD2, TET2, DNMT3A* mutations
ATLL	Memory regulator T cell	- TCR pathway: *PLCG1, PRKCB, CARD11, VAV1, IRF4, FYN, CCR4, CCR7, RHOA, CD28* mutations - Immunosurveillance: *CD58, B2M, HLA* (class I) mutations - JAK/STAT pathway: *JAK3, STAT3, PTPN1* mutations - Transcription factor: *GATA3, IKZF2, PRDM1* mutations - Epigenetics: *TET2, DNMT3A, IDH2, SETD2, EP300, KDM6A* mutations - Fusion transcripts: *ICOS_CD28* and/or *CTLA4_CD28*
ENKTCL (nasal type)	NK>>T (γδ or αβ)	- *BCOR*, *DDX3X*, *TP53*, *MGA*, *STAT3*, *STAT5B*, *MLL2*, *ARID1A*, *MSN* mutations - 3 molecular subgroups : °TSIM : mutations of genes of the JAK/STAT pathway, *TP53*, amp9p24.1(*JAK2*, *PDL1/2*), amp17q21.2 (*STAT3/5A/5B*), EBV latency type II => NK cells °HEA: mutations of *HDAC9*, *EP300*, *ARID1A*, EBV latency type II => T-cells °MB: mutations of *MGA*, del1p22.1 (*BRDT*), *MYC* overexpression, EBV latency type I => T-cells
HSTL	Tgδ> Tαβ	- *SETD2, STAT5B, INO80, ARID1B, STAT3, PIK3CD* mutations
Indolent clonal T-cell lymphoproliferative disorder of the gastro-intestinal tract	CD8+ or CD4-/CD8- (TH2)	- Structural alterations of the 3'UTR regions of IL2 coding gene
	CD4+ or CD4+/CD8+	- JAK/STAT pathway: *STAT3*, *SOCS1* mutations, *STAT3_JAK2* fusion - Epigenetics: *TET2*, *DNMT3A*, *KMT2D* mutations
EATL	Intraepithelial lymphocyte (Tαβ)	- JAK/STAT pathway: *JAK1* (p.G1097 dans 50%), *JAK3*, *STAT3*, *STAT5B*, *SOCS1* mutations - *KRAS*, *NRAS* mutations - NFκB pathway: *TNFAIP3*, *TNIP3* mutations - Epigenetics: *TET2*, *KMT2D*, *DDX3X*, *SETD2* (15%) mutations
MEITL	Intraepithelial lymphocyte (Tγδ > Tαβ)	- Alterations of *SETD2* (mutations, deletions) - *STAT5B*, *JAK3*, *TP53*, *GNAI2* mutations
T-LGLL	Tαβ (CD8) >> Tγδ	- JAK/STAT pathway: *STAT3*, less frequently *STAT5B* mutations - Epigenetics: *TET2*, *DNMT3A*
PTCL-NOS	TH1 (αβ >> δγ), common cytotoxic phenotype	- Epigenetic: *TET2*, *DNMT3A*, *KMT2D*, *SETD2* mutations - TCR pathway: *VAV1*, *PLCG1*, *PRKCB*, *CARD11* mutations; fusion transcripts involving *VAV1* - JAK/STAT pathway: *STAT3*, *STAT5B*, *JAK3*, *SOCS1* mutations
	TH2 (Tαβ)	- Deletions of *CDKN2A*, *TP53*, *PDGFA*, *STK11*, *WDR24*, *CDK4*, *CCND1*, *AKT*, *RPTOR*… - Gains/amplifications of *STAT3*, *CMYC*

TFH, T follicular helper; ALCL, anaplastic large cell lymphoma; PTCL-NOS, peripheral T-cell lymphoma, not otherwise specified; ATLL, adult T-cell leukemia/lymphoma; ENKTCL: extra-nodal NK/T-cell lymphoma, HSTL, hepatosplenic T-cell lymphoma; EATL, enteropathy associated T-cell lymphoma; MEITL, monomorphic epitheliotropic intestinal T-cell lymphoma; T-LGLL, T-cell large granular lymphocytic leukemia.

dysregulation of cell cycle arrest (111). These rearrangements were initially detected by mate-pair DNA sequencing in the context of a translocation t(6,7)(p25.3;q32.3) also involving the non-coding gene *FLJ43663* at the fragile site FRA7H of chromosome 7 (106). The prognosis is debated, favorable in most studies (112, 113) but not confirmed in others (114, 115).

- Rearrangements of *TP63*, due to the inversion inv (3) (q26q28) or translocation t(3,6)(q28;p22.3) that produce the fusion transcripts

TABLE 2 Fusion transcripts involving *ALK* in ALK-positive anaplastic large cell lymphoma.

Fusion	Translocation	Immunostaining
NPM1_ALK (90)	t (2,5)(p23.2;q35.1)	Nuclear and cytoplasmic
TPM3_ALK (91)	t(1;2)(q25;p23)	Cytoplasmic with peripheric reinforcement
ATIC_ALK (92)	inv(2)(p23q35)	Cytoplasmic diffuse
TFG_ALK (93)	t(2;3)(p23;q12.2)	Cytoplasmic diffuse
CLTCL_ALK (94)	t(2;17)(p23;q23)	Cytoplasmic granular
MSN_ALK (95)	t(X;2)(q11-12;p23)	Membranous
ALO17_ALK (96)	t(2;17)(p23;q25)	Cytoplasmic diffuse
MYH9_ALK (97)	t(2;22)(p23;q11.2)	Cytoplasmic diffuse
TRAF1_ALK (87)	t(2;9)(p23;q33)	Cytoplasmic
EEF1G_ALK (98)	t(2;11)(p23;q12.3)	Cytoplasmic
PABPC1_ALK (99)	t(2;8)(p23;q22)	Cytoplasmic

TBL1XR1_TP63 and *TP63_ATXN1* respectively, coding for oncogenic chimeric proteins, are rare and associated with a poor prognosis (32, 112, 114). The detection of P63 by immunohistochemistry may reflect P63 overexpression independently of the presence of fusion transcript (33).

- Aberrant truncated transcripts of *ERBB4* was also reported in 24% of ALK-negative ALCL in one study, associated with a Hodgkin-like morphology, without clinical relevance (116).

- Expression of pSTAT3 by immunohistochemistry, reflecting activation of the JAK/STAT pathway, is common in ALK-positive and ALK-negative ALCL, with the notable exception of those cases associated with *DUSP22* rearrangement. Among ALK-negative ALCLs, a recent study that excluded cases with rearranged *DUSP22* suggested that positive pSTAT3 cases constitute a distinct subgroup, characterized by a cytotoxic phenotype and the expression of EMA and PDL1, that is associated with a better prognosis than negative pSTAT3 cases (117). Such constitutive phosphorylation of STAT3 has been previously shown to be related to mutations in *JAK1* and/or *STAT3*, reported in 18% of ALK-negative ALCLs, as well as in fusion transcripts involving *ROS, TYK2*, and *FRK* (26, 118). More recently, fusion transcripts involving *JAK2* with several partners (*PABPC1*, *PCM1*, *ILF3*, *TFG*, *MAP7*) were detected by targeted RNAseq and associated with a Hodgkin-like morphology (119).

C) Breast-implant associated ALCL is a site-specific entity that occurs after a long latency after a breast implant for reconstruction or cosmetic reasons. Most cases are non-invasive. The disease appears to be due to chronic inflammation, with possible TH2 polarization, linked to a macro-textured implant (120). High-throughput sequencing studies have highlighted alterations of genes involved in the JAK-STAT pathway (*STAT3*, *STAT5B*, *JAK1*, *JAK3*, *SOCS1*, *SOCS3*), leading to its constitutive activation, together with recurrent mutations in epigenetic modifiers (*KMT2C*, *CREBBP*) (22, 121), the loss of chromosome 20 (122), and chromosome 9p24 gains, leading to PDL1 expression (123). Recently, a *STAT3_JAK2* fusion transcript was also reported (124).

Several immunohistochemical algorithms have been recently proposed to classify ALCL based onLEF1, P63, and pSTAT3 (117, 125), although this currently has no impact on the management of ALK-negative ALCL patients.

2.2.3 EBV-positive NK or T-cell neoplasms

EBV-related NK or T-cell neoplasms are heterogenous diseases derived from T or NK cells (126). The revised WHO and ICC classifications recognize ENKTCL, and primary nodal EBV-positive T/NK-cell lymphomas, characterized by nodal involvement, as distinct entities (1, 2). In addition to EBV, considered to be a driver of oncogenesis in these lymphomas, defined by EBV infection of virtually all neoplastic cells, as shown by *in situ* hybridization with EBER probes, the mutational landscape of ENKTCL is characterized by recurrent mutations of genes coding for RNA helicases (especially *DDX3X*), as well as *TP53*, genes of the JAK/STAT pathway (*JAK3*, *STAT5B*, *STAT3*) and epigenetic modifiers (*MLL2*, *ARID1A*, *EP300*, *ASXL3*) (20, 30, 127). The initial poor prognosis associated with *DDX3X* and *TP53* mutations for patients treated with the CHOP regimen was not confirmed for patients receiving L-asparaginase treatment (20, 30, 128). Recurrent deletions of the 6q21 locus encompassing tumor suppressor genes (*PRDM1*, *ATG5*, *AIM1*, *FOXO3* et *HACE1*) have been detected by CGH array (129–131). A recent large integrative analysis of genome, exome, and RNA sequencing, identified three molecular subgroups (30):

- the "TSIM (tumor suppressor and immunomodulator)" subgroup is characterized by frequent *TP53* mutations, deletion of the 6q21 locus, amplification of the 9p24.1 locus containing *PDL1* and *PDL2*, and the amplification of genes of the JAK/STAT pathway. This subgroup presents a gene expression signature enriched in NK-cell genes. There is an EBV latency II phenotype, with expression of the lytic gene *BALF3*, responsible for DNA damage and genomic instability.

- the "MB (MGA, BRDT)" subgroup is characterized by frequent *MGA* mutations, loss of heterozygosity of *BRDT*, and MYC overexpression, as well as activation of the MAPK, NOTCH, and WNT pathways. The EBV latency is of type I, with downregulation of *LMP1*.
- the "HEA (HDAC, EP300, ARID1A)" subgroup is characterized by mutations of epigenetic modifier genes (*HDAC9*, *EP300* et *ARID1A*), resulting in aberrant histone acetylation. The gene expression profile is enriched in T-cell genes and shows activation of the TCR and NFKB pathways. The EBV latency is of type II, with expression of the *BNRF1* lytic gene.

Although there is currently no applicability of this molecular subclassification in routine practice, the poor prognosis of the MB subgroup relative to TSIM and HEA (3-year OS rate of 38% versus 80% and 90%, respectively) may justify the evaluation of MYC expression in ENKTCL. Structural alterations of *CD274*, coding for PDL1, appear to confer sensitivity to immune checkpoint inhibitors (5, 132).

EBV-positive nodal T- and NK-cell lymphoma or primary nodal Epstein-Barr virus–positive T-cell/NK-cell lymphoma, is now recognized as a distinct entity in both the WHO and ICC classifications respectively, due to its differences with ENKTCL. This entity is morphologically characterized by the lack of necrosis and angiocentrism, a common $CD8^+$ $CD56^-$ phenotype, a frequent T-cell origin, and, finally, peculiar molecular abnormalities, with frequent *TET2*, *PIK3CD*, and *STAT3* mutations, activation of the NFKB, IFNγ, and JAK-STAT3 pathways, resulting in high PDL1 expression, and lower genomic instability (133). The prognosis is reported to be poorer than for ENKTCL.

The mutational landscape of ENKTCL is shared with that of other EBV-positive NK/T-cell neoplasms, in particular, aggressive NK-cell leukemia (134–136), as well as that of chronic active EBV-disease (137). This genetic landscape may be of clinical relevance in the rare cases that require a differential diagnosis from infectious mononucleosis.

2.2.4 Adult T-cell Leukemia/Lymphoma

This HTLV-1-associated T-cell neoplasm occurs after a long latency (more than 25-30 years) following infection, mainly due to prolonged breast feeding and, less frequently, sexual transmission (138). The histopathological diagnosis is challenging in the absence of information concerning the HTLV-1 status, as the pathological aspects of ATLL are highly heterogeneous. It can be evoked by the loss of CD7, together with the expression of CD25 and FOXP3, although the $CD25^+$/$FOXP3^+$ immunophenotype is variable and not specific to ATLL (139–142). The molecular landscape is characterized by mutations in genes of the TCR pathway (*PLCG1*, *PRKCB*, *CARD11*, *VAV1*, *IRF4*, *FYN*, *CCR4*, *CCR7*, *RHOA*, *CD28*), JAK/STAT pathway (*JAK3*, *STAT3*, *PTPN1*), immune surveillance (*CD58*, *B2M*, *HLA* class I), DNA damage (*TP53*, *CDKN2A*, *POT1*), epigenetic modifiers (*TET2*, *DNMT3A*, *IDH2*, *SETD2*, *EP300*, *KDM6A*), transcription factors (*GATA3*, *IKZF2*, *PRDM1*), and fusion transcripts involving CD28 (*ICOS_CD28* and *CTLA4_CD28*) (24, 143). The co-expression of these two fusion transcripts can occur in patients younger than 50 years of age (144). Gene mutations of the TCR/NFKB pathway, *TP53*, and *IRF4* are associated with an aggressive outcome, whereas *STAT3* mutations are frequently observed in patients with more indolent disease (143, 145). The type of *CCR4* mutation also has a specific prognostic impact (unfavorable in cases of frameshifts vs non-synonymous variations) (146).

2.2.5 Intestinal T-cell lymphomas

Enteropathy-associated T-cell lymphoma (EATL) and monomorphic epitheliotropic intestinal T-cell lymphoma (MEITL) are two distinct entities, with different morphological and immunophenotypic features. Although both are derived from intestinal intra-epithelial lymphocytes (IEL) expressing CD103, EATL and MEITL show distinct clinico-pathological and molecular characteristics (31).

a. EATL is associated with celiac disease or gluten sensitivity. Its histopathological features include the proliferation of pleomorphic to anaplastic T-cells expressing CD3 and CD30, but lacking CD4 and CD8, despite an activated cytotoxic profile. CD103 is variably expressed. Overexpression of P53 is detectable by immunohistochemistry, independently of gene alterations (147). This entity shows frequent alterations of the JAK/STAT pathway (in particular, *STAT3* and *JAK1*, as well as *SOCS1* and *SOCS3*), whereas *STAT5B* mutations are almost constantlyabsent (18, 148, 149). *TET2* and, less frequently, mutations of the RAS/MAPK pathway (149) can be observed, whereas *SETD2* mutations were almost absent in most recent series (18, 148). Gene expression profiling studies have shown enrichment for genes of the JAK/STAT (*STAT3*, *STAT5A*) and IFNγ pathways (31).
b. MEITL does not associate with celiac disease and is typically characterized by the proliferation of monomorphic medium cells, showing epitheliotropism and a $CD8^+$ $CD56^+$ phenotype. However, approximately 25% of cases may show more pleomorphism and certain phenotypic variations associated with the prognosis, in particular, a better outcome in the presence of aberrant expression of CD20 or poor outcome in the presence of *MYC* expression and *TP53* alterations, suggesting the utility of screening for these abnormalities in routine practice (150, 151). MEITL has a very homogeneous genetic landscape, with almost consistent alterations of *SETD2* (mutation +/- deletion) associated with mutations of *STAT5B* (approximately 60%) or *JAK3* and *GNAI2*, which constitute a common feature and may help pathologists in difficult cases (18, 150–152).

Indolent clonal T-cell lymphoproliferative disorder of the gastrointestinal tract (2), also designated indolent T-cell lymphoma of the gastrointestinal tract in the WHO classification (1), is now recognized as a definitive entity in both classifications due to the recent evidence of neoplastic molecular features, i.e.,

alterations of genes in the JAK/STAT pathway or epigenetic modifier genes and *JAK2_STAT3* fusions or structural alterations of the 3'UTR of the IL2 gene, depending on the CD4 or CD8 phenotype (153, 154). Despite an indolent course, some cases may relapse, spread to other sites, or transform, indicating potential aggressiveness (155, 156).

Indolent NK-cell lymphoproliferative disorder of the gastrointestinal tract is a rare condition, and a new entity in the WHO and ICC classification. Although neoplastic molecular characteristics have also been described, in particular, recurrent deletions of *STAT3*, there is no extension of this lymphoproliferation beyond the gastrointestinal tract and the outcome is favorable (157).

2.2.6 Hepatosplenic T-cell lymphoma

This rare neoplasm occurs preferentially in young males but can arise at any age, with a possible context of immunosuppression. The diagnosis is based on highly characteristic pathological features, in particular sinus infiltration in the bone marrow by small to medium lymphocytes with a $CD3^+$, $CD5^+$, $CD4^-/CD8^-$, $CD56^+$ phenotype, commonly $TCR\gamma\delta^+$. The sinusal infiltration in the liver and spleen is less specific. There is typically no lymph node involvement. This entity was initially characterized by an isochromosome 7q and chromosome 8 trisomy (158, 159), but cytogenetic material is not always available in routine practice to support the diagnosis and FISH analysis can be challenging. The mutational landscape has been reported, identifying three types of mutations involving 1/ epigenetic modifier genes (*SETD2*, *ARID1B*, *INO80*, *TET3* and *SMARCA2*), 2/*STAT5B* or *STAT3* that are mutually exclusive, and 3/*PIK3CD* (19, 160, 161). Gene expression profiling studies show a distinct signature, characterized by the overexpression of oncogenes (*FOS*, *FOSB VAV3, MAF)*, NK-cell associated genes (*KIR3DS1*, CD244 and other KIRs), the tyrosine kinase *SYK*, and *S1PR5*, and downregulation of *AIM1*, which could constitute targets for therapy in this disease that has always fatal outcome (162). A recent single-cell profiling study suggested a change in the gene expression profile of the tumor cells during disease progression under the selective pressure of therapy (163).

2.2.7 PTCL-NOS

PTCL-NOS is a diagnosis of exclusion, corresponding to cases that do not fulfill the criteria for defined PTCL entities. Thus, a large panel of immunohistochemical markers and the integration of clinical and often molecular features are required to exclude any other PTCL. Gene expression profiling studies have shown two subgroups based on expression of the TBX21 and GATA3 transcription factors associated with the immunological TH1 and TH2 signatures, respectively (39, 40), confirmed by immunohistochemistry (164). The TBX21 group is enriched in genes of IFNγ and NFKB pathway signatures and shows mutations of genes involved in epigenetic regulation (*TET1*, *TET3*, *DNMT3A*), whereas the GATA3 group shows a cell proliferation signature driven by MYC, together with enrichment in PI3K/Akt/mTOR pathway signatures, a higher number of genomic copy number abnormalities, and a poorer outcome (27, 165). In routine practice, there is no consensus concerning the proposed thresholds of immunohistochemical markers to define these two subgroups and an understanding of the clinical relevance of such immunohistochemical algorithms requires further studies.

The mutational landscape of PTCL-NOS is currently poorly defined, likely due to the heterogeneity of this category. Only a few "omic" studies focusing on PTCL-NOS have been published to date. Targeted sequencing has shown mutations of epigenetic modulator genes, notably histone methylation (*KMT2D*, *SETD2*, *KMT2A*, *KDM6A*) or acetylation (*EP300*, *CREBBP*), as well as that of genes of the TCR pathway (*TNFAIP3*, *TRAF3*, *TNFRSF14*) and tumor suppressor genes (*TP53*, *ATM*, *FOXO1*, *BCORL1*) (21, 166). Recent integrative studies based on exome and RNA sequencing have confirmed mutations of genes involved in epigenetic regulation (*TET2*, *DNMT3A*, *KMT2C*, *KMT2D*, *SETD2*, *CREBBP, ARID1A*), tumor suppressor genes (*TP53*, *TP63*, *ATM*, *FAT1*, *LATS1*, *STK3*), and genes of the NOTCH pathway (NOTCH1 and 2) (28, 167). In one study, mutations in *FAT1* were shown to be associated with a poor prognosis (167). RNAseq studies have shown fusion transcripts involving *VAV1* with various partner genes (*GSS*, *THAP4*, *MYO1F*, *S100*, *HNRNPM*) and rearrangements of *VAV1* were detected by FISH in 11% of PTCL-NOS (28, 70, 82). The *VAV1_MYO1F* transcript induces tumoral TH2 polarization and the accumulation of tumor-associated macrophages (41). Other fusion transcripts have also been reported in single cases (*ITK_FER*, *IKZF2_ERBB4*, *ETV6_FGFR3*) (82). A t (14, 19)(q11;q13) translocation, involving *TCRA* and the poliovirus receptor-related 2 gene (*PVRL2*), resulting in *BCL3* overexpression, has also been reported in PTCL-NOS, including one case with the morphological variant of Lennert's lymphoma (168, 169).

PTCL-NOS with a cytotoxic phenotype has been reported in 25 to 40% of cases, associated with impaired immunity and a poor prognosis (9, 170). This immunophenotypic subgroup has also been identified in gene expression studies within the PTCL-NOS TBX21 subgroup, enriched for genes of CD8/NK cells, the IFN response, and an immunosuppressive signature (39, 40). Targeted sequencing has shown recurrent mutations of genes involved in epigenetic regulation (*TET2*, *DNMT3A*), TCR (*VAV1*, *PLCG1*, *PRKCB*, *CARD11*) and the JAK/STAT pathways, as well as *TP53* (9). Fusion transcripts involving *VAV1* have been detected in 14% of patients. In another study, two cases of cytotoxic PTCL-NOS with diffuse cutaneous and medullary involvement showed a t (6, 14) (p25;q11.2) translocation resulting from rearrangement between the TCRα and IRF4 loci (171).

Despite these advances in our knowledge of the molecular biology of this entity, there is still an unmet need for the management of PTCL-NOS patients.

3 From biology to the diagnosis and management of PTCL patients

The diagnosis and classification of PTCLs are often challenging for pathologists, requiring experienced hematopathologists and access to molecular tests. In the absence of clear diagnostic guidelines, practices are often heterogenous between centers (172–174).

Analysis of rearrangements of the TCR loci (especially *TRG* or *TRB*) is an important element of the diagnostic process. PCR-based assays (BIOMED-2) are largely widespread in routine practice due to their reliability on FFPE samples (160–162). However, there are a number of pitfalls in the interpretation of clonality testing due to "false-negative" results in cases with low tumoral content, especially common in AITL, or due to T-cell oligoclones, as observed in AITL (175). Conversely, the presence of clonal TCR rearrangements in certain reactive conditions or even in B-cell lymphomas (notably Hodgkin lymphomas) due to TCR repertoire restriction can be misleading (176). The development of NGS-amplicon based clonality assays may improve the detection of scarce clones in a polyclonal background and allow the determination of clonotypes (177). Several authors have proposed analyzing TCR genes by whole genome sequencing, but its applicability in routine practice is still limited (178). Others have highlighted the potential interest of analyzing non-recombined T-cell receptor sequences using a digital PCR assay (179).

Recently, gene expression studies suggested molecular classifiers to discriminate the main PTCL entities, with certain limitations due to tumor cell content and the quality of the nucleic acid (40, 180, 181). Such tools should be used in routine practice with caution, as they were developed for the classification of the most common entities, their robustness has not yet been extensively evaluated, and the results need to be interpreted in the context of the histopathological analysis. Indeed, misclassification using these algorithms or discordance with the histopathological data occur for 15 to 20% of samples, likely due to a prominent microenvironment or plasticity of the tumor cells (180, 181). Sequencing of transposase-accessible chromatin (ATAC-seq) has been proposed as another innovative strategy to classify PTCL (182), but it requires fresh or frozen samples and its applicability in routine practice has not been yet evaluated.

Exome and genome sequencing studies have allowed a precise description of the mutational landscape of almost all PTCL entities. An increasing number of laboratories have developed targeted NGS panels for the molecular characterization of lymphomas or hematological neoplasms that are useful for their diagnosis and classification (183). The diagnostic performance of targeted NGS relative to that of measuring T-cell clonality by BIOMED multiplex PCR in PTCL was assessed in one study and showed similar sensitivity (approximately 95%) but significantly superior specificity (100% versus 45%) (184). However, there is currently no consensus concerning the design of the panel or the sequencing depth or coverage, which may affect the interpretation of the results. Hotspot mutations of diagnostic relevance, notably *RHOA G17V* or *IDH2 R172* mutations, can also be detected using alternative technologies, such as allele-specific PCR, digital PCR, and RTMLPA (180, 185–188).

As described above, despite highly characteristic genetic profiles for certain PTCLs, such as TFH-lymphomas and MEITL, there is no single pathognomonic molecular alteration that can define an entity, apart from ALK-positive ALCL. However, the detection of *RHOA* G17V and *IDH2* R172 mutations in routine practice strongly supports the diagnosis of TFH-PTCL (14, 16, 63, 64, 72). Although *RHOA* mutations can also be observed in 10% of ATLL, only 1% correspond to G17V (189), whereas *IDH2* R172 is almost specific to AITL.

Within ALCL, the discovery of the translocation t(2,5) led to the development and use of an anti-ALK antibody in routine practice, allowing rapid and efficient determination of the ALK status by immunohistochemistry (90, 190). The identity of the *ALK* gene partner does not appear to be important, with no prognostic relevance, with the exception of the rare *TRAF1_ALK* fusion transcript, which was shown to be associated with a poor outcome in a recent study (87). In children with ALK-positive ALCL, the prognosis also correlates with the ALK antibody titer and the copy number of the ALK fusion transcript in the blood at diagnosis (MDD: minimal disseminated disease) and after treatment (MRD: minimal residual disease) (189, 191–194). The significance of these parameters is unknown in adult patients.

In routine practice, FISH is required to diagnose DUSP22-rearranged ALK-negative ALCL, a molecularly distinct subgroup that probably merits being individualized (37). Interestingly, it is also characterized by the presence of hotspot mutations of *MSC* E116K in 35% of DUSP22-rearranged cases, a finding currently without clinical relevance (111). In the context of intestinal T-cell lymphomas, the identification of *SETD2* alterations strongly favors the diagnosis of MEITL and may be helpful in distinguishing difficult cases from EATL (18). These alterations (mutations and/or deletions) result in reduced H3K36 trimethylation, which can be detected by immunohistochemistry (195).

A number of genetic alterations may also predict the outcome of patients with a T- or NK-cell neoplasm, as observed in ENKTCL, with the poor prognosis of the MB subgroup (30), and in ATLL with CCR4 mutations or CCR7 alterations (146, 196–198). In AITL, the $DNMT3A^{R882X}$ mutation may be associated with a poor prognosis and resistance to anthracyclines (199), a finding that could influence the management of these patients in the future. MYC expression/rearrangement or TP53 alterations are associated with a poor prognosis in various PTCL entities, especially ALK-positive ALCL (104, 105), ENKTCL (30) and MEITL (151), but without a significant impact on the management of these patients.

The diagnosis of ATLL is challenging for pathologists without knowledge of the HTLV-1 serology status. Morphological and immunophenotypic features may be confusing for ALK-negative ALCL, GATA3 PTCL-NOS, or even TFH-lymphomas, with an impact on the appropriate management of these patients. There is an unmet need for the development of HTLV-1 biomarkers applicable to FFPE samples in routine practice. TAX is not expressed in most ATLL tumors, whereas HBZ is the only viral transcript expressed during disease progression and could be a good candidate (50, 51). *In situ* hybridization was proposed to detect the *HBZ* gene in FFPE tissues in a single study, but there has thus far been no development of this technology in routine practice (200). More recently, targeted gene expression studies have been developed to measure expression of the *HBZ* transcript in routinely-fixed samples (181, 201).

Thus far, the detection of fusion transcripts has not been integrated into the routine diagnosis of PTCL due to the low prevalence of known fusions (10%) and limited accessibility to available technologies. Although RNAseq is the most exhaustive technology to detect fusion transcripts, several targeted RNA sequencing alternatives have been developed (ArcherFusionPlex®, Qiaseq RNA fusion XP®, and ld-RTPCR (202)), which can be

implemented in a routine laboratory at a lower cost. Despite the current lack of clinical relevance of most fusion transcripts, the recent identification of rearrangements involving *JAK2* in systemic CD30-positive PTCL (119, 203), Bi-ALCL (124), in indolent clonal T-cell lymphoproliferative disorder of the gastrointestinal tract (153), and cutaneous T-cell lymphoma (204–208) opens the door to targeted therapies requiring the detection of such fusion transcripts. Furthermore, in addition to pathological features, the detection of certain transcripts may be of diagnostic value to support a diagnosis among several hypotheses. For example, *ICOS_CD28*, *ITK_SYK*, or *FYN_TRAF3IP2* fusions favor a diagnosis of PTCL, especially TFH-lymphoma in difficult cases, raising the possibility of the differential diagnosis from Hodgkin lymphoma or marginal zone lymphoma.

Recent studies on a limited number of cases have demonstrated the applicability of assessing circulating tumor DNA (ctDNA) by high-throughput sequencing for PTCL. In a comparison with matched tumors, ctDNA detected by HTS-sequencing of the TCR was detected for 78% of various PTCL entities (209). The detection of hotspot mutations in AITL (*RHOA* and *IDH2*) appears to be promising and sensitive, with 100% concordance between cell-free DNA and the tumors by NGS in one study (210) and a prevalence of 70% in another using allele-specific PCR (188). In ENKTCL, a concordance of 93.5% between ctDNA and tumor biopsy sequencing was observed, with a potential prognostic significance (211–213). Beyond the potential application for the detection of minimal residual disease during follow-up or at relapse, the detection of ctDNA may also be a promising tool to help for the diagnosis of difficult cases, especially those with limited tumor material, in combination with pathological analysis.

4 From molecular targets to personalized treatment: alternatives or additive therapeutic options to standard chemotherapy

The CHO(E)P-based regimen has been the standard of care for PTCL for many decades (214). To date, most alternative therapies have failed to demonstrate a better outcome and the prognosis of patients for most PTCLs is still poor (215, 216), even for stage I-II disease (217).

4.1 Frontline targeted therapies

A recent major change in frontline therapy is the use of brentuximab-vedotin (BV), in addition to CHP chemotherapy, for patients with CD30 positive PTCL. Approval for the use of BV by the US Food and Drug Administration (FDA) followed the ECHELON-2 study, which demonstrated a significant improvement in progression free survival (PFS) (median 48 months in the BV-CHP group versus 20.8 months in the CHOP group, p=0.0110), and a reduced risk of death in the BV-CHP arm, although the median overall survival (OS) was not reached (103). However, the subgroup analyses confirmed the benefit for ALCL patients receiving BV, but not for those with AITL. For PTCL-NOS, the potential benefit is unclear, probably due to the heterogeneity of the disease with respect to the percentage of CD30-positive cells (threshold ≥10% of cells by local review). The addition of BV to standard chemotherapy has also been shown to provide an improvement in event-free survival of children with ALK-positive ALCL (218). In addition, a retrospective pooled study showed a significant improvement of OS and PFS in ALK-positive ALCL with the use of CHOEP in frontline therapy compared to CHOP, independently of age (100). To date, there has been no comparison between CHOEP and BV-CHP in the frontline management of ALK-positive ALCL patients.

A second large trial compared the addition of romidepsin to CHOP versus CHOP alone in previously untreated PTCL patients (219). Although the results of the study were negative, as PFS did not statistically increase in the romidepsin CHOP group relative to the control arm, a trend towards longer PFS was observed for TFH-lymphoma patients, suggesting susceptibility of TFH-lymphomas to drugs targeting epigenetics. A phase 2 trial combining the oral form of 5-azacytidine to CHOP in the first line for 21 PTCL patients, including 17 with TFH-lymphoma, showed promising results, with an 88% complete response (CR) rate for TFH-lymphoma patients and 69% two-year PFS. However, these promising results, based on a limited number of patients, need to be confirmed in a larger series (220).

Among ENKTCL, the introduction of asparaginase has significantly improved the prognosis of patients (221, 222). Better efficacy and tolerance have been observed with the use of pegasparaginase relative to L-asparaginase (223, 224). Although there is no international consensus concerning the treatment sequence, it is generally accepted that frontline therapy should include at least pegylated-asparaginase and gemcitabine in association with various combination of other agents or strategies (including cisplatin/oxaliplatin, dexamethasone, methotrexate, and radiotherapy), depending on the staging of the lymphoma as localized or disseminated disease (223–226).

In ATLL, the characterization of a Treg/TH2 phenotype and polarization of the tumor cells led to the development of anti-CCR4 monoclonal antibodies (227). Although this targeted therapy is currently used for refractory/relapsed patients, a recent study showed better survival of aggressive transplant-ineligible ATLL using a polychemotherapy regimen containing mogalizumab in the first line (4-year OS of 46.3% versus 20.6%, p=0.033) (228). A previous study failed to demonstrate any benefit with the addition of mogalizumab in the first line for transplant-eligible patients (229). It is still unknown whether the use of mogamulizumab could be extended in the future to other PTCLs that express CCR4, in particular, GATA3-PTCL-NOS (164).

4.2 Promising therapeutic options for relapse/refractory PTCL patients

Several ALK inhibitors have been tested in refractory/relapsed ALK-positive ALCL patients, showing an improvement in PFS and long-term complete remission (230–234). However, there are no

recommendations concerning the indication or duration of treatment.

The frequent alterations of chromatin modifiers among PTCLs has led to the development of therapies to regulate epigenetic programs. Although approved by the FDA, the use of romidepsin, pralatrexate, and belinostat did not show significant efficacy in several studies, probably due to the enrollment of patients with several PTCL entities, leading to a small sample size for each (219, 235). However, subgroup analyses showed a benefit for HDAC inhibitors for TFH-lymphomas (219, 236). Prospective studies are needed to confirm these promising results for TFH-lymphoma patients and to identify predictive biomarkers of response. Several studies using hypomethylating agents, such as 5'azacytidine, have also shown promising results in AITL, usually independently of the *TET2*, *DNMT3A*, and *IDH2* mutational status, although these studies had only small numbers of patients (237, 238). A phase 3 trial comparing the use of the oral form of the 5-azacytidine to investigator-choice treatment between gemcitabine, bendamustine, or romidepsin in relapsed/refractory THF-lymphoma patients was recently reported. The primary endpoint was PFS and was not met, likely due to the trial being underpowered. However, OS was longer for patients receiving 5-azacytidine, suggesting efficacy of the drug. The combination of oral 5-azacytidine and romidepsin has shown efficacy for frontline or refractory/relapsed PTCL patients, especially those with a TFH phenotype (239). The development of IDH2 Inhibitors in acute myeloid leukemia (240, 241) suggests their potential application in TFH-lymphomas with *IDH2* mutations.

In AITL, the identification of gene alterations enhancing the TCR pathway paved the way for the use of dasatinib, a PKC inhibitor, which showed efficacy *in vitro* and *in vivo* in a mouse *RHOA* G17V mutant *TET2* deleted model, as well as in a phase 1 trial for relapsed/refractory patients (71, 242).

The identification of structural alterations of PDL1 in ENKTCL led to studies to evaluate the use of immune checkpoint inhibitors, such as PD1 inhibitors, for refractory/relapsed patients (5, 132, 243). The response to these therapies may be predicted by characterization of the tumor immune microenvironment using gene expression profiling (Nanostring technology) or immunohistochemistry (anti-PDL1, anti-FOXP3, anti-CD68) (244). Surprisingly, although similar disruption of the 3'UTR of *PDL1* was also detected in ATLL, the use of PD1 inhibitors in this entity led to rapid progression of the disease for at least some patients (245).

Translocations involving *JAK2* leads to phosphorylation of the tyrosine kinase domain, subsequent constitutive activation, and downstream JAK/STAT pathway activation (246). This pathway is now targeted using JAK inhibitors in the clinic for myeloproliferative neoplasms and cancers with high pSTAT3 levels (247), such as ALK-negative ALCL, may be a good candidate for such targeted therapy, as suggested *in vivo* in a

TABLE 3 Relevant cytogenetic or molecular findings for the management of PTCL patients.

Entity	Diagnosis	Prognosis	Therapeutic relevance	Potential targeted therapies
TFH-lymphoma	- Mutations *RHOA* G17V, *IDH2* R172, - fusions transcript *ITK_SYK*	*DNMT3A* R882X	*ITK_SYK* *CTLA4_CD28* *FYN_TRAF3IP2*	Demethylating agents PI3K inhibitors SYK inhibitors CTLA4 inhibitors IkB inhibitors
ALK-positive ALCL	ALK expression (IHC), rearrangement (FISH), fusion transcript	MYC expression *TRAF1_ALK* fusion transcript		Brentuximab-vedotin ALK inhibitors JAK/STAT inhibitors
ALK-negative ALCL		FISH for: -*DUSP22* rearrangement - *TP63* rearrangement	JAK2 fusion transcripts pSTAT3	Brentuximab vedotin JAK/STAT inhibitors Kinase inhibitor
ENKTCL (nasal type)	EBV (EBER ISH)	MYC expression	PDL1 expression	Immune checkpoint inhibitors (pembrolizumab, nivolumab)
HSTL	Iso7q (FISH)		KIR3DL2 expression	Humanized KIR3DL2 antibodies (lacutamab) JAK/STAT inhibitors
EATL	Mutations *JAK1* p.G1097, *STAT3*			JAK/STAT inhibitors
MEITL	*SETD2* mutation/deletion	CD20 expression (favorable) *TP53* alterations, *MYC* expression		JAK/STAT inhibitors Wee1 inhibitor (adavosertib)
Indolent NK-LP of the GI tract	*STAT3 K563_C565del*			JAK-STAT inhibitors
ATLL	*HBZ* transcript	Aggressive: mutations of *CCR4* (frameshift), *TP53*, *IRF4* Indolent: *STAT3* mutations	KIR3DL2 expression	Humanized antibodies against CCR4 (mogamulizumab), KIR3DL2 (lacutamab)
PTCL-NOS		TH2 polarization DNMT3A mutations		Humanized antibodies against CCR4 (mogamulizumab)

FISH, fluorescence in situ hybridization; IHC, immunohistochemistry; ISH, *in situ* hybridization.

xenograft model (248). In a recent study, ruxolitinib showed some clinical activity on PTCLs, especially those with JAK or STAT mutations or activation (249).

Recently, the KIR3DL2 killer Immunoglobulin-like receptor was identified as a useful biomarker and therapeutic target among cutaneous T-cell lymphomas, including mycosis fungoides and Sezary syndrome (250, 251) and ATLL (252, 253). Its expression in other PTCL entities has been recently evaluated (254) and lacutamab, an anti KIR3DL2 antibody, is currently under investigation for KIR3DL2-positive PTCL (NCT04984837).

Given the limited efficacy of conventional chemotherapies, such as CHOP, for most PTCL patients, in the future, it may be worthwhile considering alternative treatment options that are personalized and directed according to the molecular characterization of the tumor (Table 3). However, whether the detection of actionable alterations will be clinically important for most PTCLs, which are still an unmet medical need for most, remains unknown.

5 Conclusion

The emergence of innovative high-throughput technologies has led to a better understanding of the pathogenesis of most PTCL entities, highlighting their diversity in terms of their biology and clinical features. A large group of TFH-lymphoma patients has emerged with a unique lymphoma oncogenesis, for which the diagnosis takes advantage of robust molecular markers and for which the treatment may benefit from the emergence of novel therapies, such as those that target epigenetics. The ALCL category is still heterogenous due to its genetic diversity, which has prognostic relevance, but may now benefit from the introduction of BV targeting CD30. The recent description of the genetic landscape of PTCL offers the rationale for an association of targeted therapies, with or without conventional chemotherapy agents, in the future, although the efficient combination for each PTCL entity or molecular subgroups still needs to be identified.

Author contributions

FD and PG wrote and supervised the manuscript. FL supervised the "oncogenic mechanisms" part, wrote and supervised the therapeutic part. All authors contributed to the article and approved the submitted version.

Funding

The authors declare that this study received funding from Force Hemato. The funder was not involved in the study design, collection, analysis, interpretation of data, the writing of the article, or the decision to submit it for publication.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IB de O, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

2. Campo E, Jaffe ES, Cook JR, Quintanilla-Martinez L, Swerdlow SH, Anderson KC, et al. The international consensus classification of mature lymphoid neoplasms: a report from the clinical advisory committee. *Blood* (2022) 140(11):1229–53. doi: 10.1182/blood.2022015851

3. de Leval L, Alizadeh AA, Bergsagel PL, Campo E, Davies A, Dogan A, et al. Genomic profiling for clinical decision making in lymphoid neoplasms. *Blood* (2022) 140(21):2193–227. doi: 10.1182/blood.2022015854

4. Kataoka K, Shiraishi Y, Takeda Y, Sakata S, Matsumoto M, Nagano S, et al. Aberrant PD-L1 expression through 3′-UTR disruption in multiple cancers. *Nature* (2016) 534(7607):402–6. doi: 10.1038/nature18294

5. Kataoka K, Miyoshi H, Sakata S, Dobashi A, Couronné L, Kogure Y, et al. Frequent structural variations involving programmed death ligands in Epstein-Barr virus-associated lymphomas. *Leukemia* (2019) 33(7):1687–99. doi: 10.1038/s41375-019-0380-5

6. Genovese G, Kähler AK, Handsaker RE, Lindberg J, Rose SA, Bakhoum SF, et al. Clonal hematopoiesis and blood-cancer risk inferred from blood DNA sequence. *N Engl J Med* (2014) 371(26):2477–87. doi: 10.1056/NEJMoa1409405

7. Couronné L, Bastard C, Bernard OA. TET2 and DNMT3A mutations in human T-cell lymphoma. *N Engl J Med* (2012) 366(1):95–6. doi: 10.1056/NEJMc1111708

8. Lemonnier F, Couronne L, Parrens M, Jais JP, Travert M, Lamant L, et al. Recurrent TET2 mutations in peripheral T-cell lymphomas correlate with TFH-like features and adverse clinical parameters. *Blood* (2012) 120(7):1466–9. doi: 10.1182/blood-2012-02-408542

9. Nicolae A, Bouilly J, Lara D, Fataccioli V, Lemonnier F, Drieux F, et al. Nodal peripheral T-cell lymphoma, NOS (PTCL, NOS) with a cytotoxic phenotype frequently occurs in clinical settings suggesting immune dysregulation and discloses recurrent epigenetic alterations. *Mod Pathol* (2022) 35(8):1126–36. doi: 10.1038/s41379-022-01022-w

10. Pastoret C, Desmots F, Drillet G, Le Gallou S, Boulland ML, Thannberger A, et al. Linking the KIR phenotype with STAT3 and *TET2* mutations to identify chronic lymphoproliferative disorders of NK cells. *Blood* (2021) 137(23):3237–50. doi: 10.1182/blood.2020006721

11. Quivoron C, Couronné L, Della Valle V, Lopez CK, Plo I, Wagner-Ballon O, et al. TET2 inactivation results in pleiotropic hematopoietic abnormalities in mouse and is a recurrent event during human lymphomagenesis. *Cancer Cell* (2011) 20(1):25–38. doi: 10.1016/j.ccr.2011.06.003

12. Yang L, Rau R, Goodell MA. DNMT3A in haematological malignancies. *Nat Rev Cancer* (2015) 15(3):152–65. doi: 10.1038/nrc3895

13. Zang S, Li J, Yang H, Zeng H, Han W, Zhang J, et al. Mutations in 5-methylcytosine oxidase TET2 and RhoA cooperatively disrupt T cell homeostasis. *J Clin Invest* (2017) 127(8):2998–3012. doi: 10.1172/JCI92026

14. Cortes JR, Ambesi-Impiombato A, Couronné L, Quinn SA, Kim CS, da Silva Almeida AC, et al. RHOA G17V induces T follicular helper cell specification and promotes lymphomagenesis. *Cancer Cell* (2018) 33(2):259–73.e7. doi: 10.1016/j.ccell.2018.01.001

15. Ng SY, Brown L, Stevenson K, deSouza T, Aster JC, Louissaint A, et al. RhoA G17V is sufficient to induce autoimmunity and promotes T-cell lymphomagenesis in mice. *Blood* (2018) 132(9):935–47. doi: 10.1182/blood-2017-11-818617

16. Lemonnier F, Cairns RA, Inoue S, Li WY, Dupuy A, Broutin S, et al. The IDH2 R172K mutation associated with angioimmunoblastic T-cell lymphoma produces 2HG in T cells and impacts lymphoid development. *Proc Natl Acad Sci* (2016) 113(52):15084–9. doi: 10.1073/pnas.1617929114

17. Lemonnier F, Poullot E, Dupuy A, Couronné L, Martin N, Scourzic L, et al. Loss of 5-hydroxymethylcytosine is a frequent event in peripheral T-cell lymphomas. *Haematologica* (2018) 103(3):e115–8. doi: 10.3324/haematol.2017.167973

18. Roberti A, Dobay MP, Bisig B, Vallois D, Boéchat C, Lanitis E, et al. Type II enteropathy-associated T-cell lymphoma features a unique genomic profile with highly recurrent SETD2 alterations. *Nat Commun* (2016) 7(1):12602. doi: 10.1038/ncomms12602

19. McKinney M, Moffitt AB, Gaulard P, Travert M, De Leval L, Nicolae A, et al. The genetic basis of hepatosplenic T-cell lymphoma. *Cancer Discov* (2017) 7(4):369–79. doi: 10.1158/2159-8290.CD-16-0330

20. Jiang L, Gu ZH, Yan ZX, Zhao X, Xie YY, Zhang ZG, et al. Exome sequencing identifies somatic mutations of DDX3X in natural killer/T-cell lymphoma. *Nat Genet* (2015) 47(9):1061–6. doi: 10.1038/ng.3358

21. Ji MM, Huang YH, Huang JY, Wang ZF, Fu D, Liu H, et al. Histone modifier gene mutations in peripheral T-cell lymphoma not otherwise specified. *Haematologica* (2018) 103(4):679–87. doi: 10.3324/haematol.2017.182444

22. Laurent C, Nicolae A, Laurent C, Le Bras F, Haioun C, Fataccioli V, et al. Gene alterations in epigenetic modifiers and JAK-STAT signaling are frequent in breast implant-associated ALCL. *Blood* (2020) 135(5):360–70. doi: 10.1182/blood.2019001904

23. Vallois D, Dobay MPD, Morin RD, Lemonnier F, Missiaglia E, Juilland M, et al. Activating mutations in genes related to TCR signaling in angioimmunoblastic and other follicular helper t-cell–derived lymphomas. *Blood* (2016) 128(11):1490–502. doi: 10.1182/blood-2016-02-698977

24. Kataoka K, Nagata Y, Kitanaka A, Shiraishi Y, Shimamura T, Yasunaga Ji, et al. Integrated molecular analysis of adult T cell leukemia/lymphoma. *Nat Genet* (2015) 47(11):1304–15. doi: 10.1038/ng.3415

25. Werner MT, Zhao C, Zhang Q, Wasik MA. Nucleophosmin-anaplastic lymphoma kinase: the ultimate oncogene and therapeutic target. *Blood* (2017) 129(7):823–31. doi: 10.1182/blood-2016-05-717793

26. Crescenzo R, Abate F, Lasorsa E, Tabbo' F, Gaudiano M, Chiesa N, et al. Convergent mutations and kinase fusions lead to oncogenic STAT3 activation in anaplastic Large cell lymphoma. *Cancer Cell* (2015) 27(4):516–32. doi: 10.1016/j.ccell.2015.03.006

27. Heavican TB, Bouska A, Yu J, Lone W, Amador C, Gong Q, et al. Genetic drivers of oncogenic pathways in molecular subgroups of peripheral T-cell lymphoma. *Blood* (2019) 133(15):1664–76. doi: 10.1182/blood-2018-09-872549

28. Watatani Y, Sato Y, Miyoshi H, Sakamoto K, Nishida K, Gion Y, et al. Molecular heterogeneity in peripheral T-cell lymphoma, not otherwise specified revealed by comprehensive genetic profiling. *Leukemia* (2019) 33(12):2867–83. doi: 10.1038/s41375-019-0473-1

29. Maura F, Dodero A, Carniti C, Bolli N, Magni M, Monti V, et al. CDKN2A deletion is a frequent event associated with poor outcome in patients with peripheral T-cell lymphoma not otherwise specified (PTCL-NOS). *Haematologica* (2020) 106(11):2918–26. doi: 10.3324/haematol.2020.262659

30. Xiong J, Cui BW, Wang N, Dai YT, Zhang H, Wang CF, et al. Genomic and transcriptomic characterization of natural killer T cell lymphoma. *Cancer Cell* (2020) 37(3):403–419.e6. doi: 10.1016/j.ccell.2020.02.005

31. Moffitt AB, Ondrejka SL, McKinney M, Rempel RE, Goodlad JR, Teh CH, et al. Enteropathy-associated T cell lymphoma subtypes are characterized by loss of function of SETD2. *J Exp Med* (2017) 214(5):1371–86. doi: 10.1084/jem.20160894

32. Vasmatzis G, Johnson SH, Knudson RA, Ketterling RP, Braggio E, Fonseca R, et al. Genome-wide analysis reveals recurrent structural abnormalities of TP63 and other p53-related genes in peripheral T-cell lymphomas. *Blood* (2012) 120(11):2280–9. doi: 10.1182/blood-2012-03-419937

33. Wang X, Boddicker RL, Dasari S, Sidhu JS, Kadin ME, Macon WR, et al. Expression of p63 protein in anaplastic large cell lymphoma: implications for genetic subtyping. *Hum Pathol* (2017) 64:19–27. doi: 10.1016/j.humpath.2017.01.003

34. Atsaves V, Tsesmetzis N, Chioureas D, Kis L, Leventaki V, Drakos E, et al. PD-L1 is commonly expressed and transcriptionally regulated by STAT3 and MYC in ALK-negative anaplastic large-cell lymphoma. *Leukemia* (2017) 31(7):1633–7. doi: 10.1038/leu.2017.103

35. Kong J, Dasari S, Feldman AL. PD-L1 expression in anaplastic large cell lymphoma. *Mod Pathol* (2020) 33(6):1232–3. doi: 10.1038/s41379-019-0448-9

36. Shen J, Li S, Medeiros LJ, Lin P, Wang SA, Tang G, et al. PD-L1 expression is associated with ALK positivity and STAT3 activation, but not outcome in patients with systemic anaplastic large cell lymphoma. *Mod Pathol* (2020) 33(3):324–33. doi: 10.1038/s41379-019-0336-3

37. Luchtel RA, Dasari S, Oishi N, Pedersen MB, Hu G, Rech KL, et al. Molecular profiling reveals immunogenic cues in anaplastic large cell lymphomas with DUSP22 rearrangements. *Blood* (2018) 132(13):1386–98. doi: 10.1182/blood-2018-03-838524

38. de Leval L, Rickman DS, Thielen C, de Reynies A, Huang YL, Delsol G, et al. The gene expression profile of nodal peripheral T-cell lymphoma demonstrates a molecular link between angioimmunoblastic T-cell lymphoma (AITL) and follicular helper T (TFH) cells. *Blood* (2007) 109(11):4952–63. doi: 10.1182/blood-2006-10-055145

39. Iqbal J, Weisenburger DD, Greiner TC, Vose JM, McKeithan T, Kucuk C, et al. Molecular signatures to improve diagnosis in peripheral T-cell lymphoma and prognostication in angioimmunoblastic T-cell lymphoma. *Blood* (2010) 115(5):1026–36. doi: 10.1182/blood-2009-06-227579

40. Iqbal J, Wright G, Wang C, Rosenwald A, Gascoyne RD, Weisenburger DD, et al. Gene expression signatures delineate biological and prognostic subgroups in peripheral T-cell lymphoma. *Blood* (2014) 123(19):2915–23. doi: 10.1182/blood-2013-11-536359

41. Cortes JR, Filip I, Albero R, Patiño-Galindo JA, Quinn SA, Lin WHW, et al. Oncogenic Vav1-Myo1f induces therapeutically targetable macrophage-rich tumor microenvironment in peripheral T cell lymphoma. *Cell Rep* (2022) 39(3):110695. doi: 10.1016/j.celrep.2022.110695

42. Lamant L, McCarthy K, d'Amore E, Klapper W, Nakagawa A, Fraga M, et al. Prognostic impact of morphologic and phenotypic features of childhood ALK-positive anaplastic Large-cell lymphoma: results of the ALCL99 study. *J Clin Oncol* (2011) 29(35):4669–76. doi: 10.1200/JCO.2011.36.5411

43. Cook LB, Melamed A, Niederer H, Valganon M, Laydon D, Foroni L, et al. The role of HTLV-1 clonality, proviral structure, and genomic integration site in adult T-cell leukemia/lymphoma. *Blood* (2014) 123(25):3925–31. doi: 10.1182/blood-2014-02-553602

44. Bangham CRM, Cook LB, Melamed A. HTLV-1 clonality in adult T-cell leukaemia and non-malignant HTLV-1 infection. *Semin Cancer Biol* (2014) 26:89–98. doi: 10.1016/j.semcancer.2013.11.003

45. Melamed A, Laydon DJ, Gillet NA, Tanaka Y, Taylor GP, Bangham CRM. Genome-wide determinants of proviral targeting, clonal abundance and expression in natural HTLV-1 infection. emerman m, éditeur. *PloS Pathog* (2013) 9(3):e1003271. doi: 10.1371/journal.ppat.1003271

46. Bangham CR, Ratner L. How does HTLV-1 cause adult T-cell leukaemia/lymphoma (ATL)? *Curr Opin Virol* (2015) 14:93–100. doi: 10.1016/j.coviro.2015.09.004

47. Bangham CRM. Human T cell leukemia virus type 1: persistence and pathogenesis. *Annu Rev Immunol* (2018) 36(1):43–71. doi: 10.1146/annurev-immunol-042617-053222

48. Wattel E, Vartanian JP, Pannetier C, Wain-Hobson S. Clonal expansion of human T-cell leukemia virus type I-infected cells in asymptomatic and symptomatic carriers without malignancy. *J Virol* (1995) 69(5):2863–8. doi: 10.1128/jvi.69.5.2863-2868.1995

49. Giam CZ, Semmes O. HTLV-1 infection and adult T-cell Leukemia/Lymphoma–a tale of two proteins: tax and HBZ. *Viruses* (2016) 8(6):161. doi: 10.3390/v8060161

50. Takeda S, Maeda M, Morikawa S, Taniguchi Y, Yasunaga Ji, Nosaka K, et al. Genetic and epigenetic inactivation oftax gene in adult T-cell leukemia cells. *Int J Cancer* (2004) 109(4):559–67. doi: 10.1002/ijc.20007

51. Matsuoka M, Green PL. The HBZ gene, a key player in HTLV-1 pathogenesis. *Retrovirology* (2009) 6(1):71. doi: 10.1186/1742-4690-6-71

52. Tabiasco J, Vercellone A, Meggetto F, Hudrisier D, Brousset P, Fournié JJ. Acquisition of viral receptor by NK cells through immunological synapse. *J Immunol* (2003) 170(12):5993–8. doi: 10.4049/jimmunol.170.12.5993

53. Lee JH, Choi J, Ahn YO, Kim TM, Heo DS. CD21-independent Epstein-Barr virus entry into NK cells. *Cell Immunol* (2018) 327:21–5. doi: 10.1016/j.cellimm.2018.01.011

54. Ng SB, Selvarajan V, Huang G, Zhou J, Feldman AL, Law M, et al. Activated oncogenic pathways and therapeutic targets in extranodal nasal-type NK/T cell lymphoma revealed by gene expression profiling: gene expression profiling in NK/T lymphoma. *J Pathol* (2011) 223(4):496–510. doi: 10.1002/path.2823

55. Lebwohl B, Ludvigsson JF, Green PHR. Celiac disease and non-celiac gluten sensitivity. *BMJ* (2015) 351:h4347. doi: 10.1136/bmj.h4347

56. Deva AK, Turner SD, Kadin ME, Magnusson MR, Prince HM, Miranda RN, et al. Etiology of breast implant-associated anaplastic Large cell lymphoma (BIA-ALCL): current directions in research. *Cancers* (2020) 12(12):3861. doi: 10.3390/cancers12123861

57. Goldman S, Bron D, Tousseyn T, Vierasu I, Dewispelaere L, Heimann P, et al. Rapid progression of angioimmunoblastic T cell lymphoma following BNT162b2 mRNA vaccine booster shot: a case report. *Front Med* (2021) 8:798095. doi: 10.3389/fmed.2021.798095

58. Li Z, Xia Y, Feng LN, Chen JR, Li HM, Cui J, et al. Genetic risk of extranodal natural killer T-cell lymphoma: a genome-wide association study. *Lancet Oncol* (2016) 17(9):1240–7. doi: 10.1016/S1470-2045(16)30148-6

59. Lin GW, Xu C, Chen K, Huang HQ, Chen J, Song B, et al. Genetic risk of extranodal natural killer T-cell lymphoma: a genome-wide association study in multiple populations. *Lancet Oncol* (2020) 21(2):306–16. doi: 10.1016/S1470-2045(19)30799-5

60. Lundin EKEA, Scott H, Hansen T, Paulsen G, Halstensen TS, Fausa O, et al. Gliadin-specitlc, HLA-DQ(alpha 1*0501,beta 1*0201) restricted T cells isolated from the small intestinal mucosa of celiac disease patients. *J Exp Med* (1993) 178:187–96. doi: 10.1084/jem.178.1.187

61. Gayden T, Sepulveda FE, Khuong-Quang DA, Pratt J, Valera ET, Garrigue A, et al. Germline HAVCR2 mutations altering TIM-3 characterize subcutaneous panniculitis-like T cell lymphomas with hemophagocytic lymphohistiocytic syndrome. *Nat Genet* (2018) 50(12):1650–7. doi: 10.1038/s41588-018-0251-4

62. Amatore F, Ortonne N, Lopez M, Orlanducci F, Castellano R, Ingen-Housz-Oro S, et al. ICOS is widely expressed in cutaneous T-cell lymphoma, and its targeting promotes potent killing of malignant cells. *Blood Adv* (2020) 4(20):5203–14. doi: 10.1182/bloodadvances.2020002395

63. Luherne C, Menguy S, Ferte T, Beylot-Barry M, Seneschal J, Milpied B, et al. A high programmed cell death protein 1 hormone receptor score on skin biopsy is associated with sézary syndrome diagnosis: a study of 91 patients with erythroderma. *Acta Derm Venereol* (2022) 102:adv00773. doi: 10.2340/actadv.v102.1062

64. Picchio MC, Scala E, Pomponi D, Caprini E, Frontani M, Angelucci I, et al. CXCL13 is highly produced by sézary cells and enhances their migratory ability via a synergistic mechanism involving CCL19 and CCL21 chemokines. *Cancer Res* (2008) 68 (17):7137–46. doi: 10.1158/0008-5472.CAN-08-0602

65. Masle-Farquhar E, Jeelall Y, White J, Bier J, Deenick EK, Brink R, et al. CARD11 gain-of-function mutation drives cell-autonomous accumulation of PD-1+ ICOShigh activated T cells, T-follicular, T-regulatory and T-follicular regulatory cells. *Front Immunol* (2023) 14:1095257. doi: 10.3389/fimmu.2023.1095257

66. Cairns RA, Iqbal J, Lemonnier F, Kucuk C, De Leval L, Jais JP, et al. IDH2 mutations are frequent in angioimmunoblastic T-cell lymphoma. *Blood* (2012) 119 (8):1901–3. doi: 10.1182/blood-2011-11-391748

67. Sakata-Yanagimoto M, Enami T, Yoshida K, Shiraishi Y, Ishii R, Miyake Y, et al. Somatic RHOA mutation in angioimmunoblastic T cell lymphoma. *Nat Genet* (2014) 46(2):171–5. doi: 10.1038/ng.2872

68. Palomero T, Couronné L, Khiabanian H, Kim MY, Ambesi-Impiombato A, Perez-Garcia A, et al. Recurrent mutations in epigenetic regulators, RHOA and FYN kinase in peripheral T cell lymphomas. *Nat Genet* (2014) 46(2):166–70. doi: 10.1038/ng.2873

69. Odejide O, Weigert O, Lane AA, Toscano D, Lunning MA, Kopp N, et al. A targeted mutational landscape of angioimmunoblastic T-cell lymphoma. *Blood* (2014) 123(9):1293–6. doi: 10.1182/blood-2013-10-531509

70. Abate F, da Silva-Almeida AC, Zairis S, Robles-Valero J, Couronne L, Khiabanian H, et al. Activating mutations and translocations in the guanine exchange factor VAV1 in peripheral T-cell lymphomas. *Proc Natl Acad Sci* (2017) 114(4):764–9. doi: 10.1073/pnas.1608839114

71. Fujisawa M, Sakata-Yanagimoto M, Nishizawa S, Komori D, Gershon P, Kiryu M, et al. Activation of RHOA–VAV1 signaling in angioimmunoblastic T-cell lymphoma. *Leukemia* (2018) 32(3):694–702. doi: 10.1038/leu.2017.273

72. Lee SH, Kim JS, Kim J, Kim SJ, Kim WS, Lee S, et al. A highly recurrent novel missense mutation in CD28 among angioimmunoblastic T-cell lymphoma patients. *Haematologica* (2015) 100(12):e505. doi: 10.3324/haematol.2015.133074

73. Rohr J, Guo S, Huo J, Bouska A, Lachel C, Li Y, et al. Recurrent activating mutations of CD28 in peripheral T-cell lymphomas. *Leukemia* (2016) 30(5):1062–70. doi: 10.1038/leu.2015.357

74. Nguyen TB, Sakata-Yanagimoto M, Asabe Y, Matsubara D, Kano J, Yoshida K, et al. Identification of cell-type-specific mutations in nodal T-cell lymphomas. *Blood Cancer J* (2017) 7(1):e516–e516. doi: 10.1038/bcj.2016.122

75. Wang C, McKeithan TW, Gong Q, Zhang W, Bouska A, Rosenwald A, et al. IDH2R172 mutations define a unique subgroup of patients with angioimmunoblastic T-cell lymphoma. *Blood* (2015) 126(15):1741–52. doi: 10.1182/blood-2015-05-644591

76. Yoo HY, Sung MK, Lee SH, Kim S, Lee H, Park S, et al. A recurrent inactivating mutation in RHOA GTPase in angioimmunoblastic T cell lymphoma. *Nat Genet* (2014) 46(4):371–5. doi: 10.1038/ng.2916

77. Nakamoto-Matsubara R, Sakata-Yanagimoto M, Enami T, Yoshida K, Yanagimoto S, Shiozawa Y, et al. Detection of the G17V RHOA mutation in angioimmunoblastic T-cell lymphoma and related lymphomas using quantitative allele-specific PCR. *PloS One* (2014) 9(10):e109714. doi: 10.1371/journal.pone.0109714

78. Dobay MP, Lemonnier F, Missiaglia E, Bastard C, Vallois D, Jais JP, et al. Integrative clinicopathological and molecular analyses of angioimmunoblastic T-cell lymphoma and other nodal lymphomas of follicular helper T-cell origin. *Haematologica* (2017) 102(4):e148–51. doi: 10.3324/haematol.2016.158428

79. Leca J, Lemonnier F, Meydan C, Foox J, El Ghamrasni S, Mboumba DL, et al. IDH2 and TET2 mutations synergize to modulate T follicular helper cell functional interaction with the AITL microenvironment. *Cancer Cell* (2023) 41(2):323–39.e10. doi: 10.1016/j.ccell.2023.01.003

80. Guo. Novel fusion transcripts identified in angioimmunoblastic T cell lymphoma. *Mod Pathol* (2013) 26):330A.

81. Vallois D, Dupuy A, Lemonnier F, Allen G, Missiaglia E, Fataccioli V, et al. RNA Fusions involving CD28 are rare in peripheral T-cell lymphomas and concentrate mainly in those derived from follicular helper T cells. *Haematologica* (2018) 103(8): e360–3. doi: 10.3324/haematol.2017.186767

82. Boddicker RL, Razidlo GL, Dasari S, Zeng Y, Hu G, Knudson RA, et al. Integrated mate-pair and RNA sequencing identifies novel, targetable gene fusions in peripheral T-cell lymphoma. *Blood* (2016) 128(9):1234–45. doi: 10.1182/blood-2016-03-707141

83. Streubel B, Vinatzer U, Willheim M, Raderer M, Chott A. Novel t (5; 9)(q33; q22) fuses ITK to SYK in unspecified peripheral T-cell lymphoma. *Leukemia* (2006) 20 (2):313–8. doi: 10.1038/sj.leu.2404045

84. Attygalle AD, Feldman AL, Dogan A. ITK/SYK translocation in angioimmunoblastic T-cell lymphoma. *Am J Surg Pathol* (2013) 37(9):1456–7. doi: 10.1097/PAS.0b013e3182991415

85. Boi M, Zucca E, Inghirami G, Bertoni F. Advances in understanding the pathogenesis of systemic anaplastic large cell lymphomas. *Br J Haematol* (2015) 168 (6):771–83. doi: 10.1111/bjh.13265

86. Andraos E, Dignac J, Meggetto F. NPM-ALK: a driver of lymphoma pathogenesis and a therapeutic target. *Cancers* (2021) 13(1):144. doi: 10.3390/cancers13010144

87. Feldman AL, Vasmatzis G, Asmann YW, Davila J, Middha S, Eckloff BW, et al. Novel TRAF1-ALK fusion identified by deep RNA sequencing of anaplastic large cell lymphoma: *TRAF1-ALK* fusion in ALCL. *Genes Chromosomes Cancer* (2013) 52 (11):1097–102. doi: 10.1002/gcc.22104

88. The European T-cell Lymphoma Study Group, Abate F, Todaro M, van der Krogt JA, Boi M, Landra I, et al. A novel patient-derived tumorgraft model with TRAF1-ALK anaplastic large-cell lymphoma translocation. *Leukemia* (2015) 29 (6):1390–401. doi: 10.1038/leu.2014.347

89. Larose H, Prokoph N, Matthews JD, Schlederer M, Högler S, Alsulami AF, et al. Whole exome sequencing reveals NOTCH1 mutations in anaplastic large cell lymphoma and points to notch both as a key pathway and a potential therapeutic target. *Haematologica* (2021) 106(6):1693–704. doi: 10.3324/haematol.2019.238766

90. Morris S, Kirstein M, Valentine M, Dittmer K, Shapiro D, Saltman D, et al. Fusion of a kinase gene, ALK, to a nucleolar protein gene, NPM, in non-hodgkin's lymphoma. *Science* (1994) 263(5151):1281–4. doi: 10.1126/science.8122112

91. Lamant L, Dastugue N, Pulford K, Delsol G, Mariame B. A new fusion gene TPM3-ALK in anaplastic Large cell lymphoma created by a (1;2)(q25;p23) translocation. *Blood* (1999) 93(9):3088–95.

92. Trinei M, Lanfrancone L, Campo E, Pulford K, Mason DY, Pelicci PG, et al. A new variant anaplastic lymphoma kinase (ALK)-fusion protein (ATIC-ALK) in a case of ALK-positive anaplastic Large cell lymphoma. *Cancer Res* (2000) 60(4):793–8.

93. Hernández L, Pinyol M, Hernández S, Beà S, Pulford K, Rosenwald A, et al. TRK-fused gene (TFG) is a new partner of ALK in anaplastic Large cell lymphoma producing two structurally DifferentTFG-ALK translocations. *Blood* (1999) 94(9):3265 –8. doi: 10.1182/blood.V94.9.3265

94. Touriol C, Greenland C, Lamant L, Pulford K, Mason DY, Delsol G. Further demonstration of the diversity of chromosomal changes involving 2p23 in ALK-positive lymphoma: 2 cases expressing ALK kinase fused to CLTCL (clathrin chain polypeptide-like). *Blood* (2000) 95(10):3204–7. doi: 10.1182/blood.V95.10.3204

95. Tort F, Pinyol M, Pulford K, Roncador G, Hernandez L, Nayach I, et al. Molecular characterization of a new ALK translocation involving moesin (MSN-ALK) in anaplastic Large cell lymphoma. *Lab Invest* (2001) 81(3):419–26. doi: 10.1038/labinvest.3780249

96. Cools J, Wlodarska I, Somers R, Mentens N, Pedeutour F, Maes B, et al. Identification of novel fusion partners of ALK, the anaplastic lymphoma kinase, in anaplastic large-cell lymphoma and inflammatory myofibroblastic tumor: identification of novel ALK fusion partners. *Genes Chromosomes Cancer* (2002) 34 (4):354–62. doi: 10.1002/gcc.10033

97. Lamant L, Gascoyne RD, Duplantier MM, Armstrong F, Raghab A, Chhanabhai M, et al. Non-muscle myosin heavy chain (MYH9): a new partner fused to ALK in anaplastic large cell lymphoma. *Genes Chromosomes Cancer* (2003) 37(4):427–32. doi: 10.1002/gcc.10232

98. Palacios G, Shaw TI, Li Y, Singh RK, Valentine M, Sandlund JT, et al. Novel ALK fusion in anaplastic large cell lymphoma involving EEF1G, a subunit of the eukaryotic elongation factor-1 complex. *Leukemia* (2017) 31(3):743–7. doi: 10.1038/leu.2016.331

99. Graetz D, Crews KR, Azzato EM, Singh RK, Raimondi S, Mason J, et al. Leukemic presentation of ALK-positive anaplastic large cell lymphoma with a novel partner, poly(A) binding protein cytoplasmic 1 (PABPC1), responding to single-agent crizotinib. *Haematologica* (2019) 104(5):e218–21. doi: 10.3324/haematol.2018.215103

100. Sibon D, Nguyen DP, Schmitz N, Suzuki R, Feldman AL, Gressin R, et al. ALK-positive anaplastic large-cell lymphoma in adults: an individual patient data pooled analysis of 263 patients. *Haematologica* (2019) 104(12):e562–5. doi: 10.3324/haematol.2018.213512

101. Cederleuf H, Bjerregård Pedersen M, Jerkeman M, Relander T, d'Amore F, Ellin F. The addition of etoposide to CHOP is associated with improved outcome in ALK+ adult anaplastic large cell lymphoma: a Nordic lymphoma group study. *Br J Haematol* (2017) 178(5):739–46. doi: 10.1111/bjh.14740

102. Brink M, Meeuwes FO, van der Poel M, Kersten M, Wondergem M, Mutsaers PGNJ, et al. Impact of etoposide and ASCT on survival among patients aged,65 years with stage II to IV PTCL: a population-based cohort study. *Blood* (2022) 140(9):1009 –19. doi: 10.1182/blood.2021015114

103. Horwitz S, O'Connor OA, Pro B, Illidge T, Fanale M, Advani R, et al. Brentuximab vedotin with chemotherapy for CD30-positive peripheral T-cell lymphoma (ECHELON-2): a global, double-blind, randomised, phase 3 trial. *Lancet* (2019) 393(10168):229–40. doi: 10.1016/S0140-6736(18)32984-2

104. Moritake H, Shimonodan H, Marutsuka K, Kamimura S, Kojima H, Nunoi H. C-MYC rearrangement may induce an aggressive phenotype in anaplastic lymphoma kinase positive anaplastic large cell lymphoma: identification of a novel fusion gene ALO17/C-MYC. *Am J Hematol* (2011) 86(1):75–8. doi: 10.1002/ajh.21887

105. Raetz EA, Perkins SL, Carlson MA, Schooler KP, Carroll WL, Virshup DM. The nucleophosmin-anaplastic lymphoma kinase fusion protein induces c-myc expression in pediatric anaplastic Large cell lymphomas. *Am J Pathol* (2002) 161 (3):875–83. doi: 10.1016/S0002-9440(10)64248-4

106. Feldman AL, Dogan A, Smith DI, Law ME, Ansell SM, Johnson SH, et al. Discovery of recurrent t(6;7)(p25.3;q32.3) translocations in ALK-negative anaplastic large cell lymphomas by massively parallel genomic sequencing. *Blood* (2011) 117 (3):915–9. doi: 10.1182/blood-2010-08-303305

107. King RL, Dao LN, McPhail ED, Jaffe ES, Said J, Swerdlow SH, et al. Morphologic features of ALK-negative anaplastic large cell lymphomas with DUSP22 rearrangements. *Am J Surg Pathol* (2016) 40(1):36–43. doi: 10.1097/PAS.0000000000000500

108. Mélard P, Idrissi Y, Andrique L, Poglio S, Prochazkova-Carlotti M, Berhouet S, et al. Molecular alterations and tumor suppressive function of the DUSP22 (Dual specificity phosphatase 22) gene in peripheral T-cell lymphoma subtypes. *Oncotarget* (2016) 7(42):68734–48. doi: 10.18632/oncotarget.11930

109. Onaindia A, de Villambrosía SG, Prieto-Torres L, Rodríguez-Pinilla SM, Montes-Moreno S, González-Vela C, et al. DUSP22 -rearranged anaplastic lymphomas are characterized by specific morphological features and a lack of cytotoxic and JAK/STAT surrogate markers. *Haematologica* (2019) 104(4):e158–62. doi: 10.3324/haematol.2018.205880

110. Ravindran A, Feldman AL, Ketterling RP, Dasari S, Rech KL, McPhail ED, et al. Striking association of lymphoid enhancing factor (LEF1) overexpression and DUSP22 rearrangements in anaplastic Large cell lymphoma. *Am J Surg Pathol* (2020) 45(4):550–7. doi: 10.1097/PAS.0000000000001614

111. Luchtel RA, Zimmermann MT, Hu G, Dasari S, Jiang M, Oishi N, et al. Recurrent MSCE116K mutations in ALK-negative anaplastic large cell lymphoma. *Blood* (2019) 133(26):2776–89. doi: 10.1182/blood.2019000626

112. Parrilla Castellar ER, Jaffe ES, Said JW, Swerdlow SH, Ketterling RP, Knudson RA, et al. ALK-negative anaplastic large cell lymphoma is a genetically heterogeneous disease with widely disparate clinical outcomes. *Blood* (2014) 124(9):1473–80. doi: 10.1182/blood-2014-04-571091

113. Pedersen MBa, Hamilton-Dutoit SJ, Bendix K, Ketterling RP, Bedroske PP, Luoma IM, et al. DUSP22 and TP63 rearrangements predict outcome of ALK-negative anaplastic large cell lymphoma: a Danish cohort study. *Blood* (2017) 130(4):554–7. doi: 10.1182/blood-2016-12-755496

114. Hapgood G, Ben-Neriah S, Mottok A, Lee DG, Villa D, Sehn LH, et al. Identification of high-risk DUSP22-rearranged ALK-negative anaplastic large cell lymphoma. *Br J Haematol* (2019) 186(3):e28–31. doi: 10.1111/bjh.15860

115. Sibon D, Bisig B, Bonnet C, Bachy E, Cavalieri D, Fataccioli V, et al. Impact du DUSP22 rearrangement on the prognosis of systemic ALK-negative anaplastic large cell lymphomas: a LYSA and TENOMIC study. *Hematol Oncol* (2021) 39(S2): hon.137_2880. doi: 10.1002/hon.137_2880

116. Scarfò I, Pellegrino E, Mereu E, Kwee I, Agnelli L, Bergaggio E, et al. Identification of a new subclass of ALK-negative ALCL expressing aberrant levels of ERBB4 transcripts. *Blood* (2016) 127(2):221–32. doi: 10.1182/blood-2014-12-614503

117. Wang Jc, Zhong Lh, Lin Wq, Zhang Wf, Xi Yf, Liu Yp, et al. JAK/STAT3 signaling activation related to distinct clinicopathologic features in systemic ALK–anaplastic Large cell lymphomas. *Am J Surg Pathol* (2023) 47(1):55–64. doi: 10.1097/PAS.0000000000001995

118. Hu G, Dasari S, Asmann YW, Greipp PT, Knudson RA, Benson HK, et al. Targetable fusions of the FRK tyrosine kinase in ALK-negative anaplastic large cell lymphoma. *Leukemia* (2018) 32(2):565–9. doi: 10.1038/leu.2017.309

119. Fitzpatrick MJ, Massoth LR, Marcus C, Vergilio JA, Severson E, Duncan D, et al. JAK2 rearrangements are a recurrent alteration in CD30+ systemic T-cell lymphomas with anaplastic morphology. *Am J Surg Pathol* (2021) 45(7):895–904. doi: 10.1097/PAS.0000000000001708

120. Kadin ME, Morgan J, Xu H, Epstein AL, Sieber D, Hubbard BA, et al. IL-13 is produced by tumor cells in breast implant–associated anaplastic large cell lymphoma: implications for pathogenesis. *Hum Pathol* (2018) 78:54–62. doi: 10.1016/j.humpath.2018.04.007

121. Blombery P, Thompson ER, Jones K, Arnau GM, Lade S, Markham JF, et al. Whole exome sequencing reveals activating JAK1 and STAT3 mutations in breast implant-associated anaplastic large cell lymphoma anaplastic large cell lymphoma. *Haematologica* (2016) 101(9):e387–90. doi: 10.3324/haematol.2016.146118

122. Los-de Vries GT, de Boer M, van Dijk E, Stathi P, Hijmering NJ, Roemer MGM, et al. Chromosome 20 loss is characteristic of breast implant–associated anaplastic large cell lymphoma. *Blood* (2020) 136(25):2927–32. doi: 10.1182/blood.2020005372

123. Tabanelli V, Corsini C, Fiori S, Agostinelli C, Calleri A, Orecchioni S, et al. Recurrent PDL1 expression and PDL1 (CD274) copy number alterations in breast implant–associated anaplastic large cell lymphomas. *Hum Pathol* (2019) 90:60–9. doi: 10.1016/j.humpath.2019.05.007

124. Quesada AE, Zhang Y, Ptashkin R, Ho C, Horwitz S, Benayed R, et al. Next generation sequencing of breast implant-associated anaplastic large cell lymphomas reveals a novel STAT3-JAK2 fusion among other activating genetic alterations within the *JAK-STAT* pathway. *Breast J* (2021) 27(4):314–21. doi: 10.1111/tbj.14205

125. Feldman AL, Oishi N, Ketterling RP, Ansell SM, Shi M, Dasari S. Immunohistochemical approach to genetic subtyping of anaplastic Large cell lymphoma. *Am J Surg Pathol* (2022) 46(11):1490–9. doi: 10.1097/PAS.0000000000001941

126. Hong M, Lee T, Young Kang S, Kim SJ, Kim W, Ko YH. Nasal-type NK/T-cell lymphomas are more frequently T rather than NK lineage based on T-cell receptor gene, RNA, and protein studies: lineage does not predict clinical behavior. *Mod Pathol* (2016) 29(5):430–43. doi: 10.1038/modpathol.2016.47

127. Koo GC, Tan SY, Tang T, Poon SL, Allen GE, Tan L, et al. Janus kinase 3–activating mutations identified in natural Killer/T-cell lymphoma. *Cancer Discovery* (2012) 2(7):591–7. doi: 10.1158/2159-8290.CD-12-0028

128. Quintanilla-Martinez L, Kremer M, Keller G, Nathrath M, Gamboa-Dominguez A, Meneses A, et al. p53 mutations in nasal natural Killer/T-cell lymphoma from Mexico. *Am J Pathol* (2001) 159(6):2095–105. doi: 10.1016/S0002-9440(10)63061-1

129. Huang Y, de Reyniès A, de Leval L, Ghazi B, Martin-Garcia N, Travert M, et al. Gene expression profiling identifies emerging oncogenic pathways operating in extranodal NK/T-cell lymphoma, nasal type. *Blood* (2010) 115(6):1226–37. doi: 10.1182/blood-2009-05-221275

130. Iqbal J, Weisenburger DD, Chowdhury A, Tsai MY, Srivastava G, Greiner TC, et al. Natural killer cell lymphoma shares strikingly similar molecular features with a group of non-hepatosplenic γδ T-cell lymphoma and is highly sensitive to a novel aurora kinase a inhibitor *in vitro*. *Leukemia* (2011) 25(2):348–58. doi: 10.1038/leu.2010.255

131. Karube K, Nakagawa M, Tsuzuki S, Takeuchi I, Honma K, Nakashima Y, et al. Identification of FOXO3 and PRDM1 as tumor-suppressor gene candidates in NK-cell neoplasms by genomic and functional analyses. *Blood* (2011) 118(12):3195–204. doi: 10.1182/blood-2011-04-346890

132. Kwong YL, Chan TSY, Tan D, Kim SJ, Poon LM, Mow B, et al. PD1 blockade with pembrolizumab is highly effective in relapsed or refractory NK/T-cell lymphoma failing l-asparaginase. *Blood* (2017) 129(17):2437–42. doi: 10.1182/blood-2016-12-756841

133. Wai CMM, Chen S, Phyu T, Fan S, Leong SM, Zheng W, et al. Immune pathway upregulation and lower genomic instability distinguish EBV-positive nodal T/NK-cell lymphoma from ENKTL and PTCL-NOS. *Haematologica* (2022) 107(8):1864–79. doi: 10.3324/haematol.2021.280003

134. Dufva O, Kankainen M, Kelkka T, Sekiguchi N, Awad SA, Eldfors S, et al. Aggressive natural killer-cell leukemia mutational landscape and drug profiling highlight JAK-STAT signaling as therapeutic target. *Nat Commun* (2018) 9(1):1567. doi: 10.1038/s41467-018-03987-2

135. Huang L, Liu D, Wang N, Ling S, Tang Y, Wu J, et al. Integrated genomic analysis identifies deregulated JAK/STAT-MYC-biosynthesis axis in aggressive NK-cell leukemia. *Cell Res* (2018) 28(2):172–86. doi: 10.1038/cr.2017.146

136. El Hussein S, Patel KP, Fang H, Thakral B, Loghavi S, Kanagal-Shamanna R, et al. Genomic and immunophenotypic landscape of aggressive NK-cell leukemia. *Am J Surg Pathol* (2020) 44(9):1235–43. doi: 10.1097/PAS.0000000000001518

137. Okuno Y, Murata T, Sato Y, Muramatsu H, Ito Y, Watanabe T, et al. Defective Epstein–Barr virus in chronic active infection and haematological malignancy. *Nat Microbiol* (2019) 4(3):404–13. doi: 10.1038/s41564-018-0334-0

138. Gessain A, Cassar O. Epidemiological aspects and world distribution of HTLV-1 infection. *Front Microbiol* (2012) 3:388/abstract(388). doi: 10.3389/fmicb.2012.00388/abstract

139. Bonzheim I, Geissinger E, Tinguely M, Roth S, Grieb T, Reimer P, et al. Evaluation of FoxP3 expression in peripheral T-cell lymphoma. *Am J Clin Pathol* (2008) 130(4):613–9. doi: 10.1309/L65GWEQ803PP6VX1

140. Karube K, Aoki R, Sugita Y, Yoshida S, Nomura Y, Shimizu K, et al. The relationship of FOXP3 expression and clinicopathological characteristics in adult T-cell leukemia/lymphoma. *Mod Pathol* (2008) 21(5):617–25. doi: 10.1038/modpathol.2008.25

141. Ferreira CR, Zhao S, Sahoo MK, Pinsky B, Weber J, Lage LAPC, et al. FOXP3-positive T-cell lymphomas in non-HTLV1 carriers include ALK-negative anaplastic large cell lymphoma: expanding the spectrum of T-cell lymphomas with regulatory phenotype. *Hum Pathol* (2018) 80:138–44. doi: 10.1016/j.humpath.2018.06.001

142. Satou A, Asano N, Kato S, Katsuya H, Ishitsuka K, Elsayed AA, et al. FoxP3-positive T cell lymphoma arising in non-HTLV1 carrier: clinicopathological analysis of 11 cases of PTCL-NOS and 2 cases of mycosis fungoides. *Histopathology* (2016) 68 (7):1099–108. doi: 10.1111/his.12885

143. Marçais A, Lhermitte L, Artesi M, Laurent C, Durkin K, Hahaut V, et al. Targeted deep sequencing reveals clonal and subclonal mutational signatures in adult T-cell leukemia/lymphoma and defines an unfavorable indolent subtype. *Leukemia* (2021) 35(3):764–76. doi: 10.1038/s41375-020-0900-3

144. Yoshida N, Shigemori K, Donaldson N, Trevisani C, Cordero NA, Stevenson KE, et al. Genomic landscape of young ATLL patients identifies frequent targetable CD28 fusions. *Blood* (2020) 135(17):1467–71. doi: 10.1182/blood.2019001815

145. Kataoka K, Iwanaga M, Yasunaga Ji, Nagata Y, Kitanaka A, Kameda T, et al. Prognostic relevance of integrated genetic profiling in adult T-cell leukemia/lymphoma. *Blood* (2018) 131(2):215–25. doi: 10.1182/blood-2017-01-761874

146. Yoshida N, Miyoshi H, Kato T, Sakata-Yanagimoto M, Niino D, Taniguchi H, et al. *CCR4* frameshift mutation identifies a distinct group of adult T cell leukaemia/lymphoma with poor prognosis: *CCR4* mutation in ATLL. *J Pathol* (2016) 238(5):621–6. doi: 10.1002/path.4699

147. Obermann E, Diss T, Hamoudi R, Munson P, Wilkins B, Camozzi M, et al. Loss of heterozygosity at chromosome 9p21 is a frequent finding in enteropathy-type T-cell lymphoma: loss of heterozygosity at 9p21 in enteropathy-type T-cell lymphoma. *J Pathol* (2004) 202(2):252–62. doi: 10.1002/path.1506

148. Cording S, Lhermitte L, Malamut G, Berrabah S, Trinquand A, Guegan N, et al. Oncogenetic landscape of lymphomagenesis in coeliac disease. *Gut* (2021) 0):1–12. doi: 10.1136/gutjnl-2020-322935

149. Nicolae A, Xi L, Pham TH, Pham TA, Navarro W, Meeker HG, et al. Mutations in the JAK/STAT and RAS signaling pathways are common in intestinal T-cell lymphomas. *Leukemia* (2016) 30(11):2245–7. doi: 10.1038/leu.2016.178

150. Hang JF, Yuan CT, Chang KC, Wang RC, Chen BJ, Hsieh PP, et al. Targeted next-generation sequencing reveals a wide morphologic and immunophenotypic spectrum of monomorphic epitheliotropic intestinal T-cell lymphoma. *Am J Surg Pathol* (2022) 46(9):1207–18. doi: 10.1097/PAS.0000000000001914

151. Veloza L, Cavalieri D, Missiaglia E, Ledoux-Pilon A, Bisig B, Pereira B, et al. Monomorphic epitheliotropic intestinal T-cell lymphoma comprises morphologic and genomic heterogeneity impacting outcome. *Haematologica* (2022) 108(1):181–95. doi: 10.3324/haematol.2022.281226

152. Nairismägi ML, Tan J, Lim JQ, Nagarajan S, Ng CCY, Rajasegaran V, et al. JAK-STAT and G-protein-coupled receptor signaling pathways are frequently altered in epitheliotropic intestinal T-cell lymphoma. *Leukemia* (2016) 30(6):1311–9. doi: 10.1038/leu.2016.13

153. Sharma A, Oishi N, Boddicker RL, Hu G, Benson HK, Ketterling RP, et al. Recurrent STAT3-JAK2 fusions in indolent T-cell lymphoproliferative disorder of the gastrointestinal tract. *Blood* (2018) 131(20):2262–6. doi: 10.1182/blood-2018-01-830968

154. Soderquist CR, Patel N, Murty VV, Betman S, Aggarwal N, Young KH, et al. Genetic and phenotypic characterization of indolent T-cell lymphoproliferative disorders of the gastrointestinal tract. *Haematologica* (2020) 105(7):1895–906. doi: 10.3324/haematol.2019.230961

155. Margolskee E, Jobanputra V, Lewis SK, Alobeid B, Green PHR, Bhagat G. Indolent small intestinal CD4+ T-cell lymphoma is a distinct entity with unique biologic and clinical features. tran DQ, éditeur. *PloS One* (2013) 8(7):e68343. doi: 10.1371/journal.pone.0068343

156. Perry AM, Bailey NG, Bonnett M, Jaffe ES, Chan WC. Disease progression in a patient with indolent T-cell lymphoproliferative disease of the gastrointestinal tract. *Int J Surg Pathol* (2019) 27(1):102–7. doi: 10.1177/1066896918785985

157. Xiao W, Gupta GK, Yao J, Jang YJ, Xi L, Baik J, et al. Recurrent somatic JAK3 mutations in NK-cell enteropathy. *Blood* (2019) 134(12):986–91. doi: 10.1182/blood.2019001443

158. Jonveaux P, Daniel MT, Martel V, Maarek O, Berger R. Isochromosome 7q and trisomy 8 are consistent primary, non-random chromosomal abnormalities associated with hepatosplenic T gamma/delta lymphoma. *Leukemia* (1996) 10(9):1453–5.

159. Wlodarska I, Martin-Garcia N, Achten R, De Wolf-Peeters C, Pauwels P, Tulliez M, et al. Fluorescence in situ hybridization study of chromosome 7 aberrations in hepatosplenic T-cell lymphoma: isochromosome 7q as a common abnormality accumulating in forms with features of cytologic progression: isochromosome 7q in hepatosplenic lymphoma. *Genes Chromosomes Cancer* (2002) 33(3):243–51. doi: 10.1002/gcc.10021

160. Nicolae A, Xi L, Pittaluga S, Abdullaev Z, Pack SD, Chen J, et al. Frequent STAT5B mutations in γδ hepatosplenic T-cell lymphomas. *Leukemia* (2014) 28 (11):2244–8. doi: 10.1038/leu.2014.200

161. Küçük C, Jiang B, Hu X, Zhang W, Chan JKC, Xiao W, et al. Activating mutations of STAT5B and STAT3 in lymphomas derived from γδ-T or NK cells. *Nat Commun* (2015) 6(1):6025. doi: 10.1038/ncomms7025

162. Travert M, Huang Y, de Leval L, Martin-Garcia N, Delfau-Larue MH, Berger F, et al. Molecular features of hepatosplenic T-cell lymphoma unravels potential novel therapeutic targets. *Blood* (2012) 119(24):5795–806. doi: 10.1182/blood-2011-12-396150

163. Song W, Zhang H, Yang F, Nakahira K, Wang C, Shi K, et al. Single cell profiling of γδ hepatosplenic T-cell lymphoma unravels tumor cell heterogeneity associated with disease progression. *Cell Oncol* (2023) 46(1):211–26. doi: 10.1007/s13402-022-00745-x

164. Amador C, Greiner TC, Heavican TB, Smith LM, Galvis KT, Lone W, et al. Reproducing the molecular subclassification of peripheral T-cell lymphoma–NOS by immunohistochemistry. *Blood* (2019) 134(24):2159–70. doi: 10.1182/blood.2019000779

165. Wang T, Feldman AL, Wada DA, Lu Y, Polk A, Briski R, et al. GATA-3 expression identifies a high-risk subset of PTCL, NOS with distinct molecular and clinical features. *Blood* (2014) 123(19):3007–15. doi: 10.1182/blood-2013-12-544809

166. Schatz JH, Horwitz SM, Teruya-Feldstein J, Lunning MA, Viale A, Huberman K, et al. Targeted mutational profiling of peripheral T-cell lymphoma not otherwise specified highlights new mechanisms in a heterogeneous pathogenesis. *Leukemia* (2015) 29(1):237–41. doi: 10.1038/leu.2014.261

167. Laginestra MA, Cascione L, Motta G, Fuligni F, Agostinelli C, Rossi M, et al. Whole exome sequencing reveals mutations in FAT1 tumor suppressor gene clinically impacting on peripheral T-cell lymphoma not otherwise specified. *Mod Pathol* (2020) 33(2):179–87. doi: 10.1038/s41379-019-0279-8

168. Almire C, Bertrand P, Ruminy P, Maingonnat C, Wlodarska I, Martín-Subero JI, et al. PVRL2 is translocated to theTRA@ locus in t(14;19)(q11;q13)-positive peripheral T-cell lymphomas. *Genes Chromosomes Cancer* (2007) 46(11):1011–8. doi: 10.1002/gcc.20490

169. Leich E, Haralambieva E, Zettl A, Chott A, Rüdiger T, Höller S, et al. Tissue microarray-based screening for chromosomal breakpoints affecting the T-cell receptor gene loci in mature T-cell lymphomas. *J Pathol* (2007) 213(1):99–105. doi: 10.1002/path.2196

170. Asano N, Suzuki R, Kagami Y, Ishida F, Kitamura K, Fukutani H, et al. Clinicopathologic and prognostic significance of cytotoxic molecule expression in nodal peripheral T-cell lymphoma, unspecified. *Am J Surg Pathol* (2005) 29(10):1284–93. doi: 10.1097/01.pas.0000173238.17331.6b

171. Feldman AL, Law M, Remstein ED, Macon WR, Erickson LA, Grogg KL, et al. Recurrent translocations involving the IRF4 oncogene locus in peripheral T-cell lymphomas. *Leukemia* (2009) 23(3):574–80. doi: 10.1038/leu.2008.320

172. Hsi ED, Said J, Macon WR, Rodig SJ, Ondrejka SL, Gascoyne RD, et al. Diagnostic accuracy of a defined immunophenotypic and molecular genetic approach for peripheral T/NK-cell lymphomas: a north American PTCL study group project. *Am J Surg Pathol* (2014) 38(6):768–75. doi: 10.1097/PAS.0000000000000188

173. Hsi ED, Horwitz SM, Carson KR, Pinter-Brown LC, Rosen ST, Pro B, et al. Analysis of peripheral T-cell lymphoma diagnostic workup in the united states. *Clin Lymphoma Myeloma Leuk* (2017) 17(4):193–200. doi: 10.1016/j.clml.2016.10.001

174. Laurent C, Baron M, Amara N, Haioun C, Dandoit M, Maynadié M, et al. Impact of expert pathologic review of lymphoma diagnosis: study of patients from the French lymphopath network. *J Clin Oncol* (2017) 35(18):2008–17. doi: 10.1200/JCO.2016.71.2083

175. Smith JL, Hodges E, Quin CT, McCarthy KP, Wright DH. Frequent T and b cell oligoclones in histologically and immunophenotypically characterized angioimmunoblastic lymphadenopathy. *Am J Pathol* (2000) 156(2):661–9. doi: 10.1016/S0002-9440(10)64770-0

176. Langerak AW, Groenen PJTA, Brüggemann M, Beldjord K, Bellan C, Bonello L, et al. EuroClonality/BIOMED-2 guidelines for interpretation and reporting of Ig/TCR clonality testing in suspected lymphoproliferations. *Leukemia* (2012) 26(10):2159–71. doi: 10.1038/leu.2012.246

177. on behalf of the EuroClonality-NGS Working Group, Scheijen B, Meijers RWJ, Rijntjes J, van der Klift MY, Möbs M, et al. Next-generation sequencing of immunoglobulin gene rearrangements for clonality assessment: a technical feasibility study by EuroClonality-NGS. *Leukemia* (2019) 33(9):2227–40. doi: 10.1038/s41375-019-0508-7

178. Oon ML, Lim JQ, Lee B, Leong SM, Soon GST, Wong ZW, et al. T-Cell lymphoma clonality by copy number variation analysis of T-cell receptor genes. *Cancers* (2021) 13(2):340. doi: 10.3390/cancers13020340

179. Chen YL, Ho CL, Hung CY, Chen WL, Chang C, Hou YH, et al. Enhancing diagnosis of T-cell lymphoma using non-recombined T-cell receptor sequences. *Front Oncol* (2022) 12:1014132. doi: 10.3389/fonc.2022.1014132

180. Drieux F, Ruminy P, Abdel-Sater A, Lemonnier F, Viailly PJ, Fataccioli V, et al. Defining signatures of peripheral T-cell lymphoma with a targeted 20-marker gene expression profiling assay. *Haematologica* (2020) 105(6):1582–92. doi: 10.3324/haematol.2019.226647

181. Amador C, Bouska A, Wright G, Weisenburger DD, Feldman AL, Greiner TC, et al. Gene expression signatures for the accurate diagnosis of peripheral T-cell lymphoma entities in the routine clinical practice. *J Clin Oncol* (2022) 40(36):4261–75. doi: 10.1200/JCO.21.02707

182. Julia E, Mareschal S, Chebel A, Golfier C, Müller TA, Lours C, et al. Chromatin accessibility profiling to increase diagnostic accuracy and refine cell-of-Origin classification of mature T-cell lymphomas. *Blood* (2021) 138(Supplement 1):809–809. doi: 10.1182/blood-2021-150034

183. Bommier C, Mauduit C, Fontaine J, Bourbon E, Sujobert P, Huet S, et al. Real-life targeted next-generation sequencing for lymphoma diagnosis over 1 year from the French lymphoma network. *Br J Haematol* (2021) 193(6):1110–22. doi: 10.1111/bjh.17395

184. Syrykh C, Gorez P, Péricart S, Grand D, Escudié F, Cabarrou B, et al. Molecular diagnosis of T-cell lymphoma: a correlative study of PCR-based T-cell clonality assessment and targeted NGS. *Blood Adv* (2021) 5(22):4590–3. doi: 10.1182/bloodadvances.2021005249

185. Dupuy A, Lemonnier F, Fataccioli V, Martin-Garcia N, Robe C, Pelletier R, et al. Multiple ways to detect IDH2 mutations in angioimmunoblastic T cell lymphoma: from immunohistochemistry to next generation sequencing. *J Mol Diagn* (2018) 20 (5):677–85. doi: 10.1016/j.jmoldx.2018.05.012

186. Broccoli A, Bertuzzi C, Fiorentino M, Morigi A, Stefoni V, Agostinelli C, et al. BRAF V600E-positive monomorphic epitheliotropic intestinal T-cell lymphoma complicating the course of hairy cell leukemia. *OncoTargets Ther* (2019) 12:4807–12. doi: 10.2147/OTT.S202061

187. Tanzima Nuhat S, Sakata-Yanagimoto M, Komori D, Hattori K, Suehara Y, Fukumoto K, et al. Droplet digital polymerase chain reaction assay and peptide nucleic acid-locked nucleic acid clamp method for RHOA mutation detection in angioimmunoblastic T-cell lymphoma. *Cancer Sci* (2018) 109(5):1682–9. doi: 10.1111/cas.13557

188. Hayashida M, Maekawa F, Chagi Y, Iioka F, Kobashi Y, Watanabe M, et al. Combination of multicolor flow cytometry for circulating lymphoma cells and tests for the *RHOA* ^{G17V} and *IDH2* ^{R172} hot-spot mutations in plasma cell-free DNA as liquid biopsy for the diagnosis of angioimmunoblastic T-cell lymphoma. *Leuk Lymphoma* (2020) 61(10):2389–98. doi: 10.1080/10428194.2020.1768382

189. Mussolin L, Bonvini P, Ait-Tahar K, Pillon M, Tridello G, Buffardi S, et al. Kinetics of humoral response to ALK and its relationship with minimal residual disease in pediatric ALCL. *Leukemia* (2009) 23(2):400–2. doi: 10.1038/leu.2008.184

190. Yee H, Ponzoni M, Merson A, Goldstein M, Scarpa A, Chilosi M, et al. Molecular characterization of the t(2;5) (p23; q35) translocation in anaplastic large cell lymphoma (Ki-1) and hodgkin's disease. *Blood* (1996) 87(3):1081–8. doi: 10.1182/blood.V87.3.1081.bloodjournal8731081

191. Ait-Tahar K, Damm-Welk C, Burkhardt B, Zimmermann M, Klapper W, Reiter A, et al. Correlation of the autoantibody response to the ALK oncoantigen in pediatric anaplastic lymphoma kinase–positive anaplastic large cell lymphoma with tumor dissemination and relapse risk. *Blood* (2010) 115(16):3314–9. doi: 10.1182/blood-2009-11-251892

192. Mussolin L, Damm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, et al. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. *Leukemia* (2013) 27(2):416–22. doi: 10.1038/leu.2012.205

193. Quelen C, Grand D, Sarot E, Brugières L, Sibon D, Pradines A, et al. Minimal residual disease monitoring using a 3′ALK universal probe assay in ALK-positive anaplastic Large-cell lymphoma. *J Mol Diagn* (2021) 23(2):131–9. doi: 10.1016/j.jmoldx.2020.11.002

194. Damm-Welk C, Kutscher N, Zimmermann M, Attarbaschi A, Schieferstein J, Knörr F, et al. Quantification of minimal disseminated disease by quantitative polymerase chain reaction and digital polymerase chain reaction for NPM-ALK as a prognostic factor in children with anaplastic large cell lymphoma. *Haematologica* (2020) 105(8):2141–9. doi: 10.3324/haematol.2019.232314

195. Cavalieri D, Tournilhac O, Missiglia E, Bonnet C, Ledoux-Pilon A, Bisig B, et al. Monomorphic epitheliotropic intestinal T-cell lymphoma (MEITL): clinicopathological analysis of a multicenter european cohort. *Hematol Oncol* (2021) 39 (Supplemental 2):83–4. doi: 10.1002/hon.44_2879

196. Sakamoto Y, Ishida T, Masaki A, Murase T, Yonekura K, Tashiro Y, et al. CCR4 mutations associated with superior outcome of adult T-cell leukemia/lymphoma under mogamulizumab treatment. *Blood* (2018) 132(7):758–61. doi: 10.1182/blood-2018-02-835991

197. Tanaka N, Mori S, Kiyotani K, Ota Y, Gotoh O, Kusumoto S, et al. Genomic determinants impacting the clinical outcome of mogamulizumab treatment for adult T-cell leukemia/lymphoma. *Haematologica* (2022) 107(10):2418–31. doi: 10.3324/haematol.2021.280352

198. Sakamoto Y, Ishida T, Masaki A, Murase T, Ohtsuka E, Takeshita M, et al. CCR7 alterations associated with inferior outcome of adult T-cell leukemia/lymphoma under mogamulizumab treatment. *Hematol Oncol* (2022) 40:876–84. doi: 10.1002/hon.3072

199. Lemonnier F, Safar V, Beldi-Ferchiou A, Cottereau AS, Bachy E, Cartron G, et al. Integrative analysis of a phase 2 trial combining lenalidomide with CHOP in angioimmunoblastic T-cell lymphoma. *Blood Adv* (2021) 5(2):539–48. doi: 10.1182/bloodadvances.2020003081

200. Shimizu-Kohno K, Satou Y, Arakawa F, Kiyasu J, Kimura Y, Niino D, et al. Detection of HTLV-1 by means of HBZ gene in situ hybridization in formalin-fixed and paraffin-embedded tissues. *Cancer Sci* (2011) 102(7):1432–6. doi: 10.1111/j.1349-7006.2011.01946.x

201. Bobée V, Drieux F, Marchand V, Sater V, Veresezan L, Picquenot JM, et al. Combining gene expression profiling and machine learning to diagnose b-cell non-Hodgkin lymphoma. *Blood Cancer J* (2020) 10(5):59. doi: 10.1038/s41408-020-0322-5

202. Drieux F, Ruminy P, Sater V, Marchand V, Fataccioli V, Lanic MD, et al. Detection of gene fusion transcripts in peripheral T-cell lymphoma using a multiplexed targeted sequencing assay. *J Mol Diagn* (2021) 23(8):929–40. doi: 10.1016/j.jmoldx.2021.04.013

203. Ehrentraut S, Nagel S, Scherr ME, Schneider B, Quentmeier H, Geffers R, et al. t (8;9)(p22;p24)/PCM1-JAK2 activates SOCS2 and SOCS3 *via* STAT5. climent m, éditeur. *PloS One* (2013) 8(1):e53767. doi: 10.1371/journal.pone.0053767

204. Panagopoulos I, Gorunova L, Spetalen S, Bassarova A, Beiske K, Micci F, et al. Fusion of the genes ataxin 2 like, ATXN2L, and janus kinase 2, JAK2, in cutaneous CD4 positive T-cell lymphoma. *Oncotarget* (2017) 8(61):103775–84. doi: 10.18632/oncotarget.21790

205. Ng SY, Yoshida N, Christie AL, Ghandi M, Dharia NV, Dempster J, et al. Targetable vulnerabilities in T- and NK-cell lymphomas identified through preclinical models. *Nat Commun* (2018) 9(1):2024. doi: 10.1038/s41467-018-04356-9

206. Rodriguez-Sevilla JJ, Salido M, Rodriguez-Rivera M, Sanchez-Gonzalez B, Gallardo F, Pujol RM, et al. PCM1::JAK2 fusion associates with an atypical form of mycosis fungoides. *Virchows Arch* (2022) 481(6):967–73. doi: 10.1007/s00428-022-03372-x

207. Fernandez-Pol S, Neishaboori N, Chapman CM, Khodadoust MS, Kim YH, Rieger KE, et al. Two cases of mycosis fungoides with PCM1-JAK2 fusion. *JCO Precis Oncol* (2021) 5):646–52. doi: 10.1200/PO.20.00366

208. Wobser M, Roth S, Appenzeller S, Kneitz H, Goebeler M, Geissinger E, et al. Oncogenic mutations and gene fusions in CD30-positive lymphoproliferations and clonally related mycosis fungoides occurring in the same patients. *JID Innov* (2021) 1 (3):100034. doi: 10.1016/j.xjidi.2021.100034

209. Zhang W, Wang W, Han X, Gan Y, Qian L, Zhang Y, et al. Circulating tumor DNA by high-throughput sequencing of T cell receptor monitored treatment response and predicted treatment failure in T cell lymphomas. *Int J Lab Hematol* (2021) 43:1041–9. doi: 10.1111/ijlh.13498

210. Sakata-Yanagimoto M, Nakamoto-Matsubara R, Komori D, Nguyen TB, Hattori K, Nanmoku T, et al. Detection of the circulating tumor DNAs in angioimmunoblastic T- cell lymphoma. *Ann Hematol* (2017) 96(9):1471–5. doi: 10.1007/s00277-017-3038-2

211. Li Q, Zhang W, Li J, Xiong J, Liu J, Chen T, et al. Plasma circulating tumor DNA assessment reveals KMT2D as a potential poor prognostic factor in extranodal NK/T-cell lymphoma. *biomark Res* (2020) 8(1):27. doi: 10.1186/s40364-020-00205-4

212. Kim JJ, Kim HY, Choi Z, Hwang Sy, Jeong H, Choi JR, et al. In-depth circulating tumor DNA sequencing for prognostication and monitoring in natural killer/T-cell lymphomas. *Front Oncol* (2023) 13:1109715. doi: 10.3389/fonc.2023.1109715

213. Huang D, Li Q, Li X, Ma N, Duan Y, Zhu L, et al. The novel prognostic index model of combining circulating tumor DNA and PINK-e predicts the clinical outcomes for newly diagnosed extranodal NK/T-cell lymphoma. *HemaSphere* (2023) 7(1):e822. doi: 10.1097/HS9.0000000000000822

214. d'Amore F, Gaulard P, Trümper L, Corradini P, Kim WS, Specht L, et al. Peripheral T-cell lymphomas: ESMO clinical practice guidelines for diagnosis, treatment and follow-up. *Ann Oncol* (2015) 26:v108–15. doi: 10.1093/annonc/mdv201

215. Mak V, Hamm J, Chhanabhai M, Shenkier T, Klasa R, Sehn LH, et al. Survival of patients with peripheral T-cell lymphoma after first relapse or progression: spectrum of disease and rare long-term survivors. *J Clin Oncol* (2013) 31(16):1970–6. doi: 10.1200/JCO.2012.44.7524

216. Bellei M, Foss FM, Shustov AR, Horwitz SM, Marcheselli L, Kim WS, et al. The outcome of peripheral T-cell lymphoma patients failing first-line therapy: a report from the prospective, international T-cell project. *Haematologica* (2018) 103(7):1191–7. doi: 10.3324/haematol.2017.186577

217. Al-Mashhadi AL, Cederleuf H, Jensen RK, Nielsen TH, Pedersen MB, Mortensenl TB, et al. Outcome of limited-stage peripheral T-cell lymphoma after CHOP(-like) therapy: a population based study of 239 patients from the Nordic lymphoma epidemiology group. *Am J Hematol* (2023) 98(3):388–97. doi: 10.1002/ajh.26803

218. Lowe EJ, Reilly AF, Lim MS, Gross TG, Saguilig L, Barkauskas DA, et al. Brentuximab vedotin in combination with chemotherapy for pediatric patients with ALK+ ALCL: results of COG trial ANHL12P1. *Blood* (2021) 137(26):3595–603. doi: 10.1182/blood.2020009806

219. Bachy E, Camus V, Thieblemont C, Sibon D, Casasnovas RO, Ysebaert L, et al. Romidepsin plus CHOP versus CHOP in patients with previously untreated peripheral T-cell lymphoma: results of the ro-CHOP phase III study (Conducted by LYSA). *J Clin Oncol* (2022) 40(3):242–51. doi: 10.1200/JCO.21.01815

220. Ruan J, Moskowitz AJ, Mehta-Shah N, Sokol L, Chen Z, Kotlov N, et al. Multicenter phase 2 study of oral azacitidine (CC-486) plus CHOP as initial treatment for peripheral T-cell lymphoma. *Blood* (2023) 141(18):2194–205. doi: 10.1182/blood.2022018254

221. Yong W, Zheng W, Zhu J, Zhang Y, Wang X, Xie Y, et al. L-asparaginase in the treatment of refractory and relapsed extranodal NK/T-cell lymphoma, nasal type. *Ann Hematol* (2009) 88(7):647–52. doi: 10.1007/s00277-008-0669-3

222. Jaccard A, Gachard N, Marin B, Rogez S, Audrain M, Suarez F, et al. Efficacy of l-asparaginase with methotrexate and dexamethasone (AspaMetDex regimen) in patients with refractory or relapsing extranodal NK/T-cell lymphoma, a phase 2 study. *Blood* (2011) 117(6):1834–9. doi: 10.1182/blood-2010-09-307454

223. Li X, Cui Y, Sun Z, Zhang L, Li L, Wang X, et al. DDGP versus SMILE in newly diagnosed advanced natural Killer/T-cell lymphoma: a randomized controlled, multicenter, open-label study in China. *Clin Cancer Res* (2016) 22(21):5223–8. doi: 10.1158/1078-0432.CCR-16-0153

224. Wang X, Zhang L, Liu X, Li X, Li L, Fu X, et al. Efficacy and safety of a pegasparaginase-based chemotherapy regimen vs an l-asparaginase-Based chemotherapy regimen for newly diagnosed advanced extranodal natural Killer/T-cell lymphoma: a randomized clinical trial. *JAMA Oncol* (2022) 8(7):1035–41. doi: 10.1001/jamaoncol.2022.1968

225. Chaubard S, Marouf A, Lavergne D, Lemonnier F, Rossignol J, Clavert A, et al. Efficacy of a short sandwich protocol, methotrexate, gemcitabine, l-asparaginase and dexamethasone chemotherapy combined with radiotherapy, in localised newly diagnosed NK/T-cell lymphoma: a French retrospective study. *Br J Haematol* (2023) 201(4):673–81. doi: 10.1111/bjh.18689

226. Jaccard A, Philippe L, Couronné L, Benoist JF, Hermine O. NK/T-cell lymphoma, the french experience. *Haematol Oncol* (2017) 35(S2):126–7. doi: 10.1002/hon.2437_116

227. Ishii T, Ishida T, Utsunomiya A, Inagaki A, Yano H, Komatsu H, et al. Defucosylated humanized anti-CCR4 monoclonal antibody KW-0761 as a novel immunotherapeutic agent for adult T-cell Leukemia/Lymphoma. *Clin Cancer Res* (2010) 16(5):1520–31. doi: 10.1158/1078-0432.CCR-09-2697

228. Shichijo T, Nosaka K, Tatetsu H, Higuchi Y, Endo S, Inoue Y, et al. Beneficial impact of first-line mogamulizumab-containing chemotherapy in adult T-cell leukaemia-lymphoma. *Br J Haematol* (2022) 198:983–7. doi: 10.1111/bjh.18281

229. Ishida T, Jo T, Takemoto S, Suzushima H, Suehiro Y, Choi I, et al. Follow-up of a randomised phase II study of chemotherapy alone or in combination with mogamulizumab in newly diagnosed aggressive adult T-cell leukaemia-lymphoma: impact on allogeneic haematopoietic stem cell transplantation. *Br J Haematol* (2019) 184:440–83. doi: 10.1111/bjh.15123

230. Gambacorti-Passerini C, Messa C, Pogliani EM. Crizotinib in anaplastic Large-cell lymphoma. *N Engl J Med* (2011) 364(8):775–6. doi: 10.1056/NEJMc1013224

231. Gambacorti Passerini C, Farina F, Stasia A, Redaelli S, Ceccon M, Mologni L, et al. Crizotinib in advanced, chemoresistant anaplastic lymphoma kinase-positive lymphoma patients. *JNCI J Natl Cancer Inst* (2014) 106(2):djt378. doi: 10.1093/jnci/djt378

232. Fukano R, Mori T, Sekimizu M, Choi I, Kada A, Saito AM, et al. Alectinib for relapsed or refractory anaplastic lymphoma kinase-positive anaplastic large cell lymphoma: an open-label phase II trial. *Cancer Sci* (2020) 111(12):4540–7. doi: 10.1111/cas.14671

233. Bossi E, Aroldi A, Brioschi FA, Steidl C, Baretta S, Renso R, et al. Phase two study of crizotinib in patients with anaplastic lymphoma kinase ALK-positive anaplastic large cell lymphoma relapsed/refractory to chemotherapy. *Am J Hematol* (2020) 95(12):E319–21. doi: 10.1002/ajh.25967

234. Rindone G, Aroldi A, Bossi E, Verga L, Zambrotta G, Tarantino S, et al. A monocentric analysis of the long-term safety and efficacy of crizotinib in relapsed/refractory ALK+ lymphomas. *Blood Adv* (2023) 7(3):314–6. doi: 10.1182/bloodadvances.2022007538

235. Lunning MA. Treatment of peripheral T-cell lymphoma: many shades of Gray. *Oncol Williston Park* (2015) 29(8):545–50.

236. Rai S, Kim WS, Ando K, Choi I, Izutsu K, Tsukamoto N, et al. Oral HDAC inhibitor tucidinostat in patients with relapsed or refractory peripheral T-cell lymphoma: phase IIb results. *Haematologica* (2022) 108(3):811–21. doi: 10.3324/haematol.2022.280996

237. Cheminant M, Bruneau J, Kosmider O, Lefrere F, Delarue R, Gaulard P, et al. Efficacy of 5-azacytidine in a TET2 mutated angioimmunoblastic T cell lymphoma. *Br J Haematol* (2015) 168(6):913–6. doi: 10.1111/bjh.13170

238. Lemonnier F, Dupuis J, Sujobert P, Tournillhac O, Cheminant M, Sarkozy C, et al. Treatment with 5-azacytidine induces a sustained response in patients with angioimmunoblastic T-cell lymphoma. *Blood* (2018) 132(21):2305–9. doi: 10.1182/blood-2018-04-840538

239. Falchi L, Ma H, Klein S, Lue JK, Montanari F, Marchi E, et al. Combined oral 5-azacytidine and romidepsin are highly effective in patients with PTCL: a multicenter phase 2 study. *Blood* (2021) 137(16):2161–70. doi: 10.1182/blood.2020009004

240. Myers R A, Wirth S, Williams S, Kiel J, Enasidenib P. An oral IDH2 inhibitor for the treatment of acute myeloid leukemia. *J Adv Pract Oncol* (2018) 9(4):435–40. doi: 10.6004/jadpro.2018.9.4.7

241. de Botton S, Montesinos P, Schuh AC, Papayannidis C, Vyas P, Wei AH, et al. Enasidenib vs conventional care in older patients with late-stage mutant- IDH2 relapsed/refractory AML: a randomized phase 3 trial. *Blood* (2023) 141(2):156–67. doi: 10.1182/blood.2021014901

242. Nguyen TB, Sakata-Yanagimoto M, Fujisawa M, Nuhat ST, Miyoshi H, Nannya Y, et al. Dasatinib is an effective treatment for angioimmunoblastic T-cell lymphoma. *Cancer Res* (2020) 80(9):1875–84. doi: 10.1158/0008-5472.CAN-19-2787

243. Lim JQ, Huang D, Tang T, Tan D, Laurensia Y, Peng RJ, et al. Whole-genome sequencing identifies responders to pembrolizumab in relapse/refractory natural-killer/T cell lymphoma. *Leukemia* (2020) 34(12):3413–9. doi: 10.1038/s41375-020-1000-0

244. Cho J, Kim SJ, Park WY, Kim J, Woo J, Kim G, et al. Immune subtyping of extranodal NK/T-cell lymphoma: a new biomarker and an immune shift during disease progression. *Mod Pathol* (2020) 33(4):603–15. doi: 10.1038/s41379-019-0392-8

245. Ratner L, Waldmann TA, Janakiram M, Brammer JE. Rapid progression of adult T-cell leukemia–lymphoma after PD-1 inhibitor therapy. *N Engl J Med* (2018) 378(20):1947–8. doi: 10.1056/NEJMc1803181

246. Hoeller S, Walz C, Reiter A, Dirnhofer S, Tzankov A. PCM1–JAK2-fusion: a potential treatment target in myelodysplastic–myeloproliferative and other hemato-lymphoid neoplasms. *Expert Opin Ther Targets* (2011) 15(1):53–62. doi: 10.1517/14728222.2011.538683

247. Andersson EI, Brück O, Braun T, Mannisto S, Saikko L, Lagström S, et al. STAT3 mutation is associated with STAT3 activation in CD30+ ALK– ALCL. *Cancers Basel.* (2020) 12(3):702. doi: 10.3390/cancers12030702

248. Chen J, Zhang Y, Petrus MN, Xiao W, Nicolae A, Raffeld M, et al. Cytokine receptor signaling is required for the survival of ALK– anaplastic large cell lymphoma, even in the presence of JAK1/STAT3 mutations. *Proc Natl Acad Sci USA* (2017) 114(15):3975–80. doi: 10.1073/pnas.1700682114

249. Moskowitz AJ, Ghione P, Jacobsen E, Ruan J, Schatz JH, Noor S, et al. A phase 2 biomarker-driven study of ruxolitinib demonstrates effectiveness of JAK/STAT targeting in T-cell lymphomas. *Blood* (2021) 138(26):2828–37. doi: 10.1182/blood.2021013379

250. Battistella M, Leboeuf C, Ram-Wolff C, Hurabielle C, Bonnafous C, Sicard H, et al. KIR3DL2 expression in cutaneous T-cell lymphomas: expanding the spectrum for KIR3DL2 targeting. *Blood* (2017) 130(26):2900–2. doi: 10.1182/blood-2017-06-792382

251. Bagot M, Porcu P, Marie-Cardine A, Battistella M, William BM, Vermeer M, et al. IPH4102, a first-in-class anti-KIR3DL2 monoclonal antibody, in patients with relapsed or refractory cutaneous T-cell lymphoma: an international, first-in-human, open-label, phase 1 trial. *Lancet Oncol* (2019) 20(8):1160–70. doi: 10.1016/S1470-2045(19)30320-1

252. Hurabielle C, Leboeuf C, Ram-Wolff C, Meignin V, Rivet J, Vignon-Pennamen MD, et al. KIR3DL2 expression in patients with adult T-cell lymphoma/leukaemia. *Br J Dermatol* (2018) 179(1):197–9. doi: 10.1111/bjd.16322

253. Cheminant M, Lhermitte L, Bruneau J, Sicard H, Bonnafous C, Touzart A, et al. KIR3DL2 contributes to the typing of acute adult T-cell leukemia and is a potential therapeutic target. *Blood* (2022) 140(13):1522–32. doi: 10.1182/blood.2022016765

254. Decroos A, Cheminant M, Bruneau J, Carras S, Parinet V, Pelletier L, et al. KIR3DL2 may represent a novel therapeutic target in aggressive systemic peripheral T-cell lymphoma. *Haematologica* (2023). doi: 10.3324/haematol.2022.282220

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 17 July 2023
DOI 10.3389/fonc.2023.1175545

OPEN ACCESS

EDITED BY
Satoshi Yoshihara,
Hyogo Medical University, Japan

REVIEWED BY
Christopher C Oakes,
The Ohio State University, United States
Shih-Sung Chuang,
Chi Mei Medical Center, Taiwan

*CORRESPONDENCE
Luís Alberto de Pádua Covas Lage
luis.lage@hc.fm.usp.br
luisalberto_lage@yahoo.com.br

RECEIVED 27 February 2023
ACCEPTED 26 June 2023
PUBLISHED 17 July 2023

CITATION
Costa RdO, Pereira J, Lage LAdPC and Baiocchi OCG (2023) Extranodal NK-/T-cell lymphoma, nasal type: what advances have been made in the last decade? *Front. Oncol.* 13:1175545. doi: 10.3389/fonc.2023.1175545

Extranodal NK-/T-cell lymphoma, nasal type: what advances have been made in the last decade?

Renata de Oliveira Costa[1,2], Juliana Pereira[2,3,4], Luís Alberto de Pádua Covas Lage[3,4]* and Otávio César Guimarães Baiocchi[2,5]

[1]Department of Hematology, Faculdade de Ciências Médicas de Santos (FCMS), Centro Universitário Lusíadas (Unilus), Santos, São Paulo, Brazil, [2]Hospital Alemao Osvaldo Cruz (HAOC), São Paulo, Brazil, [3]Department of Hematology, Hemotherapy and Cell Therapy, Faculdade de Medicina da Universidade de Sao Paulo (FM-USP), São Paulo, Brazil, [4]Laboratory of Medical Investigation in Pathogenesis and Directed Therapy in Onco-Immuno-Hematology (LIM-31), University of Sao Paulo (USP), São Paulo, Brazil, [5]Department of Hematology, Universidade Federal de Sao Paulo (Unifesp), São Paulo, Brazil

Extranodal NK-/T-cell lymphoma (ENKTCL) is a rare and highly aggressive malignancy with significant racial and geographic variations worldwide. In addition to the formerly "nasal-type" initial description, these lymphomas are predominantly extranodal in origin and typically cause vascular damage and tissue destruction, and although not fully understood, Epstein–Barr virus (EBV) has an important role in its pathogenesis. Initial assessment must include a hematopathology review of representative and viable tumor areas without necrosis for adequate immunohistochemistry studies, including EBV-encoded small RNA (EBER) *in situ* hybridization (ISH). Positron emission tomography with 18-fluorodeoxyglucose (^{18}F-FDG-PET/CT) for accurate staging is essential, and most patients will have localized disease (IE/IIE) at diagnosis. Apart from other T-cell malignancies, the best treatment even for localized cases is combined modality therapy (chemotherapy plus radiotherapy) with non-anthracycline-based regimens. For advanced-stage disease, L-asparaginase-containing regimens have shown improved survival, but relapsed and refractory cases have very poor outcomes. Nowadays, even with a better understanding of pathogenic pathways, up-front therapy is completely based on chemotherapy and radiotherapy, and treatment-related mortality is not low. Future strategies targeting signaling pathways and immunotherapy are evolving, but we need to better identify those patients with dismal outcomes in a pre-emptive way. Given the rarity of the disease, international collaborations are urgently needed, and clinical trials are the way to change the future.

KEYWORDS

extranodal NK-/T-cell lymphoma, nasal type (ENKTCL-NT), Epstein-Barr virus (EBV), angiocentricity, JAK/STAT pathway, extended-field radiotherapy (EF-RT), multi-drug resistance (MDR), prognosis

1 Introduction

Peripheral T- and NK-cell lymphomas (PTCLs) are rare, heterogeneous, and commonly aggressive non-Hodgkin's lymphomas (NHLs) that originate from post-thymic T-lymphocytes and NK cells. Altogether, these disorders comprise 10%–15% of all lymphoma subtypes with great geographic differences (1). Although it has been 50 years since the recognition that NHLs are derived from either B or T cells, T- and NK-cell lymphomas are still poorly understood malignancies due to their low incidence when compared to B-cell lymphomas. In the past, lethal midline granuloma or midline malignant reticulosis were terms used to describe the extranodal NK-/T-cell lymphoma, nasal-type (ENKTCL-NT), a lymphoma subtype that typically involves the midline facial structures. Nowadays, a much better understanding regarding clinical behavior has led to the qualifier topography "nasal-type" drop off in the present fifth edition of the World Health Organization Classification of Hematolymphoid Malignancies, as many extranodal non-nasal cases were well recognized, and dismal outcomes regarding topography need yet to be addressed (2, 3).

The ENKTCL diagnosis is based on clinical aspects, in addition to histopathologic features, expression of standard cytotoxic molecules and CD56 (neural cell adhesion molecule (NCAM)]. EBV is usually present and corroborate an accurate diagnosis. More common in Asian, Central, and South American citizens, Epstein–Barr virus (EBV) DNA and its oncogenic proteins are present in virtually all cases of ENKTCL. Fortunately, the pathogenesis of this disease has been extensively studied, and the identification of different oncogenic intracellular signaling pathways, such as Janus kinase/signal transducer and activator of transcription (JAK/STAT), programmed cell death-1/programmed death ligand 1 (PD-1/PD-L1), and epigenetic dysregulation, brought new insights to better translate these biological advances into clinical practice (4, 5).

After appropriate staging with functional images, ENKTCL must be classified into localized or advanced-stage disease. Being one of the most radiosensitive NHLs, the disease stage is the most important factor to define treatment proposals and to predict survival. Prognostic parameters, such as age, disease stage, and EBV load, may be used, but predictive models are not sufficiently accurate to guide treatment. In addition to the crucial role of radiotherapy, especially for early-stage ENKTCL, it is now known that conventional anthracycline-based chemotherapy is not sufficient. Concurrent or sequential chemoradiotherapy (CCRT or SCRT) is the standard of care for localized disease, and L-asparaginase based-chemotherapy should be used for either localized or advanced stage. In this review, we summarize current practical approaches to disease staging, available treatment options, and new insights that can guide us to better future outcomes.

2 Epidemiology and clinical features

ENKTCL is an uncommon and predominantly extranodal malignancy with great racial and geographic diversity. In the *International T-cell Lymphoma Project* (ITCLP), with the participation of 22 institutions from North America, Asia, and Europe, ENKTCL was found to be responsible for 2.7% of all cases (6). When analyzed by geographic region, the differences became evident with ENKTCL corresponding to 5.1% cases in North America, 4.3% in Europe, and impressive 22.4% cases in Asia. Another population-based registry comparing hematological malignancies from Japan and the USA demonstrated an age-adjusted incidence rate of ENKTCL of 0.04 in the USA and 0.1 per 100,000 inhabitants in Japan (7). In addition, Latin America seems to be another geographic area with a high prevalence of ENKTCL, particularly in countries such as Guatemala and Brazil (8, 9). An interesting study comparing Mexican patients with other Latin American cases demonstrated that ENKTCL was the most frequent NK-/T-cell lymphoma, representing 40% of all these cases. Interestingly, Mexico is a country geographically located in North America (10). Possibly ethnic susceptibility may partly explain this pattern, as the Mongoloid race from Asia is genetically related to natives from Central and South America (11). Of note, a familial occurrence of ENKTCL affecting father and son 2 years apart was documented. Both were farmers and used large amounts of pesticides (12). Occupation, especially organophosphate exposure, may also play a potential role in its lymphomagenesis (13).

ENKTCL has a slight male predominance with a median age of 50 years at diagnosis. The most common initial symptom is nasal obstruction and discharge, explained by the localized upper aerodigestive tract presentation in most patients (14). Locally aggressive, it often causes hard palate perforation and destroys midfacial structures, such as paranasal sinuses and nasopharynx, leading to cartilage and bone destruction, with great local deformity (Figures 1A, B). Likewise, it may extend to contiguous tissues, such as the orbit or eyelid, and although not common, cranial nerves and meninges may be affected (15, 16). Systemic disease is highly aggressive but may occur, and the most common affected sites include the skin (Figure 1C), soft tissue, testis, and gut (17, 18). Extranodal primary disease may occur even without nasal involvement, which led to the recent nomenclature change in the upcoming 5th Edition of the World Health Organization Classification of Hematolymphoid Malignancies (3). In a Brazilian retrospective cohort analysis, almost 20% of cases were extranasal in origin, and, as bone marrow involvement is rare, pancytopenia should draw attention to the diagnosis of hemophagocytic syndrome. Systemic symptoms are variable and may occur, especially in advanced disease (19). Primary extranasal disease seems to confer worse outcomes, even when compared to stage III/IV nasal disease (18).

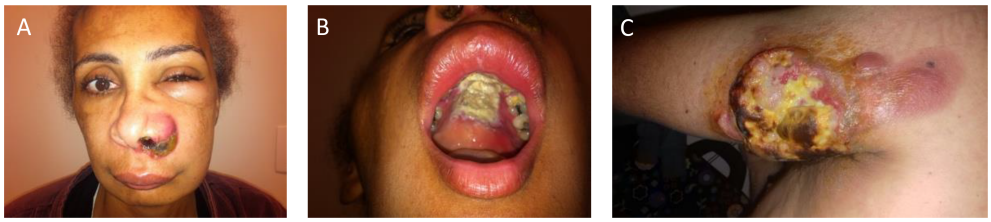


FIGURE 1
Clinical presentations of ENKTCL. **(A)** Classical nasal type with extensive and destructive lesions and eyelid swelling. **(B)** Same patient with perforation of hard palate. **(C)** Another patient with primary bulky cutaneous presentation. ENKTCL, extranodal NK-/T-cell lymphoma.

3 Diagnosis: cell of origin, pathology, and immunophenotype

Early diagnosis of ENKTCL may be challenging if extensive tumor necrosis caused by blood vessel infiltration by neoplastic cells exists (angiocentricity) and often requires multiple biopsies. Prompt clinical diagnosis is rare since most patients can be misdiagnosed and treated for more common clinical conditions, such as acute rhinosinusitis. Differential diagnoses are broad and may include cutaneous leishmaniasis, Wegener granulomatosis, South American blastomycosis, tuberculosis, leprosy, EBV-positive mucocutaneous ulcer, nasopharyngeal carcinoma, and lymphomatoid granulomatosis (20, 21). In addition, outside endemic areas and given the NK-/T-cell nature of the neoplasm, a hematopathologist is usually required for adequate diagnosis (22). ENKTCL mainly originates in activated NK cells, lacking T-cell receptor (TCR) genes, and less commonly, the cell of origin (COO) may be a cytotoxic T cell with rearranged TCR genes (23). There are little data in the literature regarding clinical, pathological, phenotypic, and molecular–genetic differences between ENKTCL cases derived from T- or NK-cell origin. Data regarding therapeutic response are virtually non-existent due to the COO. Table 1 summarizes the main differences between ENKTCL according to its COO.

Histologically, ENKTCL has a wide-ranging cytologic spectrum, with atypical and highly pleomorphic cellular infiltrate, with small, medium, or large and hyperchromatic cells. Most cases demonstrate medium-sized cells, admixed with small and large ones (Figures 2A, C, D). The presence of angioinvasion, angiodestruction, and necrosis is a hallmark of this tumor (Figures 2A, C). There may also coexist reactive inflammatory cells like lymphocytes, plasma cells, histiocytes, and eosinophils. Pseudoepitheliomatous hyperplasia, a reactive epithelial proliferation mimicking invasive squamous cell carcinoma, has been reported (24, 28). Immunophenotyping is very helpful in confirming the diagnosis, and malignant cells typically have an NK phenotype, with usually positivity for CD56. Surface CD3 negativity but cytoplasmic CD3+ (CD3ϵ) cells on paraffin samples help to support the diagnosis. There is variable expression of FAS, FASL, CD7, CD25, and CD30. Other NK- and T-cell antigens, such as CD57, CD16, CD4, and CD8, are usually negative, and a small subset of cases is truly T cell

TABLE 1 Main differences between cases of ENKTCL according to their COO.

	ENKTCL—COO: NK cell	ENKTCL—COO: T-CD8+
Frequency #	More than 75% of cases	Up to 25% of cases (αβ+, γδ+, or αβ/γδ+)
CD56 expression *	83%	33%
CXCL-13 expression *	59%	0%
PD-1 expression *	0%	40%
OCT-2 expression *	38%	0%
IRF-4/MUM-1 expression *	54%	20%
Gender predominance #	Male	Equal among both genders
Prognosis (for early-stage cases—IE/IIE) #	Poor	Tendency to better survival
Mutational landscape (SNV/CNV) ***	*STAT3*, *DDX3X*, *KMT2C*, *JAK2*, *KMT2D*, *EP300*, *STAT5B*, *STAT5A*	*EPHA1*, *TP53*, *ARID1A*, *PTPRQ*, *NCOR2*, *PPFIA2*, *BCOR*, *PTPRK*, *HDAC*
Up-regulated signaling pathways ***	JAK/STAT	RAS-MAPK and epigenetic modifiers
Potential therapeutic applications ***	JAK inhibitors	HDACi

ENKTCL, extranodal NK-/T-cell lymphoma; COO, cell of origin; NK, natural killer; OS, overall survival; SNV, single-nucleotide variant; CNV, copy number variation; HDACi, histone deacetylase inhibitors.
#According to Swerdlow SH et al. (2017) (24), Pongpruttipan et al. (2012) (23), Hong et al. (2016) (25), and Jhuang JY et al. (2015) (26).
*According to Pongpruttipan et al. (2012) (23).
***According to Xiong et al. (2020) (27).

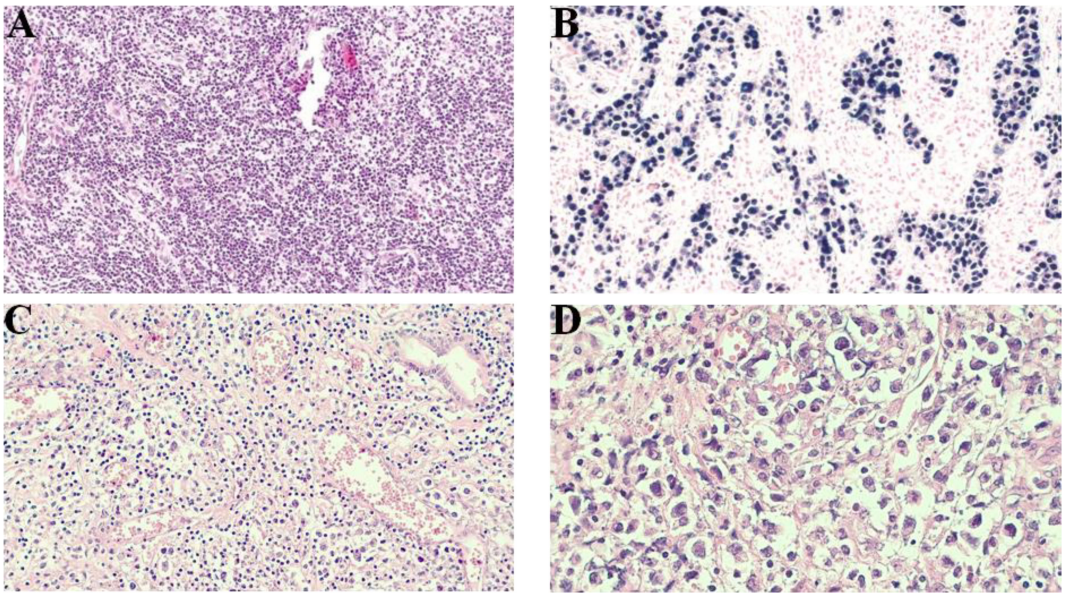


FIGURE 2
Microscopy of ENKTCL, nasal-type. **(A)** Diffuse dense tissue infiltration by small- and medium-sized atypical cells (H&E, optical microscopy, ×10 magnification). **(B)** Strong positivity for EBV-encoded small RNA by ISH for neoplastic lymphoid cells. In this staining, some large-sized atypical cells were highlighted (formalin-fixed, paraffin-embedded [FFPE] sample, optical microscopy, ×40 magnification). **(C)** Atypical lymphoid infiltration and marked vessel proliferation (optical microscopy, ×20 magnification). **(D)** High-power field demonstrating small- to medium-sized neoplastic cells and rare pleomorphic large cells infiltrating the connective tissue (optical microscopy, ×100 magnification). ENKTCL, extranodal NK-/T-cell lymphoma; EBV, Epstein–Barr virus; ISH, *in situ* hybridization.

in origin. *In situ* hybridization (ISH) for EBV-encoded small RNA (EBER) is a reliable way to demonstrate the presence of EBV (Figure 2B) (23, 24, 29). Cytotoxic granules like perforin, TIA-1, and granzyme-B are usually positive. Granulysin also seems to be a useful marker for ENKTCL, especially when lacking expression of other common markers (30, 31).

4 Pathogenesis

4.1 Recurrent genomic aberrations in ENKTCL and mutational profile

The etiopathogenesis of ENKTCL is complex and not completely elucidated. However, genetic analysis using comparative genomic hybridization (CGH), array CGH (aCGH), and loss of heterozygosity (LOH) assays showed recurrent genetic alterations in ENKTCL. The most common were gains at chromosomes 1p, 6p, 11p, 12q, 17q, 20q, and Xp and losses at chromosomes 6q, 11q, 13q, and 17p (32–37). The losses at chromosome 6q are associated with the loss of tumor-suppressor genes, such as *FOXO3*, *HACE1*, *PRDM1*, *ATG5*, and *AIM1* (38, 39). The *TP53* mutation was described in 31% to 63% of ENKTCL in two Asian population cohorts (40, 41). Therefore, ENKTCL is a malignancy associated with a high degree of chromosomal instability. Siu LL et al. reported LOH on chromosomes 6q and 13q in 80% and 66.7% of the cases, respectively, when they studied 15 patients with NK-cell lymphomas (32). Likewise, a study conducted by Chen CY et al. analyzing the pattern and distribution of recurrent karyotypic abnormalities in 200 Chinese patients with NHL demonstrated an increased frequency of 1q duplication, 6p duplication, and 11q deletion in ENKTCL in comparison to other NHL subtypes (42).

Noteworthy, the tumor mutational burden in ENKTCL is remarkably lower than in other aggressive lymphomas, similar to that found in EBV-positive nasopharyngeal carcinomas and gastric carcinoma (27, 43, 44), supporting the importance of EBV in the pathogenesis of EBV-positive neoplasms. Although recurrent somatic mutations (single-nucleotide variant (SNV)) have been reported at high frequency in ENKTCL, particularly those involving the *DDX3X* RNA helicase, *TP53*, JAK/STAT pathway genes, and epigenetic modifiers, its tumor mutational burden (TMB) is usually lower than observed in other NHLs, such as the diffuse large B-cell lymphoma (DLBCL). In this sense, a recent study accessed TMB in 188 tumor samples and 98 plasma samples from patients with different subtypes of NHLs, characterizing the landscape of somatic mutations between high-TMB (TMB-H) and low-TMB (TMB-L). The cutoff value defined to characterize TMB-H was the top quartile TMB distribution. In this study, 0% of the tumor and 0% of plasma samples from ENKTCL patients were categorized as TMB-H, compared to 34.09% (tumor) and 34.25% (plasma) of samples from DLBCL patients (45). Similarly, in another study conducted by Cho J et al. using a massively parallel sequencing panel involving 405 genes in 300 patients with different NHL subtypes, the number of SNV/indel was significantly higher ($p < 0.001$) in patients with aggressive B-cell lymphomas compared to T-lineage and natural-killer lymphomas (46). Additionally, mutations in genes involved in the JAK/STAT pathway,

epigenetic modification, RNA helicase family, RAS-MAP kinase pathway, and tumor suppressor genes contribute to ENKTCL lymphomagenesis (4, 27, 47–53). Epigenetic dysregulation involving *BCOR* and *EZH2* was also described, and its biological relevance to the ENKTCL scenario is being investigated (49, 54).

4.2 The role of EBV in ENKTCL oncogenesis

The EBV is a ubiquitous gamma human herpesvirus classified as a group 1 carcinogen by the *International Agency for Research on Cancer* (55). Mostly acquired in childhood in subclinical forms, more than 90% of adults have a lifelong asymptomatic latent disease. EBV presents tropism for different cells, including B-lymphocytes, natural killer cells, and T-lymphocytes (56, 57) being associated with several lymphoproliferative diseases (LDs) (Table 2). During primary infection, EBV usually infects epithelial cells and B-lymphocytes, but occasionally, it can infect some cells of the -T/-NK lineage. In individuals with a poor presentation by specific human leukocyte antigens (defective HLA class II molecules, such as HLA-DPB1 and HLA-DRB1) or with genetic predisposition, EBV-infected -T/-NK cells can evade host immunity and consequently survive and proliferate (63). With the participation of viral oncoproteins, such as latent membrane protein (LMP) and EBV-encoded nuclear antigens (EBNAs) and accumulation of genetic mutations, such as those affecting *DDX3X* and *TP53* genes or modifications in epigenetic targets (*KMT2C*, *KMT2D*, and *TET2*), selection and subsequent clonal expansion occur, leading to the subsequent development of ENKTCL (27, 64, 65).

The main route of EBV infection in lymphoid cells is through the CD21 receptor; however, this molecule is not expressed by -T/-NK lymphocytes. Therefore, it is believed that in -T/-NK lymphoproliferative disorders, EBV infects a lymphoid progenitor that expresses CD21, which subsequently differentiates into mature -T/-NK cells (66). Furthermore, intragenic EBV deletions are recurrently observed in chronic active EBV infection and in ENKTCL, which may represent important events for tumorigenesis (67). Additionally, certain EBV strains with a particular predisposition to infection and expansion of -T/-NK lymphocytes are more prevalent in Asia and South America, a fact that helps to explain the higher frequency of this lymphoma subtype in these geographic areas (68).

TABLE 2 EBV positivity by lymphoma subtype.

Lymphoma subtype	EBV positivity (%)	Reference
Diffuse large B-cell lymphoma	≈10-15	(3, 24, 58)
Hodgkin lymphoma	≈30–40	(3, 24, 59)
Peripheral T-cell lymphoma, NOS	≈15	(3, 24, 60)
Burkitt lymphoma	≈60	(3, 24, 61)
Extranodal NK-/T-cell lymphoma	>90	(3, 24, 62)

EBV, Epstein–Barr virus; NOS, not otherwise specified.

After infecting -T/-NK cells, viral oncoproteins, such as LMP-1, stimulate the constitutive activation of intracellular signaling pathways AKT, JAK/STAT (STAT3, JAK3, and STAT5B), MAPK, and nuclear factor kappa B (NF-κB), inhibiting apoptosis, promoting cell proliferation, and modulating the immune response, consequently regulating the interactions between the neoplastic compartment and the non-tumor immune microenvironment (69). Additionally, the LMP-1 oncoprotein promotes great genomic instability, inducing mutations and copy number alterations in several oncogenes, such as those located on the 6q21-q25 regions and in tumor suppressors, such as *TP53*, resulting in the development and progression of ENKTCL. Mutations in *DDX3X* RNA helicase gene and in *BCOR* gene, which encodes a co-repressor of the BCL-6 transcription factor, are also recurrently observed in ENKTCL, playing a fundamental role in its oncogenesis (38, 70, 71).

ENKTCL pathogenesis is strongly dependent on EBV oncoproteins, with almost all cases containing EBV genomes and encoded small RNA in neoplastic cells. Certainly, these findings provide potential targets for precision treatment (58–62). In ENKTCL lymphomagenesis, oncogenic events related to EBV are probably one of the earliest occurrences that trigger signal transduction activation, upregulation of antiapoptotic proteins like BCL2A1, and activation of various intracellular pathways. NF-κB, a transcription factor responsible for mediating the proliferation and survival of B and T cells hindering cell apoptosis and consequently promoting tumor evolution, is usually upregulated in ENKTCL (72). Alterations of the JAK/STAT pathway, especially *JAK3*, involving its mutations and aberrant phosphorylation are highly prevalent and relate to tumor cell survival (4, 73). PD-1 is an immune inhibitory receptor belonging to the CD28 family expressed by activated T and B cells, which plays an important role in tumor immune escape, with almost all EBV LDs being associated with high levels of PD-L1 expression (74). Activation of the STAT3 pathway and overexpression of LMP-1 induce the upregulation of PD-L1 in ENKTCL, highlighting the appealing treatment results with immune checkpoint inhibitors (70).

Both genetic and epigenetic factors are crucial for the pathogenesis of ENKTCL, with the latter particularly important during EBV-associated tumorigenesis (75). As stated before, EBV presents a key role as an epigenetic driver in EBV-associated cancers. EBV-encoded oncoproteins, such as LMP-1, LMP-2, and EBNA-3, modulate the host-cell epigenetic machinery, reshaping the viral and host epigenomes throughout host epigenetic modifiers, including DNA methyl transferases, histone methyl transferases, polycomb group proteins, and histone deacetylases (76–78). In addition, EBV-encoded miRNAs have epigenetic and regulatory mechanisms and regulate host-cell biology and the microenvironment, contributing to immune evasion and migration of EBV-infected cells (79, 80). In a recent study conducted by Peng RJ et al. involving whole-genome sequencing (WGS) of 27 EBV-positive NKTCL tumor samples, the authors found 0.45% (0.03%–1.06%) of similarity with the viral genome when aligned to the human and EBV reference genomes. Among the 27 viral genomes identified in these tumor samples,

approximately 1,152 SNVs and 44.8 indels (<50 bp) were revealed per sample, particularly in the BPLF-1 and BDLF-2/3 hotspot regions (81). Furthermore, different EBV strains (type A and B) and the genetic sequence of LMP-1 present distinct distributions worldwide (81). Figure 3 illustrates the main mechanisms of pathogenesis involved in ENKTCL.

4.3 Molecular pathways implicated in ENKTCL development and progression

ENKTCL molecular pathogenesis is very complex, and its main alterations involve several signaling pathways related to distinct biological functions. Among them, we found deregulation in pathways implicated in cell proliferation and survival, resistance to cell death, immune evasion, DNA repair, angiogenesis, and epigenetic control (82).

Several signaling pathways related to cell proliferation are activated in ENKTCL, often presenting gain-of-function mutations involving key genes. The JAK/STAT pathway plays a central role in ENKTCL development. Mutations involving *JAK3*, *STAT3*, and *STAT5B* genes, as well as phosphorylation of the pseudokinase domain of the *JAK3*, found in up to 40% of cases, lead to constitutive activation of the JAK/STAT pathway and consequent pro-proliferative activity (83, 84). Additionally, *PTPRK* gene, located on chromosome 6q, physiologically inactivates the STAT3 protein. However, in ENKTCL, this gene is usually inactivated by deletion or hypermethylation of its promoter region, leading to consequent activation of the JAK/STAT pathway (85).

Increased expression of genes related to the NF-κB pathway has been demonstrated in ENKTCL (86). This pathway is involved in pro-proliferative activity in several lymphoid malignancies. Furthermore, the NF-κB pathway has recently been implicated in the genesis of hemophagocytosis, a complication recurrently observed in ENKTCL (53). *DDX3X* inactivating mutations, found in up to 50% of ENKTCL, are also associated with proliferative activity via transcriptional activation of the NF-κB pathway (51). Other pathways related to proliferation and cell cycle control, such as *C-MYC*, *RUNX3*, *NOTCH1*, and *Aurora kinase*, have recently been implicated in ENKTCL oncogenesis and seem to play an important role in the proliferation and survival of its neoplastic cells (82).

Evasion of mechanisms associated with programmed cell death strongly contributes to clonal cell viability in ENKTCL. Different mechanisms are associated with apoptotic escape in this malignancy, including survivin overexpression, deregulation, and/or mutations of *TP53* tumor suppressor gene, as well as reprogramming of cellular metabolism and mechanisms associated with autophagy (82, 87, 88). Evasion of the host immune system appears to be another mechanism used by ENKTCL tumor cells to survive and subsequently proliferate. Recent studies have demonstrated overexpression of PD-L1 by immunohistochemistry in ENKTCL tumor samples. Additionally, constitutive activation of the STAT3 pathway and overexpression of the viral oncoprotein LMP1 induce upregulation of PD-L1 in ENKTCL, contributing to the tumor escape from cytotoxic T-cell activity, which has been an important biological rationale to support the development of tests with immune checkpoint inhibitors in relapsed and refractory ENKTCL (70, 89, 90).

Dysregulation of the DNA damage response is also implicated in the genesis of ENKTCL, a malignancy associated with high genomic instability. Alterations involving the *ATM/ATR* axis (*ataxia telangiectasia mutated/related*), central regulators of the

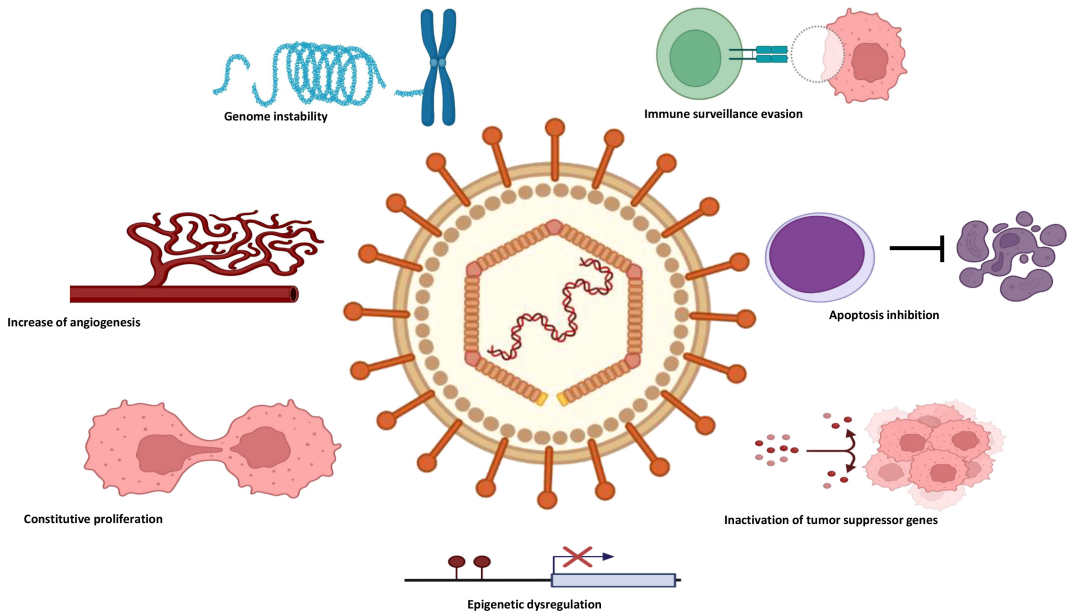


FIGURE 3
The main mechanisms of pathogenesis involved in ENKTCL. ENKTCL, extranodal NK-/T-cell lymphoma.

response to genomic damage, have been recurrently observed in a subgroup of patients with ENKTCL. *ATM/ATR* axis gene deletions, as well as mutations involving genes related to cell cycle checkpoint, are the main mechanisms implicated in the defective response to DNA damage observed in ENKTCL (91, 92).

ENKTCL is a highly vascularized tumor and is associated with a markedly angiocentric histopathological pattern. Consequently, pathways involved in neoangiogenesis seem to play a fundamental role in the survival of tumor cells and consequent progression of this neoplasm. In this sense, recent studies indicate increased expression of genes and proteins related to angiogenesis in ENKTCL samples. Among these, the *vascular endothelial growth factor-A* (*VEGF-A*), its receptor *VEGFR2*, the *hepatocyte growth factor* (*HGF*), and its receptor *MET* stand out (38, 93).

Pathways related to epigenetic modulation are also deregulated in ENKTCL. Mutations involving several epigenetic regulators have been recurrently found in ENKTCL cases, with emphasis on mutations involving the *BCOR*, *MLL2*, *ASXL3*, *ARID1A*, and *EP300* genes. These discoveries have served as a biological rationale for incorporating epigenetic-modulating drugs, such as histone deacetylase inhibitors (HDACi), into the list of new agents to be tested for ENKTCL (51, 94, 95).

Even though a better understanding of the pathogenesis of ENKTCL is evolving, it not always translates into clinical practice changes. One example is one of the earliest clinical trials using bortezomib, a proteasome inhibitor that prevents NF-κB activation. Promising results were initially demonstrated in association with the CHOP (cyclophosphamide, doxorubicin, vincristine, and prednisone) regimen, but dismal outcomes were obtained in the more modern chemotherapy association scheme era (96, 97). It is of most importance to combine molecular advances obtained in the ENKTCL recent studies with the appropriate selection of patients together with the best therapeutic combinations in clinical trials. Table 3 summarizes the main mechanisms associated with ENKTCL pathogenesis.

5 Molecular features, classification, and clinical applicability

As previously mentioned, several tumor suppressor genes located on chromosome 6q21-q25 are inactivated by the deletion of this locus, an event recurrently found in ENKTCL (32). Among these, the main ones are *FOXO3* and *PRDM1*. *FOXO3* is a *forkhead* family transcriptional factor implicated in the induction of apoptosis and cell cycle arrest in NK cells, while *PDRM1* regulates NK-cell activation and maturation (98, 99). Additionally, deletion or mutation of the tumor suppressor *TP53* has been observed in more than 30% of ENKTCL cases (100).

Genes implicated in the regulation of several signaling pathways are recurrently mutated in ENKTCL. Studies using next-generation sequencing (NGS) have shown that more than 40% of ENKTCL cases present mutations involving genes with a fundamental role in epigenetic regulation and the JAK/STAT pathway (101). Among the epigenetic modifiers, the most commonly mutated genes are *MLL2*, *MLL3*, *BCOR*, *TET2*, *EP300*, and *ARID1A*. *MLL2* and *MLL3* belong to the *KMT2* family and participate in nuclear chromatin remodeling (48). *BCOR* encodes the co-repressor for the transcription factor BCL-6 and is involved in histone modification. BCOR gene silencing promotes cell proliferation and activation of the AKT pathway (49). *TET2*, *EP300*, and *ARID1A* mutations are less frequent, occurring in approximately 7%–10% of ENKTCL cases. JAK/STAT pathway gene mutations

TABLE 3 Main mechanisms associated with ENKTCL pathogenesis.

Pathogenic mechanism	Details
Chromosomal aberrations	Gains at chromosomes 1p, 6p, 11p, 12q, 17q, 20q, and Xp Losses at chromosomes 6q, 11q, 13q, and 17p Del(6q-) are associated with losses of tumor suppressor genes *FOXO3*, *HACE1*, *PRDM1*, *ATG5*, and *AIM1* Del(17p-) is associated with loss of the tumor suppressor gene *TP53*
EBV-mediated oncogenesis	Viral oncoproteins LMP-1 and EBNAs Intragenic EBV deletions More pathogenic EBV strains—prevalent in Latin America and Asia Activation of proliferative signaling pathways: JAK/STAT, AKT, MAPK, and NF-κB; apoptosis inhibition and immune modulation via PD-1/PD-L1 axis
Deregulated molecular signaling pathways and mutational landscape	Pro-proliferative effect and cell-cycle regulation: JAK/STAT, NF-κB, *C-MYC*, *RUNX3*, NOTCH1, and Aurora kinase pathways Apoptosis inhibition: surviving overexpression, deregulation of *TP53* pathway, and reprogramming of cellular metabolism and autophagy Immune evasion: PD-1/PD-L1 axis Dysregulation of DNA repair: ATM/ATR axis Pro-angiogenic activity: *VEGFA*, *VEGFR2*, *HGF*, and *MET* Epigenetic dysregulation: *BCOR*, *MLL2*, *ASXL3*, *ARID1A*, and *EP300* Mutational landscape: *STAT3*, *JAK3*, *STAT5B*, *SOCS1*, and *PTPRK* (JAK/STAT pathway ~40%) *MAP3K5*, *BRAF*, and *EPH1A* (RAS/MAPK pathway ~15%) *ECSIT*, *IKBkB*, and *BIRC2* (NF-κB pathway ~15%) *DDX3X* (RNA helicases, ~40%–50%), *TP53* (~10%) *BCOR*, *EP300*, *TET2*, and *ARID1A* (epigenetic machinery, ~5%–10%)

ENKTCL, extranodal NK-/T-cell lymphoma; EBV, Epstein–Barr virus.

particularly affect *STAT3*, *JAK3*, *STAT5B*, *SOCS1*, and *PTPRK* genes. This pathway is crucial for the development and maturation of NK cells. Such mutations usually lead to the constitutive activation of the JAK/STAT pathway, promoting the growth, survival, and migration of tumor cells (47).

Mutations involving RAS-MAPK pathway genes are found in up to 15% of ENKTCL, commonly affecting *MAP3K5*, *BRAF*, and *EPH1A* genes, as well as NF-κB pathway activating mutations involving *ECSIT*, *IKBKB*, and *BIRC3* genes. Inactivation of RNA helicases, a negative regulator of NK-cell proliferation, usually occurs by mutations of *DDX3X* gene and less frequently by mutations involving *SHX58*, *DDX18*, and *DDX21* (51). Some mutations were associated with prognoses in ENKTCL, such as mutations in *DDX3X*, *TP53*, and *KMT2D*, which were correlated with decreased survival (50).

A recent experimental study based on phenotypic and molecular analyses of ENKTCL demonstrated tumor cell arrest at the early stages of NK maturation, suggesting that its COO is not a terminally differentiated NK cell. Additionally, ENKTCL neoplastic cells demonstrated genome-wide DNA hypermethylation, particularly in polycomb-marked regions. Such alterations were associated with extensive gene silencing, loss of transcriptional factor binding, and overexpression of *EZH2*, particularly in epigenetically more immature tumors. Based on the demonstration of this globally hypermethylated phenotype in ENKTCL, the authors investigated the potential therapeutic applications of the hypomethylating agent 5-azacytidine in a xenograft model inoculated with ENKTCL cells. The treatment led to the re-expression of NK-cell developmental genes, phenotypic NK-cell differentiation, and prolonged survival, opening precedents for the potential therapeutic application of epigenetic modifiers in this lymphoma subtype (102).

In a pioneering way, recently, Xiong et al. proposed to classify ENKTCL in three molecular subtypes based on multi-omic data, as summarized in Table 4. In this study, genomic and transcriptome analyses were performed in 128 biopsies of ENKTCL (27). The authors described the following molecular subgroup of ENKTCL: tumor suppressor–immune modulator (TSIM), MGA-BRDT (MB), and HDAC9-EP300-ARIDIA (HEA). The TSIM is characterized by deletion of chromosome 6q21, containing tumor suppressor genes, 9p24.1/*PDL1/2* overexpression, *JAK-2* amplification, 17q21.2/*STAT* amplification, JAK/STAT pathway mutation, *p53* mutation, increased expression of NK cell-associated immunity, defected immune responses associated with inappropriate antigen processing and presentation, and genomic instability. TSIM also presented higher NK gene expression, while HEA presented higher T-cell gene expression. The TSIM presented an EBV latency type II and a higher level of lytic gene *BALF3*. The MB molecular subgroup presented LOH in 1p22.1/*BRDT* and *MGA* mutations related to the upregulation of *MYC*, *MAPK*, *NOTCH*, and WNT signaling pathways, with EBV latency type I. The HEA subtype presents EBV latency patterns type II, with high levels of lytic gene *BNRF1* and is characterized by mutation of epigenetic modifiers with activation of NF-κB pathway and TCR signaling pathways. In general, the MB subtype has a worse outcome when compared to TSIM and HEA subtypes. The TSIM molecular subgroup represents the prototype of ENKTCL, typically of the NK lineage. However, the MB may correspond to ENKTCL of the T-cell lineage and represents the worst prognostic group with increased expression of *MYC*, resulting from a silencing mutation of *MGA* (27, 101). Interestingly, this molecular classification has important prognostic and therapeutic implications. While the MB subtype is associated with poor survival, the HEA group has an estimated 3-year overall survival (OS) above 90%. Likewise, individuals from the HEA group have a biological rationale for the therapeutic use of HDACi, while those with the TSIM subtype may be managed with JAK inhibitors (ruxolitinib) and/or immune checkpoint inhibitors, such as nivolumab/pembrolizumab (27, 101).

TABLE 4 Molecular subtypes of ENKTCL.

	TSIM subtype	MB subtype	HEA subtype
Main genomic alterations	Mutations in JAK/STAT and p53; amp9p24.1/JAK2 locus; amp17q21.2/STAT3/5B/5A locus. amp9P24.1/PD-L1/2 locus, del6q21(	MGM mutation and 1p22.1/BRDT LOH)	Mutations in HDAC9, EP300 and ARID1A genes
NK gene expression	+++	++	++
T-cell gene expression	++	++	+++
EBV gene expression pattern/latency	High BALF3/type II latency	Low LMP1/type I latency	High BNFR1/type II latency
Myc overexpression	–	+++	–
Signaling Pathway activated	JAK/STAT (JAK2/3, STAT3, and STAT5B)	NOTCH, MAP WNT (NOTCH3, MAP3K6, WNT2/11)	NF-κB
3-year overall survival	79.1%	38.5%	91.7%

Adapted from Xiong et al. (2020) (27).
TSIM, tumor suppressor–immune modulator; MB, MGA-BRDT; HEA: HDAC9-EP300-ARIDIA; ENKTCL, extranodal NK-/T-cell lymphoma; EBV, Epstein–Barr virus.
-, abscense of expression; +, low expression; ++, moderate expression; +++, high expression.

6 Staging and prognostic factors

The Lugano classification, derived from the Ann Arbor staging system, although routinely used for ENKTCL, lacks utility in prognostication, as these lymphomas are extranodal in origin, and this classification does not consider the adverse prognostic impact of extranasal anatomical sites (103). Different staging systems that consider local tumor invasion, disease spread pattern of local structures, lymph node, or distant sites involvement have been proposed with better accuracy, but they are not as simple as the conventionally used Ann Arbor system, which seems hard to be replaced in clinical practice (104, 105). Early-stage disease is considered nasal stage IE or contiguous stage IIE (cervical node involvement). All cases that are extranasal in origin are considered advanced-stage, with the rare exception of stage IE based on cutaneous involvement, which should be classified as a localized disease after a thorough staging for treatment purposes (2, 106). Regarding imaging modalities for staging, the high accuracy of ^{18}F-fluorodeoxyglucose–positron emission tomography/computed tomography (^{18}F-FDG-PET/CT) in ENKTCL has been demonstrated and should be included in the initial assessment (107, 108). In one of the largest cohorts in ENKTCL, ^{18}F-FDG-PET/CT detected 58 nodal and 69 extranodal lesions versus 44 and 61 detected by conventional methods, respectively ($p < 0.001$). Of note, in this study, 21.2% and 44.2% of patients had disease stage and treatment planning changed, respectively, with 97.7% sensitivity for PET/CT and 80.7% for conventional methods (109). Also, cutaneous and bone marrow infiltration by ^{18}F-FDG-PET/CT has higher sensitivity when compared to conventional methods such as bone marrow biopsy, making ^{18}F-FDG-PET/CT the modality of choice for staging (110, 111).

Additionally, prognostic information can be obtained by maximum standardized uptake value (SUV) analysis on diagnosis with worse outcomes seen in patients with higher SUV uptake (SUVmax > 15). Other adverse factors, such as bulky disease and local invasion, are independent prognostic factors for decreased progression-free survival (PFS) and OS (112). Other recent proposed refinements of ^{18}F-FDG-PET/CT are whole-body metabolic tumor volume (MTV) and whole-body level of total lesion glycolysis (LTLG). Therefore, combining tumor size with metabolic activity seems a promising prognostic tool; however, it was validated only in clinical trials and is not incorporated into clinical practice, even in much more common lymphoma subtypes (113). Noteworthy, patients from Asia are the most represented in ENKTCL studies, usually with a very short follow-up, making standardization and reproducibility a challenge, especially in middle-income countries, another geographically relevant area in this topic (114). International Prognostic Index (IPI), the most used predictive model in NHL, fails in accuracy for ENKTCL, as most patients have localized disease and good performance status and are classified as low risk by IPI; therefore, the Korean Prognostic Index (KPI) was proposed. Although KPI performed better than IPI, it was validated in most patients on anthracycline-containing regimens, and as newer treatments emerge, new prognostic factors are of paramount importance (115, 116).

Since some studies correlate pre-treatment EBV plasmatic viral load with response to treatment and overall survival, EBV serum viral load was incorporated into some prognostic scores (117). One recently proposed and useful prognostic index is the Prognostic Index for Natural Killer Lymphoma (PINK) and its variant, the Prognostic Index for Natural Killer Lymphoma plus EBV (PINK-E) scores, built from a large cohort (n = 527) of non-anthracycline-treated patients using clinical parameters (PINK) and clinical parameters plus EBV DNA data from the same cohort (n = 328 for PINK-E) (118). In this study, the authors found that age > 60 years, non-nasal type ENKTCL, distant node involvement, and advanced-stage disease adversely affect the prognosis. The patients were stratified into low risk (no risk factors), intermediate risk (one risk factor), or high risk (two or more risk factors) with 3 years OS of 81%, 62%, and 25%, respectively. The 328 patients with data for EBV DNA were stratified into three categories with different rates of overall survival. Although EBV measurements are not universally available and reference values are not standardized, PINK-E is the most reliable prognostic tool and should be adopted in clinical practice. Another promising application is the use of circulating EBV DNA as a biomarker for minimal residual disease and early relapse detection (119). In this sense, plasmatic EBV DNA measured in the interim of treatment, usually after at least two cycles of chemotherapy, has shown an important association with clinical outcomes, as well as interim imaging evaluation with ^{18}F-FDG-PET/CT. Different studies have demonstrated that ENKTCL patients with undetectable viral load and complete metabolic response (Deauville score <3) in the interim of primary therapy have markedly increased survival in comparison to patients who do not reach such goals. These same parameters are also capable of predicting an adverse prognosis if it remains positive at the end of treatment (120–122).

Aiming to overcome the limitations imposed by ENKTCL prognostic scores based on clinical and laboratory parameters, Tian XP et al. developed a molecular prognostic score based on the presence of seven single-nucleotide polymorphisms (7-SNP score). The selected seven SNPs were related to *WDR27*, *UMAD1*, *TENM2*, *LINC02463*, *KDM4C*, *FGD4*, and *FAM71A* genes. Data from 722 patients with ENKTCL from different regions of the world were analyzed and allocated into a training cohort, an internal validation cohort, and two external validation cohorts. Patients with low-risk and high-risk scores by this classifier exhibited significantly different OS and PFS ($p < 0.001$) (123). Although this score has shown high accuracy in the ENKTCL prognostic stratification, it incorporates a costly, poorly available, and complex methodology. Therefore, it has not proved to be feasible to replace PINK and PINK-E in clinical practice (124).

Recently, a Chinese multi-institutional study proposed the creation of a nomogram-revised risk index (NRI) based on the selection of risk variables obtained in a multivariate analysis of a previous cohort composed of 1,383 ENKTCL patients (125). Subsequently, the results were validated in a cohort of 1,582 cases undergoing treatment not based on anthracyclines. The variables included in the NRI were age ≥ 60 years, Eastern Cooperative Oncology Group (ECOG) score ≥2, high lactate dehydrogenase (LDH) levels, local primary tumor invasion (PTI) or stage II (1 point for each), and advanced-stage III/IV (2 points). Patients were stratified into five groups (low, low-intermediate, high-

intermediate, high, and very high) with markedly different estimated 3- and 5-year OS. The NRI showed better performance for predicting OS than the IPI, KPI, and PINK prognostic scores. Such data indicate NRI as a promising and effective tool for predicting prognosis in ENKTCL, as well as for the appropriate selection of patients for individualized therapeutic strategies (126).

7 Treatment

As in other aggressive lymphomas, ENKTCL is a type of potentially curable tumor, and all fit patients should be treated with curative intention. The most important factors regarding the choice of treatment are the stage of disease and performance status; although with novel therapies emerging, these current concepts of treatment may change in the near future. Because of the rarity of this NHL subtype, no standard treatment based on well-designed randomized trials is available. One fundamental concept is that, apart from other B-cell aggressive lymphomas, ENKTCL tumor cells have a high expression of multidrug resistance (MDR) gene *ABCB1* and its product, P-glycoprotein (Pgp), which can partly explain the poor outcomes when conventional schemes based on anthracyclines are used (127).

7.1 Limited-stage newly diagnosed disease

Historically, given the very poor outcomes with anthracycline-based chemotherapy alone (CHOP regimen) and given the radiosensitivity of ENKTCL, extended field radiotherapy (EF-RT) became a cornerstone for early-stage disease, and its omission has, since then, showed a negative impact in several studies (128, 129). Nevertheless, radiotherapy alone is not sufficient due to high rates of relapsed disease outside the radiation field (130, 131), and for limited stages, IE to IIE with nodal involvement disease combined modality therapy (radiation therapy with chemotherapy) is the standard of care. In the rare exception of stage IE primary cutaneous disease, RT alone can be considered due to anecdotal cases with more indolent clinical behavior (2, 106).

Although previous studies indicated that RT doses <50 Gy were associated with inferior response rates, more recent trials have shown that lower doses as 40–44 Gy in association with combined modalities may offer the same outcomes with good local control (132, 133). However, for clinically unfit patients unable to receive combined chemoradiotherapy (CRT), an adequate radiation dose would be >50 Gy. In combined modalities, RT doses should be given according to those established in the protocol of choice. While there is uncertainty regarding how to sequence these two modalities, a meta-analysis showed survival benefits for patients treated with RT upfront (5). Concurrent chemoradiotherapy, sequential chemoradiotherapy, and "sandwiched" chemoradiotherapy have been evaluated in prospective studies, and the choice may depend on the prompt access to RT. One commonly used RT first regimen is the RT-2/3 DeVIC (dexamethasone, etoposide, ifosfamide, and carboplatin) therapy protocol, published by the Japan Clinical Oncology Group. In this phase I/II trial, concurrent RT (50 Gy) plus carboplatin for 6–8 weeks was followed by three cycles of dexamethasone, etoposide, and ifosfamide. An updated analysis of this study revealed a PFS of 67% and an OS rate of 73% at 5 years. Although grade 3–4 hematological toxicities were observed in all patients, neutropenia was manageable, and the most common late toxicities were radiation-related, with no second malignancies reported. Of note, only patients with a performance status of 0 to 2 were included in this trial (134, 135).

The Korean group evaluated RT-cisplatin followed by VIPD (etoposide, ifosfamide, cisplatin, and dexamethasone) protocol. In this study, cisplatin as a single agent was given weekly during radiation (median 40 Gy dose), followed by three cycles of VIPD (etoposide, ifosfamide, cisplatin, and dexamethasone) after 3–5 weeks of RT. In this study, the PFS and OS rates reported were 85.1% and 86.2%, respectively. With grade 3–4 hematologic toxicities highly observed during VIPD, there were only two infection-related deaths (2/30 patients) (136). A recent observational study from South America reported inferior outcomes and toxicities with the same protocol in the real-world setting, with 57.1% (12/21) deaths during the induction phase and a 2-year OS of 53.2%. This probably reflects selection bias, as patients in clinical practice have poorer PS and may have comorbidities and infections, and these differences must be considered in clinical practice (128). Sequential and "sandwiched" chemoradiotherapy involve chemotherapy followed by interim RT. Different protocols, most using asparaginase-containing regimens, are under use with outstanding overall response rates (ORRs) >90% (137, 138). As no randomized trials comparing concurrent or sequential CRT exist, all proposals are adequate for limited-stage disease, and decisions must encompass individual and logistic issues, such as infection control, radiation availability, as previously mentioned, and even distance from the tertiary hospitals, especially in low- or middle-income countries.

7.2 Advanced-stage and relapsed/refractory disease

Approximately 25%–30% of ENKTCL patients are diagnosed with advanced-stage disease, and for those patients, although they have poor prognoses, more recent data suggest better responses for patients treated in the modern era when compared to anthracycline-based chemotherapy. In a recent large real-world retrospective analysis of 2,560 ENKTCL patients from China, 334 (13%) patients were advanced-stage, and treatment was dichotomized into non-anthracycline-based therapy (non-ANT) *vs.* ANT-based regimens, with superior PFS and OS favoring non-ANT regimens (139). Notably, an intensive treatment containing asparaginase-based chemotherapy enhances responses and is currently the standard of care as shown in a meta-analysis (140). Chemotherapy schemes vary by institution, but one of the most used is the SMILE protocol, which consists of methylprednisolone, methotrexate, ifosfamide, l-asparaginase, and etoposide, given every 28 days, for two cycles or more. In a prospective study, of 38 patients enrolled, 74% were able to complete at least two cycles.

With a median follow-up of 2 years, the ORR and complete response (CR) rate were 79% and 45%, with 55% and 53% OS and PFS, respectively (141). Although chemotherapy alone is the current practice, the role of radiation in advanced disease is not well established but may be beneficial for patients to achieve a complete response after chemotherapy. A retrospective analysis of advanced-stage disease showed a 2-year OS rate of 81.5% for post-chemotherapeutic RT patients *vs.* 40.2% for those not irradiated (142). Conversely, in another retrospective analysis of 102 advanced-stage disease patients, 23 received adjuvant radiation with no benefit in OS (p = 0.91), and of note, in this study, the best response rate was achieved with asparaginase-based therapy (SMILE), when compared to CHOP or DeVIC-like regimens (143).

Although the SMILE regimen is widely used in the treatment of advanced-stage ENKTCL, its toxicity is not negligible, particularly with regard to cytopenias and the occurrence of infectious complications. In order to minimize such adverse effects, Chinese researchers proposed the DDGP regimen (dexamethasone, cisplatin, gemcitabine, and peg-asparaginase) as an alternative to the SMILE protocol for managing ENKTCL in stages III/IV, relapsed/refractory disease, or extranasal disease. Consequently, in 2016, Xin Li et al. reported the results of a phase 3, randomized, multicenter study involving 42 Chinese patients with advanced-stage ENKTCL and ECOG ≤ 2. Patients underwent primary therapy with the DDGP (N = 21) or SMILE (N = 21) protocols. ORR and CR were increased in the DDGP arm (95% *vs.* 67% for ORR, p = 0.018; 71% *vs.* 29% for CR, p = 0.005). Similarly, 1-year PFS and 2-year OS were better in the DDGP group than in SMILE (86% *vs.* 38% for 1-year PFS, p = 0.006; 74% *vs.* 45% for 2-year OS, p = 0.027). At the same time, the group treated with SMILE developed a higher rate of adverse events, including leukopenia and allergic reactions (144). Later studies confirmed the results found in this trial, pointing to the DDGP regimen as a therapeutic strategy associated with higher response rates, increased survival, and better tolerability than SMILE chemotherapy for the treatment of patients with advanced-stage ENKTCL or with relapsed/refractory (R/R) disease (145, 146).

Since upfront autologous hematopoietic stem cell transplantation (AHSCT) results in similar response rates when compared to CCRT treatments for limited-stage disease, the current guideline from the *American Society for Blood and Marrow Transplantation* recommends against upfront AHSCT in newly diagnosed localized ENKTCL patients who achieve CR with modern therapy (147, 148). Furthermore, patients with advanced-stage disease do not seem to benefit from autologous hematopoietic cell transplantation (HCT) when treated with asparaginase-based regimens. A retrospective study demonstrated a 3-year PFS and OS of 40.1% and 52.3%, respectively, for the advanced-stage disease cohort, and patients in partial response (PR) did not benefit from the therapy, with a 3-year PFS of 13.3%. In multivariate analysis, pre-transplant PR and anthracycline-based primary chemotherapy were independent prognostic factors for reduced PFS. For OS, anthracycline-based primary chemotherapy was the only independent factor for increased risk of death (149). For patients who achieve CR and have adverse clinical variables (high prognostic score index), a retrospective analysis demonstrated notably improved survival, making AHSCT a post-induction consolidation choice for selected patients. However, more studies are needed (148).

Relapsed or refractory patients are often included in the same studies designed for advanced-stage disease, and clinical trials are the preferred treatment option after treatment with asparaginase-based chemotherapy. SMILE, AspaMetDex (asparaginase, methotrexate, and dexamethasone), P-GEMOX (peg-asparaginase, gemcitabine, and oxaliplatin), and GDP (gemcitabine, dexamethasone, and cisplatin) trials all included relapsed or refractory patients, but prognosis in this population is very poor (141, 150–152). Of note, in SMILE clinical trial, patients in the first relapse had a CR of 46%, but no refractory patient achieved CR (141). Allogenic HSCT results were reported by the *Center for International Blood and Marrow Transplant Research* (CIBMTR) study group with discouraging results. In this analysis, the 2-year PFS and OS were 20% and 24%, respectively, with a PFS of only 20% even in CR patients prior to transplant (153). A large recent analysis from the *National Spanish Group* reported the outcomes of allogenic HSCT in advanced mature T- and NK/T-cell neoplasms (6.5% ENKTCL) and showed 1-year non-relapse mortality of 21.9%, mainly due to graft-versus-host disease (GVHD) and bacterial infections (154). In addition, the current guideline from the *American Society for Blood and Marrow Transplantation* has a weak recommendation for allogeneic HSCT in this setting (147). Therefore, this group of patients represents an unmet medical need since most will not even be eligible for HSCT for inadequate functional status, organ dysfunction, comorbidities, and uncontrolled disease status, making them candidates for new agents in development.

7.3 Novel therapies

Initial impressive results involving immune therapies, especially immune checkpoint inhibitors as single-agent therapy, have been published since PD-L1 is expressed in ENKTCL (155). In one study, seven patients previously treated with asparaginase-based regimens (two with allogenic HSCT) received a median of seven cycles of the anti-PD-1 antibody pembrolizumab with an ORR of 100%, and five of them achieved CR (156). One year later, the same group reported the efficacy of another PD-1 inhibitor, nivolumab, in low doses (157). Tislelizumab in combination with chemotherapy has also proven feasible, and clinical trials with this immune checkpoint inhibitor are ongoing for both early- and advanced-stage diseases in combination with RT or chemotherapy (158, 159). Therefore, blockade of the PD-1/PD-L1 immune axis has been shown to be a safe and effective option in the management of R/R ENKTCL. However, there seems to be no direct correlation between PD-L1 antigen expression in tumor cells and therapeutic response to immune checkpoint inhibitors (160). Currently, no predictive factors of response to these drugs have been identified in ENKTCL.

In addition to initial studies demonstrating the efficacy of pembrolizumab and nivolumab in patients with ENKTCL R/R to asparaginase-based chemotherapy, new trials have evidenced the efficacy of other anti-PD-1 and anti-PD-L1 antibodies, such as

TABLE 5 Main strategies adopted for the up-front management of ENKTCL, R/R disease, and novel therapies.

ENKTCL clinical presentation	Main therapeutic strategies
1. Early stage (IE-IIE)	1.1. Very elderly (≥80 years old), unfit cases, or primary cutaneous presentation • Isolated EF-RT with 50–60 Gy 1.2. Fit patients • Chemoradiotherapy (CRT)—concurrent, sequential, or *sandwiched* modalities - 2 × P-GEMOX/EF-RT/2 × P-GEMOX - RT plus weekly cisplatin followed by 3 × VIPD (Korean protocol) - RT followed by 2/3 DeVIC
2. Advanced-stage (III-IV) or extranasal disease (except cutaneous localized disease)	- 2/3 × SMILE - 6 × DDGP - 4/6 × AspMetDex - 4 × P-GEMOX - 4/6 × GDP * Consider up-front consolidation with ASCT for cases presenting high-risk PINK or PINK-E scores
3. Relapsed/refractory (R/R) disease	- Anti-MDR-based chemotherapy (SMILE, DDGP, and P-GEMOX) followed by ASCT consolidation or allo-SCT (in cases previously submitted to ASCT)
4. Novel therapies	- Clinical trials - Anti-PD-1/PD-L1: nivolumab, pembrolizumab, tislelizumab, sintilimab, avelumab - Anti-CD30: brentuximab-vedotin - Anti-CD38: daratumumab - JAK/STAT inhibitors: ruxolitinib, tofacitinib - HDACi: chidamide - Antiviral agents: valganciclovir plus nanatinostat - CAR-T therapies

ENKTCL, extranodal NK-/T-cell lymphoma; EF-RT, extended-field radiotherapy; CRT, chemoradiotherapy; RT, radiotherapy, P-GEMOX, peg-asparaginase, gemcitabine, and oxaliplatin; VIPD, etoposide, ifosfamide, cisplatin, and dexamethasone; DeVIC, dexamethasone, etoposide, ifosfamide, and carboplatin; SMILE, methylprednisolone, methotrexate, ifosfamide, l-asparaginase, and etoposide; DDGP, dexamethasone, cisplatin, gemcitabine, and peg-asparaginase; AspMetDex, L-asparaginase, methotrexate, and dexamethasone; GDP, gemcitabine, dexamethasone, and cisplatin; ASCT, autologous stem cell transplantation; allo-SCT, allogeneic stem cell transplantation; HDACi, histone deacetylase inhibitors; CAR-T, chimeric antigen T-cell receptor.

sintilimab and avelumab, respectively (156, 157). In the ORIENT-4 trial, 28 patients with R/R ENKTCL received sintilimab at a dose of 200 mg I.V. every 3 weeks for 24 months. With a median follow-up of 30.4 months, the median OS was not reached, and the estimated 2-year OS was 78.6%. Serious adverse events occurred in only 25% of cases, and no patient died from toxicity. Thus, sintilimab proved to be a safe and effective therapeutic strategy for the management of R/R ENKTCL (161). Similarly, a recent phase 2 study evaluated the efficacy and safety of the anti-PD-L1 antibody avelumab in 21 cases of R/R ENKTCL. In this study, the responses were lower, with an ORR of 38% and a CR of 24%. No grade 4 adverse effects occurred, but there was a correlation between response and tissue expression of the PD-L1, with all patients who achieved CR presenting high PD-L1 expression (162). Although it had moderate activity as a single agent, avelumab seems to be an interesting option for trials testing it in association with other drugs, with particular potential benefit in cases of R/R ENKTCL with a high density of the PD-L1 antigen.

A proportion of ENKTCL express the transmembrane glycoprotein receptor CD30 (Ki-1) and CD38, and the clinical activity of antibodies directed against both antigens has been evaluated as possible target therapies alone or in combination. A phase 2 study with daratumumab monotherapy, a monoclonal antibody targeting CD38, demonstrated a 25% ORR in relapsed/refractory ENKTCL patients. Although feasible, no patient achieved CR, and patients presented a short duration of response (163).

Although there is a biological rationale for the use of JAK/STAT inhibitors in ENKTCL, few studies have assessed the real impact of these agents on this neoplasm. *In vitro* studies have shown that the use of the pan-JAK inhibitor CP690550 and the JAK2 inhibitor AG490 resulted in decreased phosphorylation of STAT3 and STAT5 and subsequent stimulation of apoptosis in ENKTCL cell lines (47, 164). Tofacitinib, a pan-JAK inhibitor with higher JAK-3 selectivity, has demonstrated activity, inducing cell cycle arrest and growth inhibition in both positive and negative EBV NK cell lines and may be an attractive therapy (165). Currently, an ongoing phase 2 study (NCT03598959) is evaluating the safety and efficacy of the combination composed of the pan-JAK inhibitor tofacitinib with chidamide in R/R ENKTCL.

HDACi also have a potential therapeutic effect on ENKTCL, particularly on the HEA molecular subgroup, which is enriched in mutations involving epigenetic regulators. Supported by this biological principle, monotherapy with chidamide, a selective HDAC 1, 2, and 3 inhibitor, has been tested in a phase 2 trial involving 15 patients with R/R ENKTCL. In monotherapy, its activity was modest, with CR achieved in only 33% of cases and with a short median duration of response (166). However, this agent is currently being tested in combination with other drugs.

The oncogenic EBV protein overexpression may be a target for adoptive immunotherapy with antigen-specific cytotoxic cells, and this strategy was explored as a post-remission therapy with promising results (167). In addition, the association of nanatinostat with valganciclovir showed promising results in a phase 1/2 trial, and ENKTCL patients refractory to their last therapy presented an ORR of 60%, including 27% of CR (168). Although chimeric antigen T-cell receptor (CAR-T) therapy has

been proven to be an interesting form of immunotherapy in several B-cell lymphoid malignancies, such as R/R diffuse large B-cell lymphoma, follicular lymphoma, B-cell acute lymphoblastic leukemia, and multiple myeloma, its use has been extremely limited in the setting of -T/-NK lymphoid disorders. While encouraging results were seen with some novel agents, there are still very little data about CAR-T in PTCL; limited experience and lack of controls preclude critical analyses. One of the main challenges in the use of CAR-T therapy in -T/-NK cell malignancies is due to the fact that neoplastic cells share a series of common antigens with normal T-lymphocytes, which can lead to fratricide and serious T-cell lymphoid aplasia in the receptor. To mitigate this effect, a selection of appropriate antigenic targets is essential. Currently, specific antigens have been selected for the construction of chimeric products, among which CD30, CD37, TRBC1, CCR4, and CCR9 stand out. The use of nanobody-derived or naturally selected CAR-T is an attractive strategy to overcome fratricide. Another problem intrinsic to the use of this therapeutic modality in T-cell malignancies refers to the potential contamination of the product collected for the construction of CAR-T with clonal T-cells; however, the use of allogeneic CAR-T products or CAR-NK-cells is a possible strategy with the ability to mitigate this contamination (169). Currently, data about the use of CAR-T therapy in ENKTCL are very scarce, although it may constitute an interesting therapeutic option for R/R disease in the near future. A trial with anti-CD30 CAR-T is underway for the treatment of R/R CD30-positive PTCL (NCT03049499) and its results are being eagerly awaited by the scientific community. Table 5 summarizes the main therapeutic options for ENKTCL patients in different clinical settings.

8 Conclusion and future directions

Successfully, the last decade brought us a better understanding of the importance of combined modality therapy and the relevance of asparaginase-containing protocols for ENKTCL. Nevertheless, a proportion of patients will relapse, and their prognosis is dismal. Advances in pathogenic mechanisms brought us a window of opportunity, but we need a great international, multicenter effort to design approaches capable of modifying the future.

Author contributions

RC, JP, LL, and OB reviewed the literature and organized and wrote the article. All authors contributed to the article and approved the submitted version.

Acknowledgments

We thank Dr. Sheila Aparecida Coelho Siqueira for providing photographs of the histopathological analysis (H&E and immunohistochemical stains)—Department of Pathology, University of São Paulo (USP). We thank Fabio Alessandro de Freitas and Samuel Campanelli Couto Freitas for the layout of the article and English revision, respectively.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. William BM, Armitage JO. International analysis of the frequency and outcomes of Nk/T-cell lymphomas. *Best Pract Res Clin haematology* (2013) 26(1):23–32. doi: 10.1016/j.beha.2013.04.003

2. Isobe Y, Aritaka N, Sasaki M, Oshimi K, Sugimoto K. Spontaneous regression of natural killer cell lymphoma. *J Clin Pathol* (2009) 62(7):647–50. doi: 10.1136/jcp.2008.062976

3. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

4. Lee S, Park HY, Kang SY, Kim SJ, Hwang J, Lee S, et al. Genetic alterations of Jak/Stat cascade and histone modification in extranodal Nk/T-cell lymphoma nasal type. *Oncotarget* (2015) 6(19):17764–76. doi: 10.18632/oncotarget.3776

5. Hu S, Zhou D, Zhang W. The optimal timing of radiotherapy in the combined modality therapy for limited-stage extranodal Nk/T cell lymphoma (Enktl): a systematic review and meta-analysis. *Ann Hematol* (2018) 97(12):2279–87. doi: 10.1007/s00277-018-3479-2

6. Vose J, Armitage J, Weisenburger D. International peripheral T-cell and natural Killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26(25):4124–30. doi: 10.1200/jco.2008.16.4558

7. Chihara D, Ito H, Matsuda T, Shibata A, Katsumi A, Nakamura S, et al. Differences in incidence and trends of haematological malignancies in Japan and the united states. *Br J haematology* (2014) 164(4):536–45. doi: 10.1111/bjh.12659

8. Gualco G, Domeny-Duarte P, Chioato L, Barber G, Natkunam Y, Bacchi CE. Clinicopathologic and molecular features of 122 Brazilian cases of nodal and extranodal Nk/T-cell lymphoma, nasal type, with ebv subtyping analysis. *Am J Surg Pathol* (2011) 35(8):1195–203. doi: 10.1097/PAS.0b013e31821ec4b5

9. Perry AM, Molina-Kirsch H, Nathwani BN, Diebold J, Maclennan KA, Müller-Hermelink HK, et al. Classification of non-Hodgkin lymphomas in Guatemala according to the world health organization system. *Leukemia lymphoma* (2011) 52 (9):1681–8. doi: 10.3109/10428194.2011.584254

10. Avilés A. Nasal Nk/T-cell lymphoma. a comparative analysis of a Mexican population with the other populations of Latin-America. *Mediterr J Hematol Infect Dis* (2015) 7(1):e2015052. doi: 10.4084/mjhid.2015.052

11. Tokunaga K, Ohashi J, Bannai M, Juji T. Genetic link between asians and native americans: evidence from hla genes and haplotypes. *Hum Immunol* (2001) 62(9):1001–8. doi: 10.1016/s0198-8859(01)00301-9

12. Kojya S, Matsumura J, Ting L, Hongyo T, Inazawa J, Kirihata M, et al. Familial nasal Nk/T-cell lymphoma and pesticide use. *Am J Hematol* (2001) 66(2):145–7. doi: 10.1002/1096-8652(200102)66:2<145::Aid-ajh1033>3.0.Co;2-v

13. Hu L, Luo D, Zhou T, Tao Y, Feng J, Mei S. The association between non-Hodgkin lymphoma and organophosphate pesticides exposure: a meta-analysis. *Environ pollut (Barking Essex 1987)* (2017) 231(Pt 1):319–28. doi: 10.1016/j.envpol.2017.08.028

14. Allen PB, Lechowicz MJ. Management of Nk/T-cell lymphoma, nasal type. *J Oncol Pract* (2019) 15(10):513–20. doi: 10.1200/jop.18.00719

15. Liang R, Todd D, Chan TK, Chiu E, Lie A, Kwong YL, et al. Treatment outcome and prognostic factors for primary nasal lymphoma. *J Clin Oncol* (1995) 13(3):666–70. doi: 10.1200/jco.1995.13.3.666

16. Akbar M, Clasen-Linde E, Specht L. Extranodal Nk/T-cell lymphoma, nasal type, with extranasal presentation - a case report and a review of the literature. *Acta Oncol (Stockholm Sweden)* (2020) 59(12):1480–7. doi: 10.1080/0284186x.2020.1795250

17. Chan JK, Sin VC, Wong KF, Ng CS, Tsang WY, Chan CH, et al. Nonnasal lymphoma expressing the natural killer cell marker Cd56: a clinicopathologic study of 49 cases of an uncommon aggressive neoplasm. *Blood* (1997) 89(12):4501–13.

18. Au WY, Weisenburger DD, Intragumtornchai T, Nakamura S, Kim WS, Sng I, et al. Clinical differences between nasal and extranasal natural Killer/T-cell lymphoma: a study of 136 cases from the international peripheral T-cell lymphoma project. *Blood* (2009) 113(17):3931–7. doi: 10.1182/blood-2008-10-185256

19. Huang Y, Jia B, Jiang S, Zhou S, Yang J, Liu P, et al. Different clinical characteristics and treatment strategies for patients with localized sinonasal diffuse Large b cell lymphoma and extranodal Nk/T cell lymphoma. *J Hematol Oncol* (2017) 10 (1):7. doi: 10.1186/s13045-016-0368-9

20. Miyake MM, Oliveira MV, Miyake MM, Garcia JO, Granato L. Clinical and otorhinolaryngological aspects of extranodal Nk/T cell lymphoma, nasal type. *Braz J otorhinolaryngology* (2014) 80(4):325–9. doi: 10.1016/j.bjorl.2014.05.013

21. Gurel MS, Tekin B, Uzun S. Cutaneous leishmaniasis: a great imitator. *Clinics Dermatol* (2020) 38(2):140–51. doi: 10.1016/j.clindermatol.2019.10.008

22. Jaffe ES, Chan JK, Su IJ, Frizzera G, Mori S, Feller AC, et al. Report of the workshop on nasal and related extranodal angiocentric T/Natural killer cell lymphomas. definitions, differential diagnosis, and epidemiology. *Am J Surg Pathol* (1996) 20(1):103–11. doi: 10.1097/00000478-199601000-00012

23. Pongpruttipan T, Sukpanichnant S, Assanasen T, Wannakrairot P, Boonsakan P, Kanoksil W, et al. Extranodal Nk/T-cell lymphoma, nasal type, includes cases of natural killer cell and Aβ, Γδ, and Aβ/Γδ T-cell origin: a comprehensive clinicopathologic and phenotypic study. *Am J Surg Pathol* (2012) 36(4):481–99. doi: 10.1097/PAS.0b013e31824433d8

24. Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, et al. *Who classification of tumours of haematopoietic and lymphoid tissues*, *Revised 4th ed*. (Lyon, France: IARC Press) (2017).

25. Hong M, Lee T, Young Kang S, Kim SJ, Kim W, Ko YH. Nasal-type Nk/T-cell lymphomas are more frequently T rather than nk lineage based on T-cell receptor gene, rna, and protein studies: lineage does not predict clinical behavior. *Modern Pathol* (2016) 29(5):430–43. doi: 10.1038/modpathol.2016.47

26. Jhuang JY, Chang ST, Weng SF, Pan ST, Chu PY, Hsieh PP, et al. Extranodal natural Killer/T-cell lymphoma, nasal type in Taiwan: a relatively higher frequency of T-cell lineage and poor survival for extranasal tumors. *Hum Pathol* (2015) 46(2):313–21. doi: 10.1016/j.humpath.2014.11.008

27. Xiong J, Cui BW, Wang N, Dai YT, Zhang H, Wang CF, et al. Genomic and transcriptomic characterization of natural killer T cell lymphoma. *Cancer Cell* (2020) 37(3):403–19.e6. doi: 10.1016/j.ccell.2020.02.005

28. Ling YH, Zhu CM, Wen SH, Luo RZ, Li P, Cao Y, et al. Pseudoepitheliomatous hyperplasia mimicking invasive squamous cell carcinoma in extranodal natural Killer/T-cell lymphoma: a report of 34 cases. *Histopathology* (2015) 67(3):404–9. doi: 10.1111/his.12656

29. Jaffe ES, Nicolae A, Pittaluga S. Peripheral T-cell and nk-cell lymphomas in the who classification: pearls and pitfalls. *Modern Pathol* (2013) 26 Suppl 1(Suppl 1):S71–87. doi: 10.1038/modpathol.2012.181

30. Mori N, Yatabe Y, Oka K, Kinoshita T, Kobayashi T, Ono T, et al. Expression of perforin in nasal lymphoma. additional evidence of its natural killer cell derivation. *Am J Pathol* (1996) 149(2):699–705.

31. Lo Bello G, Akarca AU, Ambrosio MR, Agostinelli C, Molina-Kirsch H, Ramsay A, et al. Granulysin, a novel marker for extranodal Nk/T cell lymphoma, nasal type. *Virchows Archiv* (2018) 473(6):749–57. doi: 10.1007/s00428-018-2434-x

32. Siu LL, Chan V, Chan JK, Wong KF, Liang R, Kwong YL. Consistent patterns of allelic loss in natural killer cell lymphoma. *Am J Pathol* (2000) 157(6):1803–9. doi: 10.1016/s0002-9440(10)64818-3

33. Yoon J, Ko YH. Deletion mapping of the long arm of chromosome 6 in peripheral T and nk cell lymphomas. *Leukemia lymphoma* (2003) 44(12):2077–82. doi: 10.1080/1042819031000123410

34. Siu LL, Wong KF, Chan JK, Kwong YL. Comparative genomic hybridization analysis of natural killer cell Lymphoma/Leukemia. recognition of consistent patterns of genetic alterations. *Am J Pathol* (1999) 155(5):1419–25. doi: 10.1016/s0002-9440(10)65454-5

35. Sun HS, Su IJ, Lin YC, Chen JS, Fang SY. A 2.6 Mb interval on chromosome 6q25.2-Q25.3 is commonly deleted in human nasal natural Killer/T-cell lymphoma. *Br J haematology* (2003) 122(4):590–9. doi: 10.1046/j.1365-2141.2003.04419.x

36. Ko YH, Choi KE, Han JH, Kim JM, Ree HJ. Comparative genomic hybridization study of nasal-type Nk/T-cell lymphoma. *Cytometry* (2001) 46(2):85–91. doi: 10.1002/cyto.1069

37. Nakashima Y, Tagawa H, Suzuki R, Karnan S, Karube K, Ohshima K, et al. Genome-wide array-based comparative genomic hybridization of natural killer cell Lymphoma/Leukemia: different genomic alteration patterns of aggressive nk-cell leukemia and extranodal Nk/T-cell lymphoma, nasal type. *Genes Chromosomes Cancer* (2005) 44(3):247–55. doi: 10.1002/gcc.20245

38. Huang Y, de Reyniès A, de Leval L, Ghazi B, Martin-Garcia N, Travert M, et al. Gene expression profiling identifies emerging oncogenic pathways operating in extranodal Nk/T-cell lymphoma, nasal type. *Blood* (2010) 115(6):1226–37. doi: 10.1182/blood-2009-05-221275

39. Iqbal J, Kucuk C, Deleeuw RJ, Srivastava G, Tam W, Geng H, et al. Genomic analyses reveal global functional alterations that promote tumor growth and novel tumor suppressor genes in natural killer-cell malignancies. *Leukemia* (2009) 23 (6):1139–51. doi: 10.1038/leu.2009.3

40. Kurniawan AN, Hongyo T, Hardjolukito ES, Ham MF, Takakuwa T, Kodariah R, et al. Gene mutation analysis of sinonasal lymphomas in Indonesia. *Oncol Rep* (2006) 15(5):1257–63.

41. Hongyo T, Hoshida Y, Nakatsuka S, Syaifudin M, Kojya S, Yang WI, et al. P53, K-ras, c-kit and beta-catenin gene mutations in sinonasal Nk/T-cell lymphoma in Korea and Japan. *Oncol Rep* (2005) 13(2):265–71.

42. Chen CY, Yao M, Tang JL, Tsay W, Wang CC, Chou WC, et al. Chromosomal abnormalities of 200 Chinese patients with non-hodgkin's lymphoma in Taiwan: with special reference to T-cell lymphoma. *Ann Oncol* (2004) 15(7):1091–6. doi: 10.1093/annonc/mdh263

43. Chalmers ZR, Connelly CF, Fabrizio D, Gay L, Ali SM, Ennis R, et al. Analysis of 100,000 human cancer genomes reveals the landscape of tumor mutational burden. *Genome Med* (2017) 9(1):34. doi: 10.1186/s13073-017-0424-2

44. Wang K, Kan J, Yuen ST, Shi ST, Chu KM, Law S, et al. Exome sequencing identifies frequent mutation of Arid1a in molecular subtypes of gastric cancer. *Nat Genet* (2011) 43(12):1219–23. doi: 10.1038/ng.982

45. Zhou H, Du X, Zhao T, Ouyang Z, Liu W, Deng M, et al. Distribution and influencing factors of tumor mutational burden in different lymphoma subtypes. *J Clin Oncol* (2019) 37(15_suppl):e19053–e. doi: 10.1200/JCO.2019.37.15_suppl.e19053

46. Cho J, Yoon SE, Kim SJ, Ko YH, Kim WS. Comparison of tumor mutation burden of 300 various non-Hodgkin lymphomas using panel based massively parallel sequencing. *BMC Cancer* (2021) 21(1):972. doi: 10.1186/s12885-021-08695-7

47. Koo GC, Tan SY, Tang T, Poon SL, Allen GE, Tan L, et al. Janus kinase 3-activating mutations identified in natural Killer/T-cell lymphoma. *Cancer Discovery* (2012) 2(7):591–7. doi: 10.1158/2159-8290.Cd-12-0028

48. Choi S, Go JH, Kim EK, Lee H, Lee WM, Cho CS, et al. Mutational analysis of extranodal Nk/T-cell lymphoma using targeted sequencing with a comprehensive cancer panel. *Genomics Inf* (2016) 14(3):78–84. doi: 10.5808/gi.2016.14.3.78

49. Dobashi A, Tsuyama N, Asaka R, Togashi Y, Ueda K, Sakata S, et al. Frequent bcor aberrations in extranodal Nk/T-cell lymphoma, nasal type. *Genes Chromosomes Cancer* (2016) 55(5):460–71. doi: 10.1002/gcc.22348

50. Gao LM, Zhao S, Zhang WY, Wang M, Li HF, Lizaso A, et al. Somatic mutations in Kmt2d and Tet2 associated with worse prognosis in Epstein-Barr virus-associated T or natural killer-cell lymphoproliferative disorders. *Cancer Biol Ther* (2019) 20 (10):1319–27. doi: 10.1080/15384047.2019.1638670

51. Jiang L, Gu ZH, Yan ZX, Zhao X, Xie YY, Zhang ZG, et al. Exome sequencing identifies somatic mutations of Ddx3x in natural Killer/T-cell lymphoma. *Nat Genet* (2015) 47(9):1061–6. doi: 10.1038/ng.3358

52. Küçük C, Jiang B, Hu X, Zhang W, Chan JK, Xiao W, et al. Activating mutations of Stat5b and Stat3 in lymphomas derived from Γδ-T or nk cells. *Nat Commun* (2015) 6:6025. doi: 10.1038/ncomms7025

53. Wen H, Ma H, Cai Q, Lin S, Lei X, He B, et al. Recurrent ecsit mutation encoding V140a triggers hyperinflammation and promotes hemophagocytic syndrome in extranodal Nk/T cell lymphoma. *Nat Med* (2018) 24(2):154–64. doi: 10.1038/nm.4456

54. Yan J, Li B, Lin B, Lee PT, Chung TH, Tan J, et al. Ezh2 phosphorylation by Jak3 mediates a switch to noncanonical function in natural Killer/T-cell lymphoma. *Blood* (2016) 128(7):948–58. doi: 10.1182/blood-2016-01-690701

55. Epstein-Barr Virus and kaposi's sarcoma Herpesvirus/Human herpesvirus 8. *IARC Monogr Eval carcinogenic risks to humans* (1997) 70:1–492.

56. Jondal M, Klein G. Surface markers on human b and T lymphocytes. ii. presence of Epstein-Barr virus receptors on b lymphocytes. *J Exp Med* (1973) 138(6):1365–78. doi: 10.1084/jem.138.6.1365

57. Barros MHM, Vera-Lozada G, Segges P, Hassan R, Niedobitek G. Revisiting the tissue microenvironment of infectious mononucleosis: identification of ebv infection in T cells and deep characterization of immune profiles. *Front Immunol* (2019) 10:146. doi: 10.3389/fimmu.2019.00146

58. Lu TX, Liang JH, Miao Y, Fan L, Wang L, Qu XY, et al. Epstein-Barr Virus positive diffuse Large b-cell lymphoma predict poor outcome, regardless of the age. *Sci Rep* (2015) 5:12168. doi: 10.1038/srep12168

59. Murray PG, Young LS. An etiological role for the Epstein-Barr virus in the pathogenesis of classical Hodgkin lymphoma. *Blood* (2019) 134(7):591–6. doi: 10.1182/blood.2019000568

60. Dupuis J, Emile JF, Mounier N, Gisselbrecht C, Martin-Garcia N, Petrella T, et al. Prognostic significance of Epstein-Barr virus in nodal peripheral T-cell lymphoma, unspecified: a groupe d'etude des lymphomes de l'adulte (Gela) study. *Blood* (2006) 108(13):4163–9. doi: 10.1182/blood-2006-04-017632

61. Pannone G, Zamparese R, Pace M, Pedicillo MC, Cagiano S, Somma P, et al. The role of ebv in the pathogenesis of burkitt's lymphoma: an Italian hospital based survey. *Infect Agents Cancer* (2014) 9(1):34. doi: 10.1186/1750-9378-9-34

62. Harabuchi Y, Takahara M, Kishibe K, Moriai S, Nagato T, Ishii H. Nasal natural killer (Nk)/T-cell lymphoma: clinical, histological, virological, and genetic features. *Int J Clin Oncol* (2009) 14(3):181–90. doi: 10.1007/s10147-009-0882-7

63. Kimura H, de Leval L, Cai Q, Kim WS. Ebv-associated nk and T-cell lymphoid neoplasms. *Curr Opin Oncol* (2022) 34(5):422–31. doi: 10.1097/cco.0000000000000889

64. Gao Y, Li Y, Ma G, Zhao G, Liu H. Kmt2d and Tp53 mutation status improve the prognostic value of the international prognostic index (Ipi) stratification in enktl patients. *Neoplasma* (2020) 67(3):636–43. doi: 10.4149/neo_2020_190307N194

65. Qi F, Cao Z, Chen B, Chai Y, Lin J, Ye J, et al. Liquid biopsy in extranodal Nk/T-cell lymphoma: a prospective analysis of cell-free DNA genotyping and monitoring. *Blood Adv* (2021) 5(11):2505–14. doi: 10.1182/bloodadvances.2020001637

66. Okuno Y, Murata T, Sato Y, Muramatsu H, Ito Y, Watanabe T, et al. Defective Epstein-Barr virus in chronic active infection and haematological malignancy. *Nat Microbiol* (2019) 4(3):404–13. doi: 10.1038/s41564-018-0334-0

67. Kimura H, Okuno Y, Sato Y, Watanabe T, Murata T. Deletion of viral micrornas in the oncogenesis of Epstein-Barr virus-associated lymphoma. *Front Microbiol* (2021) 12:667968. doi: 10.3389/fmicb.2021.667968

68. Kanda T, Yajima M, Ikuta K. Epstein-Barr Virus strain variation and cancer. *Cancer Sci* (2019) 110(4):1132–9. doi: 10.1111/cas.13954

69. Kimura H. Ebv in T-/Nk-Cell tumorigenesis. *Adv Exp Med Biol* (2018) 1045:459–75. doi: 10.1007/978-981-10-7230-7_21

70. Song TL, Nairismägi ML, Laurensia Y, Lim JQ, Tan J, Li ZM, et al. Oncogenic activation of the Stat3 pathway drives pd-L1 expression in natural Killer/T-cell lymphoma. *Blood* (2018) 132(11):1146–58. doi: 10.1182/blood-2018-01-829424

71. Montes-Mojarro IA, Chen BJ, Ramirez-Ibarguen AF, Quezada-Fiallos CM, Pérez-Báez WB, Dueñas D, et al. Mutational profile and ebv strains of extranodal Nk/T-cell lymphoma, nasal type in Latin America. *Modern Pathol* (2020) 33(5):781–91. doi: 10.1038/s41379-019-0415-5

72. Smatti MK, Al-Sadeq DW, Ali NH, Pintus G, Abou-Saleh H, Nasrallah GK. Epstein-Barr Virus epidemiology, serology, and genetic variability of lmp-1 oncogene among healthy population: an update. *Front Oncol* (2018) 8:211. doi: 10.3389/fonc.2018.00211

73. Guo Y, Arakawa F, Miyoshi H, Niino D, Kawano R, Ohshima K. Activated janus kinase 3 expression not by activating mutations identified in natural Killer/T-cell lymphoma. *Pathol Int* (2014) 64(6):263–6. doi: 10.1111/pin.12166

74. Hu B, Oki Y. Novel immunotherapy options for extranodal Nk/T-cell lymphoma. *Front Oncol* (2018) 8:139. doi: 10.3389/fonc.2018.00139

75. Li L, Ma BBY, Chan ATC, Chan FKL, Murray P, Tao Q. Epstein-Barr Virus-induced epigenetic pathogenesis of viral-associated lymphoepithelioma-like carcinomas and natural Killer/T-cell lymphomas. *Pathog (Basel Switzerland)* (2018) 7(3):1–11. doi: 10.3390/pathogens7030063

76. Hino R, Uozaki H, Murakami N, Ushiku T, Shinozaki A, Ishikawa S, et al. Activation of DNA methyltransferase 1 by ebv latent membrane protein 2a leads to promoter hypermethylation of pten gene in gastric carcinoma. *Cancer Res* (2009) 69 (7):2766–74. doi: 10.1158/0008-5472.Can-08-3070

77. Dutton A, Woodman CB, Chukwuma MB, Last JI, Wei W, Vockerodt M, et al. Bmi-1 is induced by the Epstein-Barr virus oncogene Lmp1 and regulates the expression of viral target genes in Hodgkin lymphoma cells. *Blood* (2007) 109 (6):2597–603. doi: 10.1182/blood-2006-05-020545

78. Paschos K, Parker GA, Watanatanasup E, White RE, Allday MJ. Bim promoter directly targeted by Ebna3c in polycomb-mediated repression by ebv. *Nucleic Acids Res* (2012) 40(15):7233–46. doi: 10.1093/nar/gks391

79. Albanese M, Tagawa T, Bouvet M, Maliqi L, Lutter D, Hoser J, et al. Epstein-Barr Virus micrornas reduce immune surveillance by virus-specific Cd8+ T cells. *Proc Natl Acad Sci United States America* (2016) 113(42):E6467–e75. doi: 10.1073/pnas.1605884113

80. Cullen BR. Micrornas as mediators of viral evasion of the immune system. *Nat Immunol* (2013) 14(3):205–10. doi: 10.1038/ni.2537

81. Peng RJ, Han BW, Cai QQ, Zuo XY, Xia T, Chen JR, et al. Genomic and transcriptomic landscapes of Epstein-Barr virus in extranodal natural killer T-cell lymphoma. *Leukemia* (2019) 33(6):1451–62. doi: 10.1038/s41375-018-0324-5

82. de Mel S, Hue SS, Jeyasekharan AD, Chng WJ, Ng SB. Molecular pathogenic pathways in extranodal Nk/T cell lymphoma. *J Hematol Oncol* (2019) 12(1):33. doi: 10.1186/s13045-019-0716-7

83. Bouchekioua A, Scourzic L, de Wever O, Zhang Y, Cervera P, Aline-Fardin A, et al. Jak3 deregulation by activating mutations confers invasive growth advantage in extranodal nasal-type natural killer cell lymphoma. *Leukemia* (2014) 28(2):338–48. doi: 10.1038/leu.2013.157

84. Coppo P, Gouilleux-Gruart V, Huang Y, Bouhlal H, Bouamar H, Bouchet S, et al. Stat3 transcription factor is constitutively activated and is oncogenic in nasal-type Nk/T-cell lymphoma. *Leukemia* (2009) 23(9):1667–78. doi: 10.1038/leu.2009.91

85. Chen YW, Guo T, Shen L, Wong KY, Tao Q, Choi WW, et al. Receptor-type tyrosine-protein phosphatase K directly targets Stat3 activation for tumor suppression in nasal Nk/T-cell lymphoma. *Blood* (2015) 125(10):1589–600. doi: 10.1182/blood-2014-07-588970

86. Liu X, Wang B, Ma X, Guo Y. Nf-kappab activation through the alternative pathway correlates with chemoresistance and poor survival in extranodal Nk/T-cell lymphoma, nasal type. *Japanese J Clin Oncol* (2009) 39(7):418–24. doi: 10.1093/jjco/hyp037

87. Ng SB, Selvarajan V, Huang G, Zhou J, Feldman AL, Law M, et al. Activated oncogenic pathways and therapeutic targets in extranodal nasal-type Nk/T cell lymphoma revealed by gene expression profiling. *J Pathol* (2011) 223(4):496–510. doi: 10.1002/path.2823

88. Vousden KH, Lane DP. P53 in health and disease. *Nat Rev Mol Cell Biol* (2007) 8 (4):275–83. doi: 10.1038/nrm2147

89. Jo JC, Kim M, Choi Y, Kim HJ, Kim JE, Chae SW, et al. Expression of programmed cell death 1 and programmed cell death ligand 1 in extranodal Nk/T-cell lymphoma, nasal type. *Ann Hematol* (2017) 96(1):25–31. doi: 10.1007/s00277-016-2818-4

90. Bi XW, Wang H, Zhang WW, Wang JH, Liu WJ, Xia ZJ, et al. Pd-L1 is upregulated by ebv-driven Lmp1 through nf-Kb pathway and correlates with poor prognosis in natural Killer/T-cell lymphoma. *J Hematol Oncol* (2016) 9(1):109. doi: 10.1186/s13045-016-0341-7

91. O'Connor MJ. Targeting the DNA damage response in cancer. *Mol Cell* (2015) 60(4):547–60. doi: 10.1016/j.molcel.2015.10.040

92. Liu A, Takakuwa T, Luo WJ, Fujita S, Aozasa K. Alterations in atr in nasal Nk/T-cell lymphoma and chronic active Epstein-Barr virus infection. *Cancer Sci* (2006) 97 (7):605–10. doi: 10.1111/j.1349-7006.2006.00226.x

93. Folkman J. Tumor angiogenesis: therapeutic implications. *New Engl J Med* (1971) 285(21):1182–6. doi: 10.1056/nejm197111182852108

94. Huynh KD, Fischle W, Verdin E, Bardwell VJ. Bcor, a novel corepressor involved in bcl-6 repression. *Genes Dev* (2000) 14(14):1810–23.

95. Milne TA, Briggs SD, Brock HW, Martin ME, Gibbs D, Allis CD, et al. Mll targets set domain methyltransferase activity to hox gene promoters. *Mol Cell* (2002) 10 (5):1107–17. doi: 10.1016/s1097-2765(02)00741-4

96. Tang T, Tay K, Tao M, Quek RHH, Farid M, Lim ST. A phase ii study of bortezomib-gifox (Gemcitabine, ifosfamide, oxaliplatin) in patients with newly diagnosed natural-Killer/T-Cell lymphoma. *Blood* (2016) 128(22):5353–. doi: 10.1182/blood.V128.22.5353.5353%JBlood

97. Lee J, Suh C, Kang HJ, Ryoo BY, Huh J, Ko YH, et al. Phase I study of proteasome inhibitor bortezomib plus chop in patients with advanced, aggressive T-cell or Nk/T-cell lymphoma. *Ann Oncol* (2008) 19(12):2079–83. doi: 10.1093/annonc/mdn431

98. Karube K, Tsuzuki S, Yoshida N, Arita K, Liu F, Kondo E, et al. Lineage-specific growth inhibition of nk cell lines by Foxo3 in association with akt activation status. *Exp Hematol* (2012) 40(12):1005–15.e6. doi: 10.1016/j.exphem.2012.08.005

99. Karube K, Nakagawa M, Tsuzuki S, Takeuchi I, Honma K, Nakashima Y, et al. Identification of Foxo3 and Prdm1 as tumor-suppressor gene candidates in nk-cell neoplasms by genomic and functional analyses. *Blood* (2011) 118(12):3195–204. doi: 10.1182/blood-2011-04-346890

100. Takahara M, Kishibe K, Bandoh N, Nonaka S, Harabuchi Y. P53, n- and K-ras, and beta-catenin gene mutations and prognostic factors in nasal Nk/T-cell lymphoma from Hokkaido, Japan. *Hum Pathol* (2004) 35(1):86–95. doi: 10.1016/j.humpath.2003.08.025

101. Kim H, Ko YH. The pathologic and genetic characteristics of extranodal Nk/T-cell lymphoma. *Life (Basel Switzerland)* (2022) 12(1):1–19. doi: 10.3390/life12010073

102. Mundy-Bosse BL, Weigel C, Wu YZ, Abdelbaky S, Youssef Y, Casas SB, et al. Identification and targeting of the developmental blockade in extranodal natural Killer/T-cell lymphoma. *Blood Cancer Discovery* (2022) 3(2):154–69. doi: 10.1158/2643-3230.Bcd-21-0098

103. Jo JC, Yoon DH, Kim S, Lee BJ, Jang YJ, Park CS, et al. Clinical features and prognostic model for extranasal Nk/T-cell lymphoma. *Eur J haematology* (2012) 89 (2):103–10. doi: 10.1111/j.1600-0609.2012.01796.x

104. Yan Z, Huang HQ, Wang XX, Gao Y, Zhang YJ, Bai B, et al. A tnm staging system for nasal Nk/T-cell lymphoma. *PloS One* (2015) 10(6):e0130984. doi: 10.1371/journal.pone.0130984

105. Kim TM, Heo DS. Extranodal nk / T-cell lymphoma, nasal type: new staging system and treatment strategies. *Cancer Sci* (2009) 100(12):2242–8. doi: 10.1111/j.1349-7006.2009.01319.x

106. Chang SE, Yoon GS, Huh J, Choi JH, Sung KJ, Moon KC, et al. Comparison of primary and secondary cutaneous Cd56+ Nk/T cell lymphomas. *Appl immunohistochemistry Mol morphology AIMM* (2002) 10(2):163–70. doi: 10.1097/00129039-200206000-00012

107. Elstrom R, Guan L, Baker G, Nakhoda K, Vergilio JA, Zhuang H, et al. Utility of fdg-pet scanning in lymphoma by who classification. *Blood* (2003) 101(10):3875–6. doi: 10.1182/blood-2002-09-2778

108. Zhou X, Lu K, Geng L, Li X, Jiang Y, Wang X. Utility of Pet/Ct in the diagnosis and staging of extranodal natural Killer/T-cell lymphoma: a systematic review and meta-analysis. *Medicine* (2014) 93(28):e258. doi: 10.1097/md.0000000000000258

109. Moon SH, Cho SK, Kim WS, Kim SJ, Chan Ahn Y, Choe YS, et al. The role of 18f-fdg Pet/Ct for initial staging of nasal type natural Killer/T-cell lymphoma: a comparison with conventional staging methods. *J Nucl Med* (2013) 54(7):1039–44. doi: 10.2967/jnumed.112.113399

110. Zhou Z, Chen C, Li X, Li Z, Zhang X, Chang Y, et al. Evaluation of bone marrow involvement in extranodal Nk/T cell lymphoma by fdg-Pet/Ct. *Ann Hematol* (2015) 94(6):963–7. doi: 10.1007/s00277-014-2289-4

111. Liu C, Zhang Y, Zhang Y, Wang M, Liu R, Liu X, et al. Diagnostic value of 18f-fdg Pet/Ct for cutaneous extranodal natural Killer/T-cell lymphoma, nasal type. *Nucl Med Commun* (2016) 37(5):446–52. doi: 10.1097/mnm.0000000000000463

112. Bai B, Huang HQ, Cai QC, Fan W, Wang XX, Zhang X, et al. Predictive value of pretreatment positron emission Tomography/Computed tomography in patients with newly diagnosed extranodal natural Killer/T-cell lymphoma. *Med Oncol (Northwood London England)* (2013) 30(1):339. doi: 10.1007/s12032-012-0339-0

113. Vercellino L, Cottereau AS, Casasnovas O, Tilly H, Feugier P, Chartier L, et al. High total metabolic tumor volume at baseline predicts survival independent of response to therapy. *Blood* (2020) 135(16):1396–405. doi: 10.1182/blood.2019003526

114. Kim CY, Hong CM, Kim DH, Son SH, Jeong SY, Lee SW, et al. Prognostic value of whole-body metabolic tumour volume and total lesion glycolysis measured on ^{18}F-fdg Pet/Ct in patients with extranodal Nk/T-cell lymphoma. *Eur J Nucl Med Mol Imaging* (2013) 40(9):1321–9. doi: 10.1007/s00259-013-2443-6

115. A predictive model for aggressive non-hodgkin's lymphoma. *New Engl J Med* (1993) 329(14):987–94. doi: 10.1056/nejm199309303291402

116. Lee J, Suh C, Park YH, Ko YH, Bang SM, Lee JH, et al. Extranodal natural killer T-cell lymphoma, nasal-type: a prognostic model from a retrospective multicenter study. *J Clin Oncol* (2006) 24(4):612–8. doi: 10.1200/jco.2005.04.1384

117. Suzuki R, Yamaguchi M, Izutsu K, Yamamoto G, Takada K, Harabuchi Y, et al. Prospective measurement of Epstein-Barr virus-DNA in plasma and peripheral blood mononuclear cells of extranodal Nk/T-cell lymphoma, nasal type. *Blood* (2011) 118 (23):6018–22. doi: 10.1182/blood-2011-05-354142

118. Kim SJ, Yoon DH, Jaccard A, Chng WJ, Lim ST, Hong H, et al. A prognostic index for natural killer cell lymphoma after non-Anthracycline-Based treatment: a multicentre, retrospective analysis. *Lancet Oncol* (2016) 17(3):389–400. doi: 10.1016/s1470-2045(15)00533-1

119. Wang XX, Li PF, Bai B, Gao Y, Rong QX, Cai QQ, et al. Differential clinical significance of pre-, interim-, and post-treatment plasma Epstein-Barr virus DNA load in Nk/T-cell lymphoma treated with p-gemox protocol. *Leukemia lymphoma* (2019) 60 (8):1917–25. doi: 10.1080/10428194.2018.1563690

120. Kwong YL, Pang AW, Leung AY, Chim CS, Tse E. Quantification of circulating Epstein-Barr virus DNA in Nk/T-cell lymphoma treated with the smile protocol: diagnostic and prognostic significance. *Leukemia* (2014) 28(4):865–70. doi: 10.1038/leu.2013.212

121. Khong PL, Huang B, Lee EY, Chan WK, Kwong YL. Midtreatment ^{18}F-fdg Pet/Ct scan for early response assessment of smile therapy in natural Killer/T-cell lymphoma: a prospective study from a single center. *J Nucl Med* (2014) 55(6):911–6. doi: 10.2967/jnumed.113.131946

122. Kim SJ, Choi JY, Hyun SH, Ki CS, Oh D, Ahn YC, et al. Risk stratification on the basis of deauville score on pet-ct and the presence of Epstein-Barr virus DNA after completion of primary treatment for extranodal natural Killer/T-cell lymphoma, nasal type: a multicentre, retrospective analysis. *Lancet Haematology* (2015) 2(2):e66–74. doi: 10.1016/s2352-3026(15)00002-2

123. Tian XP, Ma SY, Young KH, Ong CK, Liu YH, Li ZH, et al. A composite single-nucleotide polymorphism prediction signature for extranodal natural Killer/T-cell lymphoma. *Blood* (2021) 138(6):452–63. doi: 10.1182/blood.2020010637

124. Tse E, Zhao WL, Xiong J, Kwong YL. How we treat Nk/T-cell lymphomas. *J Hematol Oncol* (2022) 15(1):74. doi: 10.1186/s13045-022-01293-5

125. Yang Y, Zhang YJ, Zhu Y, Cao JZ, Yuan ZY, Xu LM, et al. Prognostic nomogram for overall survival in previously untreated patients with extranodal Nk/T-cell lymphoma, nasal-type: a multicenter study. *Leukemia* (2015) 29(7):1571–7. doi: 10.1038/leu.2015.44

126. Chen SY, Yang Y, Qi SN, Wang Y, Hu C, He X, et al. Validation of nomogram-revised risk index and comparison with other models for extranodal nasal-type Nk/T-cell lymphoma in the modern chemotherapy era: indication for prognostication and clinical decision-making. *Leukemia* (2021) 35(1):130–42. doi: 10.1038/s41375-020-0791-3

127. Yamaguchi M, Kita K, Miwa H, Nishii K, Oka K, Ohno T, et al. Frequent expression of p-Glycoprotein/Mdr1 by nasal T-cell lymphoma cells. *Cancer* (1995) 76(11):2351–6. doi: 10.1002/1097-0142(19951201)76:11<2351::aid-cncr2820761125>3.0.co;2-1

128. de Pádua Covas Lage LA, Machado PPF, Reichert CO, Miranda E, Culler HF, da Siqueira SAC, et al. High-dose extended-field radiotherapy plus chemotherapy improved survival in extranodal Nk/T-cell lymphoma in a real-life setting: results from the multicenter T-cell Brazil project. *Sci Rep* (2022) 12(1):20557. doi: 10.1038/s41598-022-25034-3

129. Vargo JA, Patel A, Glaser SM, Balasubramani GK, Farah RJ, Marks SM, et al. The impact of the omission or inadequate dosing of radiotherapy in extranodal natural killer T-cell lymphoma, nasal type, in the united states. *Cancer* (2017) 123(16):3176–85. doi: 10.1002/cncr.30697

130. Yang Y, Cao JZ, Lan SM, Wu JX, Wu T, Zhu SY, et al. Association of improved locoregional control with prolonged survival in early-stage extranodal nasal-type natural Killer/T-cell lymphoma. *JAMA Oncol* (2017) 3(1):83–91. doi: 10.1001/jamaoncol.2016.5094

131. Kim GE, Cho JH, Yang WI, Chung EJ, Suh CO, Park KR, et al. Angiocentric lymphoma of the head and neck: patterns of systemic failure after radiation treatment. *J Clin Oncol* (2000) 18(1):54–63. doi: 10.1200/jco.2000.18.1.54

132. Huang MJ, Jiang Y, Liu WP, Li ZP, Li M, Zhou L, et al. Early or up-front radiotherapy improved survival of localized extranodal Nk/T-cell lymphoma, nasal-type in the upper aerodigestive tract. *Int J Radiat oncology biology Phys* (2008) 70 (1):166–74. doi: 10.1016/j.ijrobp.2007.05.073

133. Kim SJ, Yang DH, Kim JS, Kwak JY, Eom HS, Hong DS, et al. Concurrent chemoradiotherapy followed by l-Asparaginase-Containing chemotherapy, vidl, for localized nasal extranodal Nk/T cell lymphoma: Cisl08-01 phase ii study. *Ann Hematol* (2014) 93(11):1895–901. doi: 10.1007/s00277-014-2137-6

134. Yamaguchi M, Tobinai K, Oguchi M, Ishizuka N, Kobayashi Y, Isobe Y, et al. Phase I/Ii study of concurrent chemoradiotherapy for localized nasal natural Killer/T-cell lymphoma: Japan clinical oncology group study Jcog0211. *J Clin Oncol* (2009) 27 (33):5594–600. doi: 10.1200/jco.2009.23.8295

135. Yamaguchi M, Tobinai K, Oguchi M, Ishizuka N, Kobayashi Y, Isobe Y, et al. Concurrent chemoradiotherapy for localized nasal natural Killer/T-cell lymphoma: an updated analysis of the Japan clinical oncology group study Jcog0211. *J Clin Oncol* (2012) 30(32):4044–6. doi: 10.1200/jco.2012.45.6541

136. Kim SJ, Kim K, Kim BS, Kim CY, Suh C, Huh J, et al. Phase ii trial of concurrent radiation and weekly cisplatin followed by vipd chemotherapy in newly diagnosed, stage ie to iie, nasal, extranodal Nk/T-cell lymphoma: consortium for improving survival of lymphoma study. *J Clin Oncol* (2009) 27(35):6027–32. doi: 10.1200/jco.2009.23.8592

137. Jing XM, Zhang ZH, Wu P, Zhang SC, Ren YR, Xiong ZJ, et al. Efficacy and tolerance of pegaspargase, gemcitabine and oxaliplatin with sandwiched radiotherapy in the treatment of newly-diagnosed extranodal nature killer (Nk)/T cell lymphoma. *Leukemia Res* (2016) 47:26–31. doi: 10.1016/j.leukres.2016.05.004

138. Zhu Y, Tian S, Xu L, Ma Y, Zhang W, Wang L, et al. Gelad chemotherapy with sandwiched radiotherapy for patients with newly diagnosed stage Ie/Iie natural Killer/T-cell lymphoma: a prospective multicentre study. *Br J haematology* (2022) 196(4):939–46. doi: 10.1111/bjh.17960

139. Qi SN, Yang Y, Song YQ, Wang Y, He X, Hu C, et al. First-line non-Anthracycline-Based chemotherapy for extranodal nasal-type Nk/T-cell lymphoma: a retrospective analysis from the clcg. *Blood Adv* (2020) 4(13):3141–53. doi: 10.1182/bloodadvances.2020001852

140. Pokrovsky VS, Vinnikov D. L-asparaginase for newly diagnosed extra-nodal Nk/T-cell lymphoma: systematic review and meta-analysis. *Expert Rev Anticancer Ther* (2017) 17(8):759–68. doi: 10.1080/14737140.2017.1344100

141. Yamaguchi M, Kwong YL, Kim WS, Maeda Y, Hashimoto C, Suh C, et al. Phase ii study of smile chemotherapy for newly diagnosed stage iv, relapsed, or refractory extranodal natural killer (Nk)/T-cell lymphoma, nasal type: the nk-cell tumor study group study. *J Clin Oncol* (2011) 29(33):4410–6. doi: 10.1200/jco.2011.35.6287

142. Bi XW, Jiang WQ, Zhang WW, Huang JJ, Xia Y, Wang Y, et al. Treatment outcome of patients with advanced stage natural Killer/T-cell lymphoma: elucidating the effects of asparaginase and postchemotherapeutic radiotherapy. *Ann Hematol* (2015) 94(7):1175–84. doi: 10.1007/s00277-015-2336-9

143. Suzuki R, Yamaguchi M, Oguchi M, Asano N, Tomita N, Amaki J, et al. Extranodal Nk/T-cell lymphoma, nasal type of advanced stage: nkea (Next-generation therapy for Nk/T-cell lymphoma in East Asia) project. *22nd Congress Eur Hematol Association: EHA Library* (2016).

144. Li X, Cui Y, Sun Z, Zhang L, Li L, Wang X, et al. Ddgp versus smile in newly diagnosed advanced natural Killer/T-cell lymphoma: a randomized controlled, multicenter, open-label study in China. *Clin Cancer Res* (2016) 22(21):5223–8. doi: 10.1158/1078-0432.Ccr-16-0153

145. Wang X, Zhang L, Liu X, Li X, Li L, Fu X, et al. Efficacy and survival in newly diagnosed advanced extranodal natural Killer/T-cell lymphoma: a randomized, controlled, multicenter and open-labled study with ddgp regimen versus smile regimen. *Blood* (2019) 134(Supplement_1):463–. doi: 10.1182/blood-2019-127811%JBlood

146. Wang X, Hu J, Dong M, Ding M, Zhu L, Wu J, et al. Ddgp vs. smile in Relapsed/Refractory extranodal natural Killer/T-cell lymphoma, nasal type: a retrospective study of 54 patients. *Clin Trans Sci* (2021) 14(1):405–11. doi: 10.1111/cts.12893

147. Kharfan-Dabaja MA, Kumar A, Ayala E, Hamadani M, Reimer P, Gisselbrecht C, et al. Clinical practice recommendations on indication and timing of hematopoietic cell transplantation in mature T cell and Nk/T cell lymphomas: an international collaborative effort on behalf of the guidelines committee of the American society for blood and marrow transplantation. *Biol Blood marrow Transplant* (2017) 23(11):1826–38. doi: 10.1016/j.bbmt.2017.07.027

148. Lee J, Au WY, Park MJ, Suzumiya J, Nakamura S, Kameoka J, et al. Autologous hematopoietic stem cell transplantation in extranodal natural Killer/T cell lymphoma: a

multinational, multicenter, matched controlled study. *Biol Blood marrow Transplant* (2008) 14(12):1356–64. doi: 10.1016/j.bbmt.2008.09.014

149. Yhim HY, Kim JS, Mun YC, Moon JH, Chae YS, Park Y, et al. Clinical outcomes and prognostic factors of up-front autologous stem cell transplantation in patients with extranodal natural Killer/T cell lymphoma. *Biol Blood marrow Transplant* (2015) 21(9):1597–604. doi: 10.1016/j.bbmt.2015.05.003

150. Jaccard A, Gachard N, Marin B, Rogez S, Audrain M, Suarez F, et al. Efficacy of l-asparaginase with methotrexate and dexamethasone (Aspametdex regimen) in patients with refractory or relapsing extranodal Nk/T-cell lymphoma, a phase 2 study. *Blood* (2011) 117(6):1834–9. doi: 10.1182/blood-2010-09-307454

151. Wang JH, Wang L, Liu CC, Xia ZJ, Huang HQ, Lin TY, et al. Efficacy of combined gemcitabine, oxaliplatin and pegaspargase (P-gemox regimen) in patients with newly diagnosed advanced-stage or Relapsed/Refractory extranodal Nk/T-cell lymphoma. *Oncotarget* (2016) 7(20):29092–101. doi: 10.18632/oncotarget.8647

152. Wang JJ, Dong M, He XH, Li YX, Wang WH, Liu P, et al. Gdp (Gemcitabine, dexamethasone, and cisplatin) is highly effective and well-tolerated for newly diagnosed stage iv and Relapsed/Refractory extranodal natural Killer/T-cell lymphoma, nasal type. *Medicine* (2016) 95(6):e2787. doi: 10.1097/md.0000000000002787

153. Hamadani M, Kanate AS, DiGilio A, Ahn KW, Smith SM, Lee JW, et al. Allogeneic hematopoietic cell transplantation for aggressive nk cell leukemia. a center for international blood and marrow transplant research analysis. *Biol Blood marrow Transplant* (2017) 23(5):853–6. doi: 10.1016/j.bbmt.2017.01.082

154. Novelli S, Bento L, Garcia I, Prieto L, López L, Gutierrez G, et al. Allogeneic stem cell transplantation in mature T cell and natural Killer/T neoplasias: a registry study from Spanish Geth/Geltamo centers. *Transplant Cell Ther* (2021) 27(6):493. doi: 10.1016/j.jtct.2021.03.014

155. Chen BJ, Chapuy B, Ouyang J, Sun HH, Roemer MG, Xu ML, et al. Pd-L1 expression is characteristic of a subset of aggressive b-cell lymphomas and virus-associated malignancies. *Clin Cancer Res* (2013) 19(13):3462–73. doi: 10.1158/1078-0432.Ccr-13-0855

156. Kwong YL, Chan TSY, Tan D, Kim SJ, Poon LM, Mow B, et al. Pd1 blockade with pembrolizumab is highly effective in relapsed or refractory Nk/T-cell lymphoma failing l-asparaginase. *Blood* (2017) 129(17):2437–42. doi: 10.1182/blood-2016-12-756841

157. Chan TSY, Li J, Loong F, Khong PL, Tse E, Kwong YL. Pd1 blockade with low-dose nivolumab in Nk/T cell lymphoma failing l-asparaginase: efficacy and safety. *Ann Hematol* (2018) 97(1):193–6. doi: 10.1007/s00277-017-3127-2

158. Xia L, Zhang HS, Bao J, Zhao YC, Xia HL. Successful outcome of programmed death 1 blockade plus gemox for Epstein-Barr virus-associated primary nodal T/Nk cell lymphoma: a case report. *Front Oncol* (2021) 11:706865. doi: 10.3389/fonc.2021.706865

159. Zhang L, Liu X, Wang X, Chang Y, Fu X, Li X, et al. P1240: anti-Pd-1-Antibody (Tislelizumab) combined with deacetylase inhibitor (Chidamide), lenalidomide and etoposide for the treatment of Refractory/Relapsed extranodal natural Killer/T cell lymphoma, nasal. *Hemasphere*. (2022) 6(Suppl):1125–6. doi: 10.1097/01.HS9.0000847824.69595.f0

160. Lim JQ, Huang D, Tang T, Tan D, Laurensia Y, Peng RJ, et al. Whole-genome sequencing identifies responders to pembrolizumab in Relapse/Refractory natural-Killer/T cell lymphoma. *Leukemia* (2020) 34(12):3413–9. doi: 10.1038/s41375-020-1000-0

161. Tao R, Fan L, Song Y, Hu Y, Zhang W, Wang Y, et al. Sintilimab for Relapsed/Refractory extranodal Nk/T cell lymphoma: a multicenter, single-arm, phase 2 trial (Orient-4). *Signal transduction targeted Ther* (2021) 6(1):365. doi: 10.1038/s41392-021-00768-0

162. Kim SJ, Lim JQ, Laurensia Y, Cho J, Yoon SE, Lee JY, et al. Avelumab for the treatment of relapsed or refractory extranodal Nk/T-cell lymphoma: an open-label phase 2 study. *Blood* (2020) 136(24):2754–63. doi: 10.1182/blood.2020007247

163. Huang H, Zhu J, Yao M, Kim TM, Yoon DH, Cho SG, et al. Daratumumab monotherapy for patients with relapsed or refractory natural Killer/T-cell lymphoma, nasal type: an open-label, single-arm, multicenter, phase 2 study. *J Hematol Oncol* (2021) 14(1):25. doi: 10.1186/s13045-020-01020-y

164. Geng L, Li X, Zhou X, Fang X, Yuan D, Wang X. Wp1066 exhibits antitumor efficacy in Nasal–Type natural Killer/T-cell lymphoma cells through downregulation of the Stat3 signaling pathway. *Oncol Rep* (2016) 36(5):2868–74. doi: 10.3892/or.2016.5091

165. Ando S, Kawada JI, Watanabe T, Suzuki M, Sato Y, Torii Y, et al. Tofacitinib induces G1 cell-cycle arrest and inhibits tumor growth in Epstein-Barr virus-associated T and natural killer cell lymphoma cells. *Oncotarget* (2016) 7(47):76793–805. doi: 10.18632/oncotarget.12529

166. Yan G, H-q H, Li P, Bing B, Xiaoxiao W, Qixiang R, et al. Chidamide ,Oral subtype-selective histone deacetylase inhibitor (Hdaci) monotherapy was effective on the patients with relapsed or refractory extranodal natural killer (Nk)/T-cell lymphoma. *Blood* (2017) 130(Supplement 1):2797–. doi: 10.1182/blood.V130.Suppl_1.2797.2797%JBlood

167. Cho SG, Kim N, Sohn HJ, Lee SK, Oh ST, Lee HJ, et al. Long-term outcome of extranodal Nk/T cell lymphoma patients treated with postremission therapy using ebv Lmp1 and Lmp2a-specific ctls. *Mol Ther* (2015) 23(8):1401–9. doi: 10.1038/mt.2015.91

168. Haverkos BM, Alpdogan O, Baiocchi R, Brammer JE, Feldman TA, Capra M, et al. Nanatinostat (Nstat) and valganciclovir (Vgcv) in Relapsed/Refractory (R/R) Epstein-Barr virus-positive (Ebv +) lymphomas: final results from the phase 1b/2 Vt3996-201 study. *Blood* (2021) 138(Supplement 1):623–. doi: 10.1182/blood-2021-152603%JBlood

169. Luo L, Zhou X, Zhou L, Liang Z, Yang J, Tu S, et al. Current state of car-T therapy for T-cell malignancies. *Ther Adv Hematol* (2022) 13:20406207221143025. doi: 10.1177/20406207221143025

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 30 August 2023
DOI 10.3389/fonc.2023.1195759

OPEN ACCESS

EDITED BY
Ryan Wilcox,
University of Michigan, United States

REVIEWED BY
Amit Khot,
Peter MacCallum Cancer Center, Australia
Haiwen Huang,
Suzhou University, China

*CORRESPONDENCE
Vanderson Rocha
Vanderson.rocha@hc.fm.usp.br

†These authors have contributed equally to this work

RECEIVED 28 March 2023
ACCEPTED 31 July 2023
PUBLISHED 30 August 2023

CITATION
Couto SCF, Kowes A, Aurabi CS, Oliveira TGM, Klinger P and Rocha V (2023) Autologous, allogeneic hematopoietic cell transplantation and CAR-T/NK therapy: what is their real importance in PTCL? *Front. Oncol.* 13:1195759. doi: 10.3389/fonc.2023.1195759

Autologous, allogeneic hematopoietic cell transplantation and CAR-T/NK therapy: what is their real importance in PTCL?

Samuel C. F. Couto[1,2†], Ariel Kowes[2†], Camila S. Aurabi[2], Theo G. M. Oliveira[1,2], Paulo Klinger[1] and Vanderson Rocha[1,2,3*]

[1]Laboratory of Medical Investigation in Pathogenesis and Directed Therapy in Onco-Immuno-Hematology (LIM-31), Department of Hematology and Cell Therapy, Hospital das Clínicas HCFMUSP, Faculdade de Medicina, University of São Paulo, São Paulo, Brazil, [2]Fundação Pró-Sangue–Hemocentro de São Paulo, São Paulo, Brazil, [3]Instituto D'Or de Ensino e Pesquisa, São Paulo, Brazil

Peripheral T cell lymphoma (PTCL) is a rare and aggressive type of non-Hodgkin's lymphoma that affects mature T cells. This type of cancer is characterized by the abnormal growth of T cells, which can accumulate in the lymph nodes, spleen, bone marrow, and other organs, leading to a variety of symptoms. PTCLs are often difficult to diagnose and treat, and they have a poorer prognosis than other types of lymphoma. However, recent advancements in treatment options, such as targeted therapies have shown promise in improving outcomes for patients with PTCL. Here, we discuss the use of autologous and allogeneic hematopoietic cell transplantation (HCT) as a treatment strategy for patients with PTCL, as well as the recent treatment approaches based on advanced cellular therapy. The current evidence for the use of HCT in PTCL is mainly derived from registry data, retrospective studies, and expert opinion, as randomized trials are limited due to the low incidence and histological heterogeneity of PTCL subtypes.

KEYWORDS

hematopoietic cell transplantation, peripheral T-cell lymphoma, CAR-T cell, CAR-NK cell, immunotherapy

1 Introduction

Peripheral T-cell lymphomas encompass a biologically and clinically heterogeneous group of lymphoproliferative disorders derived from mature T-cells (post-thymic lymphocytes). Much less frequent than B-cell lymphomas, they represent about 10-15% of the non-Hodgkin lymphomas (NHL) in the Western hemisphere (1, 2). On the most recent World Health Organization Classification of Hematolymphoid Tumors (WHO-HAEM5), the mature T-cell and NK-cell neoplasms have been grouped into 9 families based on characteristics like cell of origin/differentiation state, clinical scenario, disease

localization and cytomorphology (3). PTCL most common subtypes are, in order of incidence, PTCL-not otherwise specified (PTCL-NOS), angioimmunoblastic T-cell lymphoma (AITL), anaplastic large cell lymphoma (ALCL), anaplastic lymphoma kinase–positive (ALCL ALK-positive), anaplastic large cell lymphoma, anaplastic lymphoma kinase–negative (ALCL ALK-negative), and enteropathy-associated T-cell lymphoma (EATL), and each of them is characterized by unique genetic, molecular, histopathologic, and clinical features (2, 4). Overall, PTCL patients have dismal prognosis, and the currently available treatment strategies are still unsatisfactory for both front line and relapsed/refractory (R/R) settings. ALCL, ALK-positive subtype, might be an exception according to the International T-Cell Project, which reported a 5-year overall survival (OS) of 70% (5). Regimens derived from the existing protocols for B-cell non-Hodgkin lymphomas, like CHOP (cyclophosphamide, doxorubicin, vincristine, prednisone) and CHOP-like, are the ones used as first-line therapy and depending on the patient's performance status and response to chemotherapy, consolidation with high-dose chemotherapy and hematopoietic cell transplantation (HCT) can improve the outcomes. Nevertheless, the risk of refractoriness or relapse is high for most patients (1). Due to the PTCL disorders heterogeneity and their low incidence, it is more difficult to conduct prospective studies, and for that reason most of the current evidence for the treatment comes from phase II trials, retrospective studies, and expert opinion. Therefore, PTCL still represents a therapeutic challenge. The aim of this study is to discuss the existing data and present a review over the role of autologous and allogeneic HCT for PTCL, and the role of more recent cellular therapy, such as chimeric antigen receptor-T and -NK cell (CAR-T and CAR-NK) in the treatment of these diseases. To comprehensively summarize the data, we created two tables incorporating patient characteristics, transplant settings, histology subtypes, conditioning regimens, and outcomes, extracted from diverse studies (Tables 1, 2).

2 First-line HCT in PTCL

The first nation-wide survey conducted in transplanted PTCL patients was in Finland during 1990-2001. Following induction therapy, patients were submitted to high dose therapy (HDT) conditioning regimen before autologous HCT (autoHCT). Of thirty-seven patients assessed, four (11%) died from treatment related mortality (TRM), 76% (28/37) achieved complete response (CR) and 5% (2/37) were in partial remission (PR). Eight percent of patients (3/37) were refractory to HDT and died from progressive lymphoma. The 5-year OS was 63% after autoHCT in frontline CR/PR vs 45% beyond second treatment. This study included fourteen patients (37%) with ALCL, and these patients had a significantly better OS compared to other PTCL subtypes (85% vs 35%). Although relatively high TRM rate (11%), this nation-wide survey supports the idea that HDT followed by autoHCT is feasible and that higher survival rates correlates with response to treatment and with specific PTCL subtypes. However, prospective randomized trials are needed to better determine the impact in up-front PTCL therapy (6). Rodriguez et al. described the GEL-TAMO (Grupo Español de Linfomas/Trasplante Autólogo de Médula Ósea) experience in patients with PTCL who underwent HDT and autoHCT. One hundred and fifteen patients were included and given mostly anthracycline-based regimens prior to transplant. Complete response was achieved in 86% of patients (98/114). Six patients (5%) achieved PR, 3% (3/114) had stable disease and 6% (7/114) disease progression. At a median follow-up of 37 months, 73 (64%) patients were alive with an estimated 5-year OS of 56% and disease-free survival (DFS) of 60%. Forty-two patients (37%) had died, with the main cause being disease progression in 32 of these patients (76%). The 5-year OS in first-line CR was significantly higher in comparison to second-line or more CR prior to transplant (80% and 50%, respectively). Data from this study shows a higher survivability in chemosensitive PTCL patients. Prognostic criteria, like adjusted-IPI (a-IPI) higher than 1 and altered LDH were associated with lower 5-year OS (a-IPI 0-1: 65% vs a-IPI>1: 13%; LDH normal 55% vs high LDH: 22%) in univariate analysis and therefore, might have utility in predicting clinical outcomes (14).

A meta-analysis published in 2016 by El-Asmar et al. (25) collected and analyzed data from 27 studies (3 prospectives and 16 retrospectives for front-line) and assessed the efficacy of HDT with autoHCT in front-line and R/R consolidation in PTCL. The pooled prospective and retrospective trials included a total of 179 and 599 patients, respectively. Interestingly, as expected, there was a clear difference between the OS and progression-free survival (PFS) on the different types of studies, with OS rates of 54% and PFS 33% in prospective, 68% and 55% in the retrospective studies, respectively. TRM pooled rates in prospective were 2% versus 6% in retrospective studies. These results are probably due to better control of confounding factors and accurately defining death causality in prospective trials. Although with several limitations in this study and the lack of randomized trials, the meta-analysis allowed strong evidence to support HDT and autoHCT as a reasonable option in front-line treatment for PTCL (25).

Moreover, Yam and colleagues compared PFS in first complete response (CR1) PTCL patients submitted to autoHCT or active observation after CHOP-like regimens. In a total of 48 patients (28 in the observation group and 20 submitted to consolidation transplant), the median follow-up duration was 26.4 months. The median PFS was 15.8 months for the observation patients and 12.8 months for patients who underwent autoHCT. The estimated 3-year PFS was 37% and 41% for the observation and transplantation groups, respectively. The results of this study revealed no improvement in PFS and OS in patients who achieved CR1 with observation or autoHCT. Even though these findings have several limitations, including the nature of the study, it shows the diversified data between different trials in PTCL patients, and prospective randomized trials could provide stronger evidence in this matter (7).

A prospective PTCL-restricted multicenter study that evaluated the role of frontline therapy with myeloablative chemoradiotherapy (CRT) and autoHCT was conducted by Reimer et al. From June 2000 to April 2006, a group of 83 patients with PTCL were given four to six cycles of CHOP, and if at least PR was reached, they proceeded to mobilization and were submitted to CRT followed by autoHCT. After CHOP therapy, patients had an overall response

rate (ORR) of 79% (39% CR and 40% PR), and 66% (55/83) patients completed myeloablative therapy and proceeded to autoHCT. The main reason for not undergoing transplant was disease progression (22 patients). Following transplantation, 48 out of 55 patients achieved CR and seven patients achieved PR. In the intention to treat analysis, ORR was 66% (58% CR and 8% PR). After a median follow-up of 33 months, 43 patients (52%) were still alive either in remission (35) or with evidence of disease (8). The estimated 3-year

TABLE 1 Studies evaluating autologous HCT in PTCL as first-line treatment.

Study	Year	Patients (n)	Histology subtypes (most common/ALK+ ALCL)	Pre-transplant Response (CR/PR)	Conditioning, Regimen	TRM	Survival	Median follow-up
RETROSPECTIVE STUDIES								
Jantunen et al. (6)	2004	37	14 PTCL-NOS 14 ALCL 9 other (ALK+ ALCL included)	28 CR 2 PR	BEAC (22%) BEAM (15%)	4 (11%)	5-year: OS 54% PFS 44% (OS ALCL 85 *vs* 35% other subtypes)	24 months
Yam et al. (7)	2016	48 (20 autoHCT / 28 observation)	6 PTCL-NOS 6 AITL 8 other (ALK+ ALCL excluded)	20 CR	No description	–	3-year: OS 72% PFS 41%	26.4 months
PROSPECTIVE STUDIES								
Reimer et al. (8)	2009	83 (55 autoHCT)	32 PTCL-NOS 27 AITL 24 other (ALK+ ALCL excluded)	40 CR 15 PR	Cy/TBI	3 (3.6%)	3-year: OS 48%	33 months
Corradini et al. (9)	2006	62 (46 autoHCT)	28 PTCL-NOS 19 ALK+ ALCL 15 other (ALK+ ALCL included)	32 CR 10 PR	Mitoxantrone + Melphalan BEAM	3 (4.8%)	12-year: OS 34% DFS 55% EFS 30%	76 months
Rodríguez et al. (10)	2007	26 (19 autoHCT)	11 PTCL-NOS 8 ALK+ ALCL 7 AITL (ALK+ ALCL included)	17 CR 2 PR	BEAM	–	3-year: OS 73% PFS 53%	35 months
Mercadal et al. (11)	2008	41 (17 autoHCT)	20 PTCL-NOS 12 AITL 9 other (ALK+ ALCL excluded)	20 CR 4 PR	BEAM BEAC	1 (2.4%)	4-year: OS 39%	3.2 years
D'Amore et al. (12)	2012	166 (115 autoHCT)	62 PTCL-NOS 31 ALK- ALCL 30 AITL 21 EATL 16 other (ALK+ ALCL excluded)	82 CR 49 PR	BEAM BEAC	7 (4%)	5-year: OS 51% PFS 44%	60.5 months
Wilhelm et al. (13)	2016	111 (75 autoHCT)	42 PTCL-NOS 37 AITL 16 ALK- ALCL 16 other (ALK+ ALCL included)	69 CR 22 PR	Cy/TBI BEAM	(3,6%)	5-year: OS 44%, DFS 54% PFS 39%	59 months
Registry studies Rodríguez et al. (14)	2003	115	72 PTCL-NOS 25 ALCL 18 other (ALK+ ALCL included)	37 CR 28 CR2 44 PR	BEAM (43%) BEAC (32%) Cy/TBI (12%) Other (12%)	9 (8%)	5-year: OS 56% DFS 60%	37 months
Park et al. (15)	2019	119 (36 autoHCT)	- **autoHCT** 17 AITL 15 PTCL-NOS 4 ALK- ALCL - **non-autoHCT** 18 AITL	CR1	BEAM BEAM variation	–	2-year: - **autoHCT** OS 87.8% (95% CI, 77.3%-99.8%) PFS 57.6 (*P*=0.23)	2.8 years

(Continued)

TABLE 1 Continued

Study	Year	Patients (n)	Histology subtypes (most common/ALK+ ALCL)	Pre-transplant Response (CR/PR)	Conditioning, Regimen	TRM	Survival	Median follow-up
			39 PTCL-NOS 26 ALK- ALCL				- **non-autoHCT** OS 70.2% (95% CI, 60.9%-80.9%) PFS 47.5%	
Al-Mansour et al.	2019	28 (15 autoHCT, 3 did not undergo transplant)	11 PTCL-NOS 10 ALCL 7 AITL (ALK+ ALCL included)	CR/PR	CBV TBI + Etoposide/Cy	–	5-year: - **autoHCT** OS 40% PFS 40% - **non-autoHCT** OS 45% PFS 38%	7.8 years

PTCL-NO, Peripheral T-cell Lymphoma, not Otherwise Specified; ALCL, Anaplastic Large Cell Lymphoma; AITL, Angioimmunoblastic T-Cell Lymphoma, CR, Complete Response; PR, Partial Response; CR1, First Complete Remission; CR2, Second Complete Remission; OS, Overall survival; PFS, Progression-Free Survival; EFS, Event-Free Survival; DFS, Disease-Free Survival; Cy, Cyclophosphamide; TBI, Total Body Irradiation; Beam (Carmustine, etoposide, citarabyne, melphalan); BEAC (Bleomycin, etoposide, doxorubicin, cyclophosphamide; CBV (Carmustine, etoposide, Cyclophosphamide).

TABLE 2 Studies evaluating autologous and allogeneic HCT in R/R PTCL.

Study	Type of transplant	Patients (n)	Histology subtypes	Disease status at HCT	Conditioning, regimen	TRM	Outcomes	Median follow-up
RETROSPECTIVE STUDIES								
Chen et al. (16)	Autologous	53	18 ALCL 16 Unspecified 9 AITL 7 nNK/T 2 HSTL 1 ATLL	15 CR1/PR1 28 CR2/PR2 + 10 Refractory	CR1/PR1: 13 chemotherapy only 2 TBI-based CR2/PR2+: 24 chemotherapy only 4 TBI-based Refractory: 7 chemotherapy only 3 TBI-based	4% NRM	5-year PFS: CR1/PR1 51% CR2/PR2+ 12% Refractory 0 5-year OS: CR1/PR1 76% CR2/PR2 40% Refractory 30%	60 months
Rodriguez et al. (14)	Autologous	115	72 PTCL-NOS 25 ALCL 8 Lymphoepiteloid 6 AITL 3 HSTL 1 EATL	37 CR1 28 CR2+ 44 PR 6 Refractory	50 BEAM 37 BEAC 14 Cy-TBI 10 CVB 4 Others	8% TRM	5-year OS: CR1 80% CR2+ 50% PR1+ 49% Refractory 0	37 months
Huang et al. (17)	Autologous Allogeneic	67 (43 autologous, 24 allogeneic)	Autologous: 20 PTCL-NOS 18 ALCL, ALK- 5 nNK/T Allogeneic: 17 PTCL-NOS 1 ALCL, ALK- 1 AITL 5 nNK/T	Autologous: 20 CR1 6 CR2 7 PR 10 Refractory Allogeneic: 0 CR1 2 CR2 6 PR 16 Refractory	Autologous: 38 BEAM 5 Others Allogeneic: 6 Cy-TBI 18 Bu-Cy	Autologous: 1-year NRM 7% Allogeneic: 1-year NRM 18%	Autologous: Median time from HCT to relapse: 6 months 5-year PFS 49% 5-year OS 59% 3-year PFS 20% (primary refractory specifically) 3 year OS 20% (primary refractory specifically) Allogeneic: Median time from HCT to relapse: 8 months 5-year PFS 54% 5-year OS 55%	Autologous: 31 months Allogeneic: 25.5 months

(Continued)

TABLE 2 Continued

Study	Type of transplant	Patients (n)	Histology subtypes	Disease status at HCT	Conditioning, regimen	TRM	Outcomes	Median follow-up
							3-year PFS 49% (primary refractory specifically) 3 year OS 53% (primary refractory specifically)	
Rohlfing et al. (18)	Autologous Allogeneic	117 (89 R/R)	34 PTCL-NOS 31 ALCL,ALK- 28 AITL 11 nNK/T 10 EATL 3 HSTL	No description	Autologous: Dexa-BEAM Allogeneic: 18 Myeloablative 12 RIC 1 Unknown	Autologous: 0 Allogeneic: 23% TRM	Autologous (n=7): Median survival 10 months Death from PD 100% Allogeneic (n=31): Median survival not reached 5-year OS 52% No transplant (n=51): Median survival 3 months Death from PD 92%	5.8 years
Smith et al. (19)	Autologous Allogeneic	241 (115 autologous, 126 allogeneic)	Autologous: 61 ALCL (ALK +, - and unknown) 39 PTCL-NOS 15 AITL Allogeneic: 51 ALCL (ALK +, - and unknown) 63 PTCL-NOS 12 AITL	Autologous: 40 CR1 24 CR2+ 16 PR1 17 PR2+ 16 Refractory 2 no data Allogeneic: 18 CR1 20 CR2+ 23 PR1 21 PR2+ 41 Refractory 3 no data	Autologous: 26 TBI-based 65 BEAM 14 Cy 4 Bu-Mel / Bu-Cy 6 Other Allogeneic: 74 Myeloablative 45 NST/RIC 7 Unknown	Autologous: 6% NRM Allogeneic: 34% NRM	Autologous: 3-year PFS 42% 3-year OS 53% (excluded CR1) Allogeneic: 3-year PFS 31% 3-year OS 41% (excluded CR1)	48 months
Czajczynska et al. (20)	Allogeneic	24	9 PTCL-NOS 5 AITL 4 ALCL (1 positive / 3 negative) 2 EATL 2 nNK/T 1 T-PLL 1 LTCL	2 CR1 5 CR2 2 CR2+ 6 PR1 4 PR2 1 SD 1 Refractory 1 Resistance relapse 2 Responding relapse	21 BEAM-Alemtuzumab 3 Other	25%	100-day OS 87.5% 1-year OS 58.3% 3-year OS 42.4%	44.8 months
Wulf et al., 2019 (21)	Allogeneic	84	30 PTCL-NOS 17 AITL 15 ALCL 4 nNK/T 5 LTCL 6 T-PLL 7 Other	13 CR 35 PR 14 SD 22 PD/ Refractory	FBC-12 (Myeloablative)	13.1% at 1 year 32.3% at 3 years 46% at 5 years	38.2% OS at median follow-up 37.2% DFS at median follow-up	14.5 months
PROSPECTIVE STUDIES								
Shustov et al. (22)	Allogeneic	17 (14 R/R)	7 PTCL-NOS 4 AITL 3 T-PLL 1 ALCL 2 Other	8 CR 5 PR 2 SD 2 PD	Flu-TBI (non myeloablative)	3-year NRM 19%	3-year OS 59% 3-year PFS 53%	3.3 years
Jacobsen et al. (23)	Allogeneic	52	20 PTCL-NOS 6 ALCL 5 AITL	10 CR1 7 CR2 6 CR3	31 Myeloablative 21 RIC	3-year NRM: 36%	3-year OS 41% 3-year PFS 30% Relapse at 3 years:	49 months

(Continued)

TABLE 2 Continued

Study	Type of transplant	Patients (n)	Histology subtypes	Disease status at HCT	Conditioning, regimen	TRM	Outcomes	Median follow-up
			4 HSTL 4 nNK/T 1 ATLL 1 EATL 9 Other	16 PR 5 Relapse		Myeloablative 14% RIC	Myeloablative 33% RIC 57%	
Corradini et al., 2004 (24)	Allogeneic	17	9 PTCL-NOS 4 AITL 4 ALCL ALK-	1 CR2 12 PR2+ 1 CR3 2 PD 1 untested	RIC	2-year NRM 6%	3-year OS 81% 3-year PFS 64%	28 months

AITL, angioimmunoblastic T-cell lymphoma; ALCL, anaplastic large cell lymphoma; ATLL, adult T-cell leukemia/lymphoma; BEAC, bleomycin, etoposide, doxorubicin and cyclophosphamide; BEAM, carmustine, etoposide, cytarabine and melphalan; Bu-Cy, busulfan-cyclophosphamide; Bu-Mel, busulfan-melphalan; CR1/PR1, first complete or partial response; CR2/PR2+, second or more complete or partial response; CVB, cyclophosphamide, etoposide, BCNU; Cy-TBI, cyclophosphamide-TBI; Dexa-BEAM, dexamethasone-BEAM; DFS, disease-free survival; EATL, enteropathy-associated T-cell lymphoma; FBC-12, fludarabine, busulfan and cyclophosphamide; Flu, fludarabine; HCT, hematopoietic cell transplantation; HSTL, hepatosplenic T-cell lymphoma; LTCL, Lymphoblastic T cell lymphoma; nNK/T, nasal type extranodal NK/T cell lymphoma; NRM, nonrelapse mortality; NST, nonmyeloablative; OS, overall survival; PD, progressive disease; PFS, progression free survival; PR, partial response; PTCL, peripheral T-cell lymphoma; PTCL-NOS, PTCL-not otherwise specified; RIC, reduced intensity conditioning; R/R, relapsed/refractory; SD, stable disease; TBI, total body irradiation; T-PLL, T-cell prolymphocytic leukemia; TRM, treatment related mortality.

OS rate was 48%. The estimated 3-year OS rate was 71% for patients who underwent autoHCT compared with only 11% for patients who did not undergo autoHCT. These findings, although with limitations, suggest a favorable outcome in autoHCT compared with conventional chemotherapy alone (8).

Corradini et al. reported a median 76-month follow-up of 62 patients with PTCL in Italy healthcare institutions. Patients were submitted to high-dose sequential chemotherapy regimen followed by autoHCT. Prior to the autologous transplant, 56% (32/62) patients were in CR, 10 (16%) were in PR and 15 patients (24%) had progressive disease (PD). The intention-to-treat analysis showed that forty-six out of the sixty-two (74%) of patients underwent autologous transplantation, whereas sixteen patients did not undergo transplant, mainly due to disease progression. After autoHCT, 89% (41/46) patients were in CR, with 5 patients (11%) in PR that died shortly after disease progression. At a median follow-up of 66 months, the estimated 12-year OS, DFS and event-free survival (EFS) of 34%, 55% and 30%, respectively. Due to inclusion of ALK-positive ALCL (31%), with overall better prognosis, these results should be interpreted with caution. Multivariate analysis showed that patients with CR before autoHCT had a statistically significant benefit in terms of OS and EFS. These findings suggest that achieving CR before autoHCT could offer a greater chance of long-term survival (9).

The GEL-TAMO Study Group published a phase II trial for front-line treatment for high-risk nodal PTCL with MegaCHOP (higher-dose CHOP) regimen and consolidation with autoHCT. Twenty-six patients were enrolled to the study, and after the first three courses of MegaCHOP, response was evaluated with computed tomography (CT) and gallium scans. In case of CR and negative gallium scan, patients were assigned to receive one to two additional courses of MegaCHOP and the conditioning regimen followed by autoHCT. The remaining patients were given salvage therapy with IFE (ifosfamide, etoposide) and those who achieved at least PR in re-evaluation would proceed to autoHCT. The remaining patients were considered as primary failure and were excluded from the protocol. After a median follow-up of 35 months after diagnosis, the 3-year OS and PFS was 73% and 53%, respectively. In addition, 73% (19/26) of patients who received autoHCT presented an estimated 2-year OS, PFS and DFS of 84%, 56% and 63%, respectively. The only variable in this study that showed significant impact on OS was the chemosensitive status, both after initial MegaCHOP and before transplant, with a 3-year OS of 83% for patients with complete or partial response after three courses of MegaCHOP, compared to 43% OS in patients with primary refractory disease. Even patients that were chemosensitive after IFE salvage therapy appeared to have an improvement in the estimated 3-year OS. Univariate analysis of prognostic score systems, like Prognostic Index for PTCL (PIT) and a-IPI, showed no difference in outcome with this treatment strategy. They concluded that frontline autoHCT might have overcome the poor prognosis determined by prognostic scoring systems. The approach of salvage therapy in high-risk aggressive nodal PTCL that does not achieve CR in initial treatment may improve outcomes following autoHCT (10).

Mercadal et al. studied 41 patients with newly diagnosed PTCL and submitted them to either MegaCHOP or ESHAP (etoposide, cisplatin, cytarabine and prednisone) regimen, with consolidation autoHCT if at least PR was reached. Seventeen patients failed therapy, with sixteen cases because of disease progression and one due to an early death by severe infection. Twenty-four patients (16 CR, 4 CR/unconfirmed (CRu) and 4 PR) were candidates for autoHCT. The authors discuss an important selection bias in studies aimed at evaluating the role of autoHCT, mainly because a significant proportion of patients do not respond to induction chemotherapy and therefore, are not submitted to transplantation. Within this study, among the 58% patients eligible for transplantation, only 41% eventually received a transplant out of the initial. These results might be due to a moderate CR rate obtained and the small sample size, and novel therapies and clinical trials should be encouraged to improve outcomes in PTCL therapy (11).

The Nordic Lymphoma Group (NLG) conducted one of the largest prospective phase II studies that addressed the role of up-front HDT and autoHCT in PTCL. With a total of 166 patients enrolled, treatment consisted of CHOEP-14 (CHOP with inclusion of etoposide biweekly), or CHOP-14 (CHOP biweekly) in patients aged over 60 years. Patients who achieved PR or CR were candidates for conditioning chemotherapy and autoHCT. Patients were evaluated for treatment response, and at the end of induction treatment, 82 of 156 patients achieved CRu and 49 PR, with an ORR of 82%. Twenty-five patients developed primary refractory disease. For various reasons, 16 patients were excluded before HDT and autoHCT, finally resulting in 115 patients who continued to transplantation. With a median follow-up of 60.5 months, the 5-year OS and PFS was 51% and 44%, respectively. Mortality due to treatment toxicity resulted in seven deaths, corresponding to a 4% TRM. ALCL ALK-positive, primary cutaneous, and primary leukemic subtypes were excluded. The highest OS and PFS after subtype-specific analysis were seen in patients with ALCL ALK-negative (5-year OS 70% and PFS 61%). The median 5-year follow-up allowed analysis of the time to disease relapse, with 11 patients (7%) developing relapsed disease after two years from transplantation, and therefore, provide rationale for a future maintenance therapy. Almost one fourth of patients (26%) failed treatment prior to transplant, reinforcing the need for better and novel therapies in the first-line treatment for PTCL. The results of this large cohort revealed encouraging outcomes and strengthened the recommendation of HDT and autoHCT for first-line therapy in PTCL patients (12).

The German study from Willhelm et al. reported a median 5-year follow-up study with 111 patients with newly diagnosed PTCL. Patients received conventional chemotherapy regimen with CHOP and if at least PR was reached, patients would proceed to myeloablative therapy and autoHCT. Treatment response revealed 91 (82%) ORR, with 69 patients (62%) achieving CR and 22 a PR (20%), with the remaining 20 patients (18%) failing primary treatment. Only 75 (68%) completed the entire study protocol, with disease progression/relapse as the main cause for not undergoing autoHCT. The estimated 5-year OS, DFS and PFS were 44%, 54% and 39% respectively. Considering patients who underwent autoHCT, OS rate was estimated at 57%, in comparison to 23% OS in those who did not. TRM rate resulted in 3.6%, similar to previous prospective studies. Long follow-up analysis showed that 43 (39%) patients achieved continuous remission, but early relapse and disease progression remained as major issues and demand novel treatment strategies to improve response rates in PTCL, allowing more patients to proceed with transplantation (13).

A registry study from Park et al. analyzed data from the prospective multicenter cohort study COMPLETE (Comprehensive Oncology Measures for Peripheral T-Cell Lymphoma Treatment), analyzed the impact of autoHCT on outcomes of patients with nodal PTCL in first-line complete remission. This was the first published study to report findings from prospective enrolled patients comparing clinical outcomes with or without autoHCT. Of the 119 patients with nodal PTCL who achieved CR1, 36 (30%) underwent autoHCT, while 83 (70%) were treated without autoHCT. Physician choice was the main reason for not considering transplantation, accounting for 55% of cases, which could serve a potential bias, as higher proportions of AITL histology subtype, advanced-stage disease (III/IV), and patients younger than 65 years were reported in the autoHCT group. ALCL subtype, which is associated with a favorable prognosis, was more frequently present in the non-autoHCT group. Overall, after a median follow-up of 2,8 years, there was no significant difference in survival between patients with CR1 that underwent and who did not undergo autoHCT. However, subgroup analysis suggested better outcomes in patients with AITL that proceeded to autoHCT compared to other nodal PTCL subtypes, with improved outcomes in OS in advanced-stage disease and intermediate to high IPI. The estimated 2-year OS and PFS rates for CR1 patients were 75.3% and 63.4%, respectively; while patients who did not achieve CR after first-line chemotherapy was 41.9% and 19.3%, respectively. Although with a notable selection bias limitation, these findings strengthen the importance of achieving CR1 in OS and serve as guidance for new and larger studies to determine the real benefit of autoHCT in PTCL (15).

Rarer entities such as hepatosplenic T-cell lymphoma (HSTL), AITL and EATL are underrepresented in clinical trials, making it difficult to find data regarding treatment approaches and outcomes in a real-world setting. As described in registry studies and other retrospective and prospective studies, consolidative autoHCT should be considered for AITL cases in first CR (7, 8, 10–13). The supporting evidence for utilizing autoHCT as a consolidation tactic is most compelling for EATL. One study conducted by the Scotland and Newcastle Lymphoma Group involved 26 patients who underwent a unique treatment regime that consisted of one round of CHOP followed by three rounds of IVE (ifosfamide, vincristine, etoposide), with alternating intermediate-dose methotrexate. Subsequently, autoHCT was administered if the patients were in remission. The 5-year PFS and OS rates were found to be 52% and 60% respectively, which displayed significant improvement when compared to the historical group treated with anthracycline-based chemotherapy (26). In a UK phase II study, 21 patients (including 11 with EATL) were assessed using the induction regimen applied by the Scotland and Newcastle Lymphoma Group. For EATL patients, both the 1-year OS and PFS rates were 45%. Among the five EATL patients who underwent autoHCT, only one experienced relapse (27). Furthermore, the already presented NLG study had 21 EATL patients enlisted, demonstrating a 5-year PFS rate of 38% and an OS rate of 48% for this specific group of patients (12). Supporting these findings, a retrospective analysis of the European Society for Bone and Marrow Transplantation (EBMT) registry and a retrospective cohort study both highlighted a survival advantage with autoHCT (28, 29). To date, from the limited data available for HSTL, it appears that the best therapeutic approach is non-CHOP induction therapy followed by consolidation with allo or auto (if limited donor availability) HCT (30, 31).

The SWOG S9704 trial designed by lymphoma committees of the United States and Canada was the first randomized trial to address consolidative autoHCT for high-risk patients with diffuse aggressive NHL. After patients received five cycles of induction chemotherapy with CHOP or R-CHOP (CHOP plus rituximab), they were randomized either to receive three more cycles of

chemotherapy (control group) versus one additional cycle followed by autoHCT with prior myeloablative radio or chemotherapy-based regimen. In a total of 370 eligible patients, 40 had an aggressive T-cell phenotype NHL. Of these, 28 (70%) patients were randomized after induction therapy, with 9 out of 12 patients excluded due to early disease progression. Thirteen were in the control arm and 15 in the transplantation arm, with three patients that did not undergo transplant due to patient refusal (2) or mobilization failure (1). At a median follow-up of 7.8 years after randomization, there was no statistical significance observed in the 5-year estimated OS (40% versus 45%) and PFS (40% versus 38%) for the transplant and control group, respectively. While results were discouraging for the first randomized trial, these findings should be analyzed with caution due to the small sample size and the retrospective analysis of a specific subgroup from the study (32).

Savage et al. reported a subgroup analysis of the role of HCT in CD30+ PTCL in the double-blind randomized phase III ECHELON-2 study after frontline Brentuximab vedotin (BV) plus cyclophosphamide doxorubicin and prednisone (CHP) versus CHOP regimen. From the BV plus CHP arm, 114 of 177 (64%) patients were in CR at the end of the treatment regimen. Thirty-eight of 114 (33%) underwent consolidative HCT (2 allo and 36 auto). With a median follow-up of 47.57 months, there was no difference in adverse event profile for those who did or did not undergo transplantation, with an estimated 3-year PFS of 80.4% vs 54.9%, respectively. The estimated 5-year PFS was 65.3% after transplant vs 46.4% without transplant. Of the CHOP arm, 97 of 177 (55%) were in CR at end of treatment. Twenty-nine out of 97 patients underwent consolidative transplant. At a median follow-up of 53.72 months, the estimated 3-year PFS was 67,2% in favor for consolidative transplant vs 54.1% in those who did not undergo transplant. The results for 5-year PFS were 48.9% and 40.9%, respectively. Even though the ECHELON-2 trial was not focused on transplantation, analysis of HCT seems to support a benefit in consolidative HCT for patients that received BV plus CHP, with less pronounced benefit in the CHOP arm. The low sample size of the study limits the statistical power and the overall impact of consolidative HCT (33).

The French Lymphoma Study Association (LYSA) and the German Lymphoma Alliance (GLA) designed a prospective, randomized, multicenter, phase III trial that evaluated the role of allogeneic HCT (alloHCT) against autoHCT in untreated patients with high-risk PTCL. Patients were randomized to either receive four courses of CHOEP-14, one course of DHAP (dexamethasone, cytosine-arabinoside, and cisplatin or carboplatin) and auto or alloHCT after conditioning regimen. From 103 patients in the intention-to-treat analysis, 54 were assigned to autoHCT and 49 to alloHCT. Thirty-four of 54 patients (63%) underwent autoHCT. Twenty patients were unable to proceed to transplant, 15 due to early progression. Twenty-six of 49 patients (53%) underwent alloHCT, while 14 patients did not undergo transplant due to early progression. Eight patients randomized to alloHCT had no compatible donor and were rescheduled to receive autoHCT. In total, 41 patients were consolidated with autologous HCT and 26 with alloHCT. At a median follow-up of 42 months, there were no significant differences between auto and alloHCT in OS (70% vs 57%), PFS (39% vs 43%) and EFS (38% vs 43%), respectively. TRM rate was 0% in the autoHCT setting, while eight deaths (31% TRM) were related to alloHCT. Almost a third of patients did not undergo transplant due to disease progression, supporting the rationale for more effective and novel therapies for T-cell lymphoma. The significant high TRM observed in first-line alloHCT is not acceptable in current treatment settings, and therefore, the recommendation for alloHCT should be mainly in specific histologic subtypes or patients with R/R disease, while autoHCT continues to be the preferred option for patients with newly diagnosed PTCL (34).

Given the conflicting data, small sample size, numerous histology subtypes and different treatment regimens, the impact of consolidation therapy with autologous HCT in patients with PTCL is controversial. Many retrospective and prospective studies tend to suggest benefits for autoHCT in up-front therapy. Current guidelines and expert opinion recommend this strategy as the main treatment choice in most of the more common subtypes, excluding ALCL ALK-positive, extranodal NK/T-cell lymphoma and Adult T-cell Leukemia/Lymphoma (ATLL) (2, 35). Treatment related toxicity is generally manageable, showing feasibility of chemotherapy and autoHCT in newly diagnosed patients. The predictive capability of prognostic scoring systems prior to transplant might be surpassed with this first-line treatment strategy, although still debatable. The value of achieving the best ORR, mainly CR, is crucial for improving clinical outcomes and OS and PFS, and therefore, up-front autologous HCT might benefit with a long-term disease remission in chemosensitive patients. With novel and promising therapies emerging, defining the real role for autologous HCT in PTCL frontline therapy is still challenging, and larger and transplant-focused randomized trials are required to establish it.

3 HCT in R/R PTCL

Unfortunately, disease refractoriness and relapse are common outcomes for patients with peripheral T-cell lymphoma. Data collected from patients enrolled in the International T-cell Project from 2006 to 2016 showed that out of the patients that received first line therapy, 32% reached and sustained CR and 68% were refractory or relapsed. Among those labeled as refractory/relapsed, 69% represented the refractory and 31% the relapsed (1). Median time from diagnosis to relapse ranges from 8 to 12.1 months (1, 36). Regarding survival rates, the COMPLETE Registry showed that the median OS were 29.1 months for relapsed patients and 12.3 months for refractory patients, which suggests that patients with chemosensitive disease have higher survival rates. They also found that 30% of this population had T-cell lymphomas with extra nodal involvement (36). In the International T-cell Project, after a median follow up of 38 months, 70% of the R/R patients had died, and the median survival time after relapse was only 5.8 months. Three-year OS were 21% and 28% for the refractory and relapsed, respectively. They also demonstrated that refractory disease was associated with higher risk of death, while later relapse (> 12 months) and salvage therapy with stem cell transplant were associated with better OS (1).

A retrospective study from Stanford University showed that disease status by the time of transplant had a great impact on OS and PFS. Fifty-three patients with PTCL underwent autoHCT in different stages of disease. Five-year PFS rates were 51%, 12% and 0 for patients in CR1/PR1, CR2/PR2 and primary refractory disease, respectively. Corresponding 5-year OS rates were 76%, 40% and 30% (16). Similarly, the GEL-TAMO presented data from 115 PTCL patients treated with autologous HCT in CR1, CR2+, PR1+ or refractory disease, and they also found higher survival rates in patients who were transplanted in CR1 in comparison to other groups. For patients in CR1, CR2+, PR1+ and refractory disease, the 5-year OS were 80%, 50%, 49% and 0, respectively. They also concluded that transplant in first line or chemosensitive disease, age < 41 years old, ECOG 0 or 1, absence of extranodal involvement, among other factors, are associated with higher survival (14). Both studies show that autoHCT performed in earlier stages of remission was associated with better outcomes, highlighting those patients with refractory disease had much worse performance, and those relapsed with chemosensitive disease were more likely to benefit from this strategy.

A systematic review and meta-analysis evaluated HDT and autoHCT for PTCL. They included 27 studies, in which 15 reported autoHCT in the R/R PTCL setting. PFS, OS, relapse/progression and TRM pooled rates were 36%, 47%, 51% and 10%, respectively. These data point to HDT/autoHCT as a reasonable strategy for R/R PTCL, given its 47% OS rate, but it also represented higher TRM when compared to HDT/autoHCT in first-line. An important discussion raised by these authors is the fact that most studies assess patients with relapsed or refractory disease combined, and therefore the outcomes for them seem to be the same, however, they believe that patients with refractory disease may not achieve the same outcomes than those that have relapsed disease, but have previously presented some chemosensitivity to salvage therapy, since this is a predictor of response to HDT/auto-HCT in other types of lymphoma (25).

Allogeneic HCT data for PTCL is mostly based on retrospective and prospective single-arm studies. However, the recommended treatment strategy in the R/R setting of non-ALK+ PTCL is salvage chemotherapy followed by HCT (autoHCT or alloHCT) (37). For patients with primary refractory PTCL, or PTCL that has relapsed after autoHCT or multiple prior lines of therapy, alloHCT provides the only potential curative therapy with survival rates of 40 to 50%. Due to the high risk of NRM, particularly with myeloablative conditioning in patients who have recently received an autograft or who have received extensive salvage chemotherapy, reduced intensity regimens are preferred (19–23, 38, 39).

AITL presents a rather unique scenario where alloHCT in the R/R setting appears to hold more promise compared to other nodal PTCLs. According to data from the CIBMTR, R/R AITL patients demonstrated a 4-year PFS and OS of 47% and 56%, respectively. Notably, relapse rates maintained a steady level at the 2-year mark post alloHCT, indicating sustained disease control even in patients who had experienced a failed prior autoHCT and those with refractory disease at the time of alloHCT (4-year PFS: 38%, OS: 52%) (40). Furthermore, the French registry data showed a favorable survival advantage for R/R AITL when compared to other histological subtypes (PTCL and ALCL), showing 5-year OS and EFS rates of 80% for R/R AITL. Despite the differences in survival rates, the univariate analysis did not reveal any statistical significance in OS, EFS, and TRM among the various histological subtypes (41).

A retrospective study from Huang et al. comparing auto and alloHCT for PTCL showed that primary refractory patients that underwent autoHCT had 3-year PFS of 20% and 3-year OS of 20%, while alloHCT provided rates of 49% and 53% for 3-year PFS and 3-year OS, respectively (17). Additionally, in a study aimed at evaluating salvage strategies after relapse in PTCL ALK-, all seven patients that underwent autoHCT at relapse died from disease progression, with median survival of 10 months (18). On the other hand, Smith et al. described a cohort of 241 patients from the Centre for International Blood and Marrow Transplant Research (CIBMTR) database that showed better OS and PFS in patients that underwent autoHCT in comparison to alloHCT, although there were differences in baseline characteristics (patients from the autologous group were more likely to be in CR1, have chemosensitive disease, ALCL subtype and fewer previous lines of therapy, which means they were of lower risk than the patients from the allogeneic group). Multivariate analysis did not show a difference between autologous and allogeneic in concern to relapse/progression, but NRM was higher in the allogeneic group and in patients with two or more lines of pretransplantation chemotherapy. Patients who underwent autoHCT in CR1 showed the highest survival rates. Moreover, patients presenting ALCL subtype had better survival rate (55% vs 35% PFS and 68% vs 41% OS) and reduced NRM in the autoHCT setting in comparison to alloHCT. This is the largest retrospective study regarding alloHCT in PTCL, with 126 patients undergoing this modality of transplantation, and they found that patients not in CR or after two or more chemotherapy regimens were at higher risk of overall mortality and treatment failure. Also, they described no impact of donor source, conditioning intensity, or graft-versus-host disease (GvHD) on relapse or survival (19).

A systematic review and meta-analysis published in 2021 was designed with the aim of comparing the efficacy and safety of auto and alloHCT in patients with R/R PTCL. Thirty studies were analyzed, comprising a total of 1765 patients. The rate of 3-year OS was relatively higher on the auto group (55% vs 50%, in comparison to allo), although CR rate prior to transplant was higher on patients who underwent autologous transplant, indicating that these patients were more chemosensitive. Therefore, when taking this enrollment bias into consideration, it is possible to speculate that alloHCT showed survival advantage in comparison to autoHCT. However, 3-year TRM was lower on the autologous group (7% vs 32%), showing that despite providing greater effectiveness, alloHCT may be riskier than autoHCT (42).

As presented in the articles reviewed, the evidence to support the use of autologous and allogeneic HCT in the R/R PTCL are limited and conflicting, meaning this is still a challenging scenario for physicians. Additional prospective trials and new therapeutic approaches, including cell therapy techniques, are sorely needed in this population.

4 Advanced cellular therapies for PTCL

CAR technology enables the cytotoxic immune cells to specifically recognize and target a surface antigen in a major histocompatibility complex (MHC)-independent manner. The CAR extracellular single-chain variable fragment (scFv) is specific for the target antigen and is linked by hinge and transmembrane regions to CD28 and/or 4-1BB co-activation domains and finally the CD3ζ intracellular signaling domain (43). Developing a safe and effective CAR-T cell therapy relies on identifying an ideal surface target antigen that is highly sensitive for the underlying malignancy and uniformly specific to avoid on-target off-tumor toxicities. Hematological malignancies are generally heterogeneous, and an optimal antigen that is exclusively expressed on all malignant cells with robust intensity is rarely found. Although not as studied as CD19+ leukemia and lymphomas and other hematological malignancies, the number of CAR-T cell therapy clinical trials for T cell malignancies have been increasing considerably in the past few years (Table 3).

CAR transgenes can be introduced into cells either transiently using mRNA electroporation or permanently using lentiviral, gammaretroviral or transposon-based gene delivery (44, 45). CAR-T products generated through viral vector transduction can lead to robust expansion and persistence *in vivo*, which heightens the risk of T-cell aplasia(46). Conversely, mRNA-engineered CAR-T cells have demonstrated similar anti-tumor activity but with limited persistence following administration (47, 48). This strategy shows promise for treating T-cell malignancies, but stable and sufficient tumoricidal activity may require sequential CAR-T administration or bridging to HCT. To address T-cell aplasia, equipping CAR-T products with safety switches (or suicide switches) may allow the control of transduced T-cells after infusion into patients (49).

Due to the similar biological structures and functions shared by B- and T-cells, chimeric antigen receptor (CAR) T-cell therapy was initially considered a natural approach for treating T-cell neoplasms. However, practical concerns such as fratricide and possibility of immunosuppression due to aplasia of normal T-cells have been raised. These concerns are further complicated by an immunosuppressive microenvironment that promotes the development and progression of T-cell malignancies, particularly TCLs (50, 51). Moreover, autologous T-cells harvested from patients with T-cell malignancies for CAR-T cell manufacture may be contaminated with malignant cells. To circumvent that issue, the use of allogeneic T-cells, Natural Killer (NK) cells, iNKT cells and macrophages are currently being explored. Indeed, several studies have evaluated these different cell types as allogeneic "off-the-shelf" products. In the following sections we present recent studies and discuss its advantages.

5 CAR-T cell for PTCL

Most targeted antigens for CAR-T products against T-cell malignancies, such as CD3, CD5, and CD7, are commonly expressed by healthy T-cells (52, 53). This shared expression makes it difficult to isolate healthy T-cells from patients with T-cell malignancies to engineer autologous CAR-T products, where normal and malignant T-cells might be collected during leukapheresis. A CAR-T construct targeting a tumor-associated antigen (TAA) expressed by different populations of T-cells may present on-target off-tumor effect, attacking and destroying malignant and normal T-cells, as well as other CAR T-cells, leading to disruption of CAR-T cells' expansion, persistence, and tumoricidal function. Such fratricide could result in T cell dysfunction before or after exposure, leading to resistance to CAR-T therapy or disease relapse (54, 55). These observations prompted several groups to develop various products and strategies, including targeting more restricted T-cell antigens, such as CD4, CD30, CD37, and CCR4. Selecting the appropriate target and considering potential adverse events remains a challenge in CAR-T therapy for T-cell malignancies. Other alternative antigens, such as the myeloid markers CD13 and CD33, are emerging as possible targets due to their aberrant expression on precursor T-cell leukemia, which could indicate a worse disease prognosis (56). A quick search in the ClinicalTrials.gov repository using the terms "Peripheral T cell lymphoma", "PTCL", "T cell Lymphoma", "CAR" and "chimeric antigen receptor" returned 40 studies of which 39 are CAR-T cell studies and only one is a CAR-NK clinical trial. It is possible to note from these studies that several T cell membrane markers are being investigated as possible targets to CAR-T cell therapy for PTCL (Figure 1). The most applied target is CD7, a protein of the immunoglobulin superfamily expressed both in mature T cells and thymocytes. Currently, eight different markers are being tested as targets for CAR therapy for PTCL: CD4, CD5, CD7, CD30, CD37, CD70, CD147 and TRBC1 (Table 3).

Even though malignant T-cells significantly decrease the expression of T cell receptor (TCR), this important receptor is still expressed in about 30% of T-ALLs and the vast majority of PTCLs (57). Since a normal population of T cells expresses both TRBC1 and TRBC2, and a malignant subset expresses only one, targeting either receptor individually could have antitumor effects while preserving a substantial portion of normal T-cells. *In vitro* studies on the generation of TRBC1CAR-T have demonstrated the ability to leave TRBC2+ cells untouched. In addition, treatment with these cells was associated with a significant decrease in tumor burden and prolonged survival in comparison to the control group (58). This led to the use of CARs specific for the TCR beta chain constant regions TCRB1 or TRCB2, which is being evaluated in a phase I/II clinical trial for T-non-Hodgkin lymphomas (NCT03590574). Malignant Sezary cells express a unique TCR gene rearrangement, which could be a good tumor-specific antigen to be targeted. However, this unique TCR seems to vary from patient to patient, making TCR targeting a complex approach (59). Certain highly expressed cell surface molecules such as CCR4, programmed cell death protein 1 (PD-1), and CD47 could provide a common targeting approach. CD47 is significantly increased on Sezary cells and inhibits phagocytosis by macrophages (60). Studies showed that targeting of universal T cell antigens such as CD3 and CD7 using CAR-T cells leads to fratricide of CAR-T cells. Other tumor-restricted antigens targeted with CAR-T cells include CCR4,

CD4, and CD30. PTCLs are frequently characterized by the presence of CD4+ cells, with consistent and elevated expression levels of CD4 making it an optimal candidate for targeted therapy using CAR technology (61). Targeting of CD4 led to fratricide of CD4+ CAR-T cells, but the remaining CD8+ CAR-T cells may have therapeutic potential (62).

In 2021, Pan et al. (63) reported the first-in-human Phase I trial to evaluate the safety and efficacy of donor-derived CD7 CAR T-cell

TABLE 3 Registered clinical trials using CAR-T/NK cells to treat PTCL patients.

Target	Number of Studies	Source	Trial Number	Status	Phase	N	Start Year
CD4	5	Auto (5)	NCT04162340	Recruiting	1	12	2019
			NCT03829540	Active, not recruiting	1	20	2019
			NCT04712864	Active, not recruiting	1	50	2021
			NCT04973527	Terminated	1	9	2021
			NCT04219319	Terminated	1	4	2021
CD5	4	Auto (3)	NCT03081910	Recruiting	1	42	2017
			NCT04767308	Not yet recruiting	Early 1	18	2021
			NCT05138458	Recruiting	1/2	40	2021
		Unknown (1)	NCT04594135	Recruiting	1	20	2020
CD7	17	Allo (6)	NCT02742727	Recruiting	1	21	2016
			NCT04538599	Completed	1	12	2020
			NCT05127135	Recruiting	1	24	2020
			NCT04689659	Recruiting	2	50	2021
			NCT04264078	Recruiting	2	20	2021
			NCT05377827	Recruiting	1	20	2022
		Auto (5)	NCT04004637	Not yet recruiting	1	48	2019
			NCT04480788	Recruiting	1	9	2020
			NCT05059912	Recruiting	Early 1	30	2021
			NCT04840875	Recruiting	1	30	2021
			NCT03690011	Recruiting	1	20	2021
		Unknown (6)	NCT04572308	Completed	1	20	2020
			NCT04934774	Recruiting	1	4	2020
			NCT04823091	Recruiting	1	20	2021
			NCT05620680	Unknown status	1/2	10	2022
			ISRCTN15323014	Recruiting	1	10	2022
			NCT05290155	Unknown status	1	10	2022
CD30	9	Auto (7)	NCT03049449	Completed (Has results)	1	26	2017
			NCT03602157	Recruiting	1	59	2018
			NCT04008394	Recruiting	1	50	2019
			NCT04083495	Recruiting	2	20	2019
			NCT04653649	Recruiting	1/2	30	2020
			NCT04526834	Active, not recruiting	1	21	2021
			NCT05208853	Not yet recruiting	Early 1	9	2022
		Allo (2)	NCT04288726	Recruiting	1	18	2020
			NCT04952584	Not yet recruiting	1	18	2023

(Continued)

TABLE 3 Continued

Target	Number of Studies	Source	Trial Number	Status	Phase	N	Start Year
CD37	1	Auto	NCT04136275	Recruiting	1	18	2020
CD70	1	Allo	NCT04502446	Recruiting	1	45	2020
CD147	1	Unknown	NCT05013372	Not yet recruiting	Early 1	12	2023
TRBC1	2	Auto (1)	NCT03590574	Recruiting	1/2	200	2018
		Unknown (1)	NCT04828174	Recruiting	1	9	2021

therapy for patients with R/R T-ALL. Twenty patients diagnosed with CD7-positive r/r T-ALL were enrolled. The median age of the participants was 11 years (range, 2-43), and they had received at least two previous therapies. To address the issue of CD7 CAR T-cell fratricide, the researchers designed a CAR construct using IntraBlock technology, which effectively prevented CD7 cell surface expression. To overcome the challenge of low quantity and quality of patients' autologous T cells, CAR-T cells were manufactured using T cells harvested from previous SCT donors and new donors. Most patients experienced Grade 1-2 CRS, with only 10% of patients experiencing Grade 3 CRS. Neurotoxicities associated with the treatment were mild and self-limiting. The treatment demonstrated a high response rate, with 95% of the participants responding to the therapy. Furthermore, 90% of the patients achieved complete remission, including 85% who achieved MRD-negative CR by day 15. Within 15 days post-infusion, the therapy led to a rapid depletion of CD7-positive normal lymphocytes, including T cells. However, CD7-negative T cells dramatically increased in all patients, indicating a gradual recovery of total T cells and NK cells. These T cells also displayed lower T-cell receptor diversity. Nevertheless, they demonstrated the ability to react to fungal and viral stimulations, producing interferon-gamma, suggesting potential immune-protecting functions. The trial demonstrated an acceptable safety profile, with most adverse events occurring within 30 days post-infusion. Early GVHD was observed in 60% of the participants but was low grade and manageable. CAR T cells efficiently proliferated and persisted in patients without evidence of rejection, which may be attributed to complete chimerism in those who received donor-derived cells after previous stem cell transplantation. A multi-center, phase II study is currently in progress to investigate the use of donor-derived CD7 CAR-T cells in patients with R/R T-cell malignancies (63).

More recently, Tan et al. reported the outcomes of the same cohort described in 63, after a median follow-up time of 27 months post-treatment. Non-relapse mortality occurred in 25% of the 20 patients, with a median time of 6.8 months after treatment. The causes of non-relapse mortality included infections in patients without SCT consolidation and engraftment syndrome in a patient after SCT consolidation. Of the 19 patients who responded, seven proceeded to SCT consolidation, two withdrew to take alternative therapies, and ten did not receive further therapy. Among the ten patients who did not receive further treatment, three were in remission, three relapsed (including one with CD7-positive

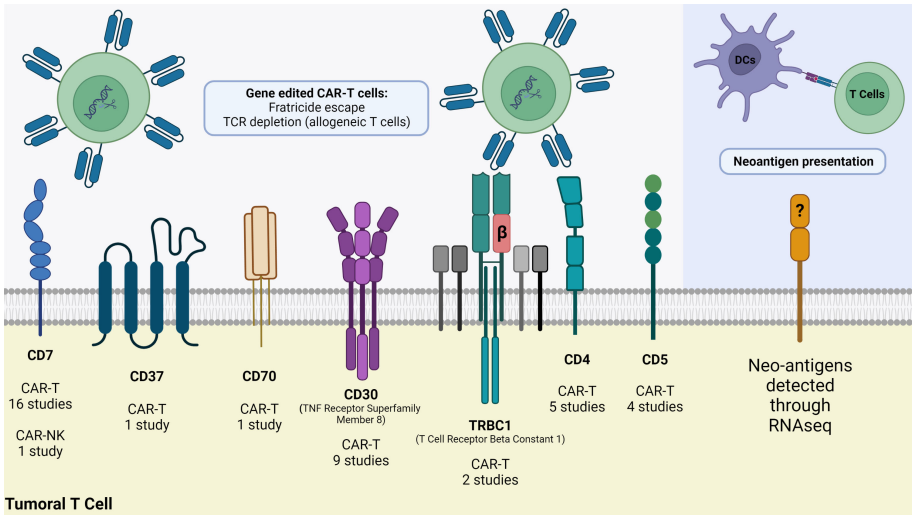


FIGURE 1
Strategies to fight PTCL using advanced cellular therapies. At least seven T cell membrane markers are being currently targeted in clinical trials, but CAR-T cells demand gene editing whether to avoid fratricide (CAR-T/NK cells self-destruction) or Graft-versus-Host-Disease (GvHD) when using allogeneic T cells. As an alternative strategy, RNA-seq of malignant T cells reveal neoantigens that can be synthesized and delivered to dendritic cells for presentation through MHC molecules to T cells. Image was created using Biorender.

extramedullary disease), and four died of infection. Among the seven patients who underwent SCT consolidation, three maintained in remission, three relapsed, and one patient died of transplant-related complications. The relapse rate among responders was 33.3%, with a median time to relapse of 6 months post-infusion. The median PFS and duration of response (DOR) were 11.0 months and 10.5 months, respectively, among the 19 responders. The median OS was 18.3 months. The 2-year PFS rate for responders and OS rate for all 20 treated patients were 36.8% and 42.3%, respectively. Subgroup analysis revealed that patients without SCT consolidation had a 2-year PFS rate of 31.8% and an OS rate of 35%, with a median PFS and OS of 11.0 months and 18.3 months, respectively. In contrast, patients with SCT consolidation had a 2-year PFS rate of 42.9% and an OS rate of 58%, with a median PFS of 9.1 months. The study did not reach the median OS for patients who received SCT consolidation. Long-term monitoring of T-cell phenotype and function showed that CD7+ T and NK cells remained undetectable in all patients until the last visit, except for one patient who experienced recovery of CD7+ T and NK cells 25.6 months post-infusion following the loss of flow-cytometry-detectable CD7 CAR T cells at 22.7 months. The number of CD7 – T, total T, and NK cells progressively increased, with total T-cell counts recovering to normal levels in 7 out of 12 patients at a median time of 1.9 months. Furthermore, long-term monitoring of T-cell phenotype and function in two patients revealed that the central memory T-cell subpopulation gradually increased, while low levels of naive and stem-cell memory T-cell subpopulations were detectable in one patient after 15 months. TCR diversity in patients after CD7 CAR T-cell infusion remained lower compared to healthy donors. The study identified late-onset GVHD as the most common long-term adverse event, with an incidence of 58% among the 12 patients without SCT consolidation. The authors suggest that early bridging to SCT consolidation may reduce the risk of severe infections, which were observed in a lower incidence (14.3%) in patients with SCT consolidation after CAR T-cell infusion. Relapse analysis revealed that among the patients who relapsed, four had CD7-negative relapses, and two had CD7-positive relapse. Frameshift or missense mutations were detected in the four CD7-negative relapse patients, suggesting that mutations may be a main cause of CD7 loss in tumor cells. The study also observed CD7-positive relapse following the loss of CAR T cells in a patient without SCT consolidation, suggesting that insufficient persistence of CAR T cells may contribute to relapse. The study acknowledges several limitations, including its phase I trial design with a small sample size. Consequently, the authors emphasize the need for larger phase II studies to confirm the safety, efficacy, and prognostic factors associated with CD7 CAR T-cell therapy. Despite these limitations, the study's strength lies in its provision of the first long-term follow-up in patients with T-ALL after CAR T-cell treatment. The durable responses observed, along with new signals of long-term adverse events, support the feasibility of using donor-derived CD7 CAR T cells as salvage treatment for children or adults with R/R T-ALL. Notably, severe infection emerged as a significant side effect associated with this therapy, underscoring the importance of early SCT consolidation and vigilant monitoring and prevention of infections for patients for whom subsequent SCT is not feasible (64).

Zhang et al. have recently presented the outcomes of a single-center phase I study evaluating autologous and allogeneic anti-CD7 CAR-T cell therapy in patients with R/R T-cell malignancies. The trial enrolled 11 patients aged 16 to 69, with CD7+ T-cell malignancies, including adult T-ALL, T-cell lymphoblastic lymphoma, angioimmunoblastic T-cell lymphoma, and mycosis fungoides. Of these patients, 10 received treatment, after one patient achieved complete remission prior to CAR-T cell infusion. The median lines of previous therapies were 4. CAR-T cells were produced using PBMCs from either patients or donors. The authors observed that 70% of patients achieved CR with mild CRS and no ICANS. Patients who received allogeneic CAR-T cells did not experience severe CRS, ICANS, or GVHD. In addition to these common complications, hematological toxicities, hemophagocytic lymphohistiocytosis (HLH), infections, and T-cell aplasia were also observed. The choice of allogeneic CAR-T cells for patients with highly aggressive and rapidly progressing malignancies eliminated the need for a one-month drug elution period before leukapheresis, reducing the likelihood of rapid disease progression. The CR rate was 80% for patients receiving allogeneic cells compared to 40% for those with autologous products. Notably, the relapse rate showed significant differences, with only one patient (25%) experiencing CD7- recurrence after allogeneic CAR-T cell therapy, while all patients treated with autologous CAR-T cells relapsed, either in the bone marrow or as extramedullary disease. Some patients experienced CD7+ recurrence despite the absence of detectable CAR copies *in vivo*. The study highlighted that the lower persistence of autologous CAR-T cells may contribute to treatment failure. Allogeneic CAR-T cells demonstrated stable survival in 75% of patients at month 2, whereas only 33% of patients receiving autologous cells showed persistent CAR-T cell presence. Two patients with Epstein-Barr virus (EBV) activation died of pneumonia during the study, emphasizing the need for caution when enrolling patients with a history of EBV infection and the importance of close monitoring. Limitations of the study include its non-randomized controlled trial design due to patient conditions and the small sample size. The authors emphasized the need for extended follow-up and larger studies to further evaluate long-term outcomes of anti-CD7 CAR-T cell therapies in T-cell malignancies (65).

Using a different approach to avoid fratricide during anti-CD7 CAR-T manufacture, Lu et al. showed the development of naturally selected CD7 CAR T cells (NS7CAR) for the treatment of CD7+ T-cell malignancies, focusing on T-ALL/LBL. The authors employed lentiviral transduction and selection techniques to generate NS7CAR T cells that retain CD7 expression while avoiding self-targeting and fratricide. A first-in-human phase 1 clinical study was conducted, utilizing NS7CAR T cells derived from patients with treatment-refractory T-ALL/LBL or their hematopoietic stem cell donors. Twenty patients, aged 3 to 47 years, received NS7CAR T cell therapy. Most patients experienced mild CRS, with only one patient developing grade 3 CRS, and neurotoxicity was minimal. At the 28-day evaluation, 19 patients achieved measurable residual disease-negative complete remission or incomplete CR, including responses

in patients with extramedullary disease. Subsequent transplantation was performed in most patients, and no relapses were documented during a median follow-up of 142.5 days. Importantly, NS7CAR T cell therapy resulted in rapid ablation of circulating CD7+ T cells and natural killer (NK) cells, which were replenished by CD7-negative subsets, preventing prolonged T-cell and NK-cell aplasia. The study concluded that NS7CAR T cells are well-tolerated and effective against CD7+ T-cell malignancies. Importantly, the generation of naturally fratricide-resistant CD7 CAR-T cells simplifies the manufacturing process and enhances the cost-effectiveness of this therapy (66).

6 "Universal" CAR-T cell for T-cell malignancies

Gene-editing methods have been gaining prominence in the cellular therapy field, such as the transcription activator-like effector nuclease (TALEN), which is being used to disrupt the CD3/TCR complex and prevent the expression of endogenous TCR in T-cells. Once TCR is inhibited, the cells are then modified to express CD3ϵ-targeting CARs. This approach has shown impressive results, with specific and significant antitumor activity against pediatric T-ALL samples demonstrated in preclinical models with the CD3+ Jurkat cell line (67). In a separate study, gene editing technologies were employed to simultaneously remove the expression of one of the TRBC genes, resulting in the elimination of endogenous TCR from the cell surface, indicating that this strategy could be used to prevent fratricide when producing autologous TRBCCAR-T (68). Other gene-editing techniques, such as CRISPR-Cas9, and Zinc-finger nucleases (ZFN), have been investigated for their potential to develop off-the-shelf CAR-T products for T-cell neoplasms. CRISPR-Cas9 has also been used to knock-out CD5 in T cells before embedding the CAR transgene into primary patient cells and Jurkat cells (69). This approach has resulted in limited fratricide and subsequent CAR persistence. CRISPR-Cas9 genome editing to disrupt the CD7 expression and engineer CAR-T cells lacking CD7 and TCR alpha chain (TRAC) expression have demonstrated significant antitumor activity against T-ALL cell lines and primary human samples, as well as tumor regression in preclinical models with the absence of GvHD (55).

Hu et al. presented the results of a phase I clinical study investigating the safety, efficacy, and pharmacokinetics of genetically modified CD7-targeting allogeneic CAR-T cell therapy (RD13-01) in patients with R/R CD7-positive hematological malignancies. The RD13-01 CAR-T cells were designed to enhance persistence and potency by genetically depleting CD7, TCR, and HLA class II, and incorporating an NK cell inhibitor (NKi) and the common cytokine receptor γ chain (γc). Preclinical assessments demonstrated potent antitumor activity of RD13-01 CAR-T cells. In the clinical trial, RD13-01 CAR-T cells were manufactured from allogeneic healthy donor PBMCs, and residual TCR/CD3+ T cells were removed to minimize GvHD. Twelve eligible patients with relapsed or refractory hematological malignancies were enrolled, including T-ALL, T-cell lymphoma, and acute myeloid leukemia (AML) cases. No dose-limiting toxicity, GvHD, or ICANS occurred during the trial. Grade 1-2 CRS was observed in 10 patients, with no severe CRS (grade ≥3) reported. Among the 11 patients evaluated for efficacy, 82% achieved an objective response, with 64% achieving complete remission or CR with incomplete hematological recovery at day 28 post-infusion. At a median follow-up of 10.5 months, four responders remained in CR, while one patient underwent salvage HCT and remained in CR. However, relapse or disease progression occurred in three leukemia and one lymphoma patient at a median time of 82 days after infusion. The expansion of CD8+CD7− T cells, which may recognize the HLA antigen of infused CAR-T cells, was associated with decreased CAR-T cell numbers and antigen-positive relapse. On the other hand, however, as 63 have shown, CD7− T cells generated from CD7-depleted hematopoietic stem cells may contribute to maintaining T cell function and controlling infections. Strategies to prolong CAR-T cell persistence while preserving CD7− normal T cell expansion, such as the expression of inhibitory ligands (e.g., PD-L1) on allogeneic CAR-T cells, warrant further investigation. Additionally, the authors report inter-patient variability in clinical responses following infusion of cells manufactured from the same batch, suggesting that endogenous factors and recipient immune landscape may influence therapeutic outcomes in the context of a universal CAR-T therapy (70).

In a more recent work, 71 demonstrated the use of base editing, mediated by CRISPR technology, for precise DNA modifications without inducing double-stranded DNA breaks, to generate base-edited allogeneic CAR7 (BE-CAR7) T cells for a phase I feasibility and safety trial in pediatric patients with R/R T-ALL. Healthy donor T cells were electroporated with specific single-guiding RNAs (sgRNAs) and a codon-optimized cytidine base editor (coBE) mRNA to target TRBC1, TRBC2, CD7, and CD52 genes. The edited T cells were then transduced with a lentivirus vector encoding a CAR targeting CD7. The trial aims to recruit 10 children in the United Kingdom for an initial cohort. The initial report covers data from lympho-depletion to day 28 after CAR-T cell infusion for three patients. Patients in molecular remission at day 28 underwent allogeneic stem-cell transplantation, depleting any persisting BE-CAR7 cells through the conditioning regimen before the transplant. Molecular analysis confirmed the precise editing of targeted cytosine positions in TRBC, CD7, and CD52. Karyotyping showed normal karyotypes, and PCR assays had negative results for translocations. The 28-day treatment period led to significant antileukemic responses and deep remission in two of the three patients. Cytokine release syndrome (CRS), fever, rash, and multilineage cytopenia were observed in all patients, with infectious complications managed with antiviral medications. The authors acknowledge the substantial immunosuppressive and cytopenic effects of the protocol and the risks associated with immune-cell manipulation. Subsequent allogeneic transplantation was performed to ensure immune reconstitution and limit the persistence of engineered cells (71).

7 CAR-NK cell for PTCL

Prior pre-clinical investigations have employed CAR-modified primary human natural killer (NK) cells against CD19, CD20, CD244, and HER2 for both hematological and solid tumors (72–75). Additionally, clinical trials have demonstrated successful application of anti-CD19 CAR-modified umbilical cord blood-derived and haploidentical NK cells in patients with CD19 lymphoid tumors and acute myeloid leukemia, respectively (76, 77). The use of CAR-modified NK cells can potentially eliminate the risk of fratricide, T-cell aplasia, and GvHD associated with CAR-T therapy (78). It may also remove the need for an inducible safety switch, as CAR-modified NK cells are eliminated shortly after administration (79). In contrast to CAR T-cells, CAR NK cells offer the benefit of engaging tumor cells through diverse mechanisms, while exhibiting a relatively reduced production of pro-inflammatory cytokines (80, 81).

The human NK cell line NK-92 has been utilized in multiple clinical studies for both hematologic malignancies and solid tumors, as well as in pre-clinical CAR applications (82–84). Given its versatility in both clonal NK cells and autologous/allogeneic NK cell immunotherapy, NK-92 serves as a valuable model. A third-generation CD5-CAR incorporating NK-92 cell lines, which do not express CD5 on their surface, showed selective and significant tumoricidal activity towards various T-cell lines, including Jurkat, CCRF-CEM, and MOLT-4, as well as against primary CD5+ cells from human T-ALL and PTCL samples (85). To mitigate the risk of T-cell aplasia and related infections, researchers engineered a CD4-redirected CAR-NK using the NK-92 cell line (61). Ex vivo experiments have shown that CD4CAR NK-92 cells possess potent anti-tumor cytotoxicity against various adult and pediatric CD4+ lymphoma/leukemia cell lines, as well as primary CD4+ T-cell malignancies from both adult and pediatric patients. In xenogeneic mouse models, CD4CAR NK-92 cells also demonstrated strong *in vivo* anti-CD4 activity. Notably, CD4CAR NK-92 cells did not affect the CFU capacity of CD34+ cord blood granulocyte/macrophage or erythroid cells in ex vivo assays, indicating that they do not compromise the hematopoietic stem cell and progenitor compartment. This promising approach suggests that CD4CAR NK cells could be utilized as part of a bridge-to-transplant strategy or as a stand-alone curative treatment for patients who are not eligible for HCT. Moreover, CD3CAR transduced NK-92 cells showed significant dose-dependent *in vitro* and *in vivo* cytotoxic against CD3-expressing PTCL samples and several T-ALL cell lines, with prolonged survival in preclinical models engrafted with the Jurkat cell line (86). In a recent study, CAR-NKs with the 2B4 and 4-1BB costimulatory domains demonstrated similar selective tumoricidal activity *in vitro*, while CAR-NKs with the 2B4 co-stimulatory domain showed an improved antileukemic activity in T-ALL preclinical models (87).

8 Future perspectives

CAR-T cell therapy has faced challenges in achieving optimal trafficking to challenging tumor sites, such as the skin. This is thought to be due to poor infiltration of such areas by αβ T cell subsets. γδ T cells, which constitute a smaller population of circulating lymphocytes (1-5%), are present in the skin, intestine, and reproductive organs and express chemokine receptors that attract them to these inaccessible tumor locations (88, 89). Additionally, γδ T cells can proliferate ex vivo and do not induce GvHD due to MHC-independent activation of their TCR. Therefore, γδ T cells could be considered as potential alternative effectors for allogeneic CAR-T therapy in T-cell malignancies, following thorough evaluation in studies for other malignancies (90, 91). Multi-virus-specific T (VST) cells have also been utilized as effector cells for CAR expression. Such cells can be genetically engineered to lack CAR target antigen expression and be fratricide-resistant, offering potential antiviral activity in the case of T-cell aplasia (92, 93). Studies have shown that allogeneic VST cells with HLA alloreactivity do not cause GvHD in humans, suggesting they may provide an alternative for producing off-the-shelf CAR-Ts (94).

The potential of CAR-iNKT (Invariant Natural Killer T) cell therapy as an "off-the-shelf" allogeneic immunotherapy for the treatment of T cell lymphoma is also being investigated. iNKT cells are a rare subset of T cells with innate and adaptive immune features (95). They express an invariant TCRVa24Ja18 chain that pairs with diverse TCRVb11 chains (96). By targeting specific TCRVb chains associated with ATL/TCL, CAR-iNKT cells can be engineered for effective and precise immunotherapy. In a very elegant pre-clinical study, (97) generated lentiviral CAR constructs to target TCRVb1, Vb2, Vb9, and Vb11 expressed on T cells. CAR-iNKT cells engineered with these constructs demonstrated potent anti-tumor activity against primary ATL cancer cells. TCRVb2 CAR-iNKT cells significantly inhibited tumor growth without causing adverse effects such as weight loss or signs of acute GVHD in animal models. The findings suggest that anti-TCRVb CAR-iNKT cells can offer both autologous and allogeneic treatment options as "off-the-shelf" cellular immunotherapy for T cell malignancies. Early clinical experience with allogeneic CAR-iNKT cells against B cell lymphoma have already indicated minimal toxicity and aGVHD (98). Overall, this study provides a promising rationale for the clinical development of anti-TCRVb CAR-iNKT cells as an effective and highly selective immunotherapy for currently incurable T cell lymphomas.

Another interesting approach to target PTCL that has already been evaluated in clinical studies is based on the use of neoantigen-activated haploidentical T cell therapy (NAHTC) (99). Whole-exome sequencing was used to identify non-synonymous mutations in matched tumor and normal cells, as well as tumor RNA sequencing to identify neoantigen candidate epitope sequences. The predicted binding affinity of peptides to individual HLA molecules was then determined, and synthetic RNAs encoding potential neo-epitopes were designed based on the selected mutations. Haploidentical donor's monocytes were isolated and differentiated into mature dendritic cells (DCs) that were electroporated with selected synthetic RNA and then co-cultured with haploidentical T cells. Preliminary findings demonstrate that the NAHTC regimen was well-tolerated, with no incidence of CRS or GvHD in the treated patients. Of the five PTCL patients evaluated, four (80%) achieved a complete response, and three of

the four CRs were sustained until the last analysis. Thus, treatment with NAHTC was safe and resulted in durable clinical responses. Despite the small sample size and preliminary nature of these findings, single-dose NAHTC therapy demonstrated superior activity compared to recently FDA-approved drugs for R/R PTCL (100–102). The effectiveness of NAHTC therapy for PTCL may stem from its distinct mechanism of action. Tumor neoantigens represent *de novo* epitopes derived from somatic mutations and are therefore tumor-specific and highly immunogenic, as they lack central tolerance. NAHTC cells activated by multiple patient-specific neoantigens *in vitro* are likely to target a diverse array of malignant clones within each patient, with the potential to address tumor heterogeneity, reducing the likelihood of tumor escape by single neoantigen loss (103, 104). The use of T cells from healthy donors in haploidentical T-cell therapy ensures their manufacture quality and quantity, making it a simple, safe, and reliable process that saves time and reduces costs, ensuring the accessibility of this treatment to more patients.

9 Conclusion

Autologous HCT has been the core of consolidation therapy in most of the more common PTCL subtypes for chemoresponsive patients (2, 35). Only recently, after the first randomized phase III ECHELON-2 study, there is evidence of improvement in PFS and OS in comparison to standard induction regimen CHOP (105). The expression of ALK in ALCL correlates with a favorable prognosis compared with other histological subtypes of PTCL, therefore, autoHCT may be considered in high-risk IPI patients or in second-line therapy. On the other hand, acute and lymphomatous subtypes of ATLL have a poor prognosis, with recommendation of alloHCT even in first-line therapy (2, 35). Overall, there is limited data and a lack of randomized trials for PTCL treatment, mainly due to the low incidence and heterogeneity of histology subtypes, and therefore, current treatments rely mostly on phase II trials, retrospective studies, and expert opinion (106). Overall, it is possible to conclude that HCT is a reasonable strategy for PTCL patients in the R/R context, but it seems that main factors to take into consideration may be the type of transplant (auto/allo) and the stage of the disease to be performed. Currently, HCT is the only potentially curative therapy for patients with R/R PTCL, and the evidence for the role of auto and alloHCT for this population has only been evaluated in registry data and retrospective studies (4). It is noteworthy that conclusions were not homogeneous among authors. In general, it might be safe to assume that autoHCT often results in lower durable benefit for patients with R/R disease in comparison to alloHCT, however, for patients with ALCL subtype and chemosensitive disease, autoHCT may provide survival benefit. These findings corroborate with the current recommendations from the American Society for Blood and Marrow Transplantation, that recommends autoHCT for nodal PTCL subtypes with relapsed chemosensitive disease, if it has not been done as upfront consolidation, or alloHCT when an autologous transplantation was made in first line (35). CAR-T cell therapy is currently being considered as a possible approach for the treatment of PTCL, but challenges like fratricide, failure in manufacture and proper antigen targeting still need to be addressed. Clinical studies that use allogeneic anti-CD7 CAR-T have shown promising results, indicating it as an effective "bridge-to-transplant" strategy for patients with available hematopoietic cell donors. Novel approaches such as the use of other immune cells, like NK and myeloid cells equipped with a CAR as "off-the-shelf" products are being studied, as well as the modification of donor derived T cells through gene editing techniques (107). With the rapid evolving knowledge of PTCL molecular and pathogenic properties, we will be able to develop efficient and personalized therapies for the treatment of these hard-to-treat diseases.

Author contributions

SC, AK, CA, TO, PK and VR reviewed the literature, organized, and wrote the manuscript. All authors contributed to the article and approved the submitted version.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Bellei M, Foss FM, Shustov AR, Horwitz SM, Marcheselli L, Kim WS, et al. The outcome of peripheral T-cell lymphoma patients failing first-line therapy: a report from the prospective, International T-Cell Project. *Hematologica* (2018) 103:1191–7. doi: 10.3324/haematol.2017.186577

2. Horwitz SM, Ansell S, Ai WZ, Barnes J, Barta SK, Brammer J, et al. T-cell lymphomas, version 2.2022, NCCN clinical practice guidelines in oncology. *J Natl Compr Cancer Netw* (2022) 20:285–308. doi: 10.6004/jnccn.2022.0015

3. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IB, de O, et al. The 5th edition of the world health organization classification of hematolymphoid tumors: lymphoid neoplasms. *Leukemia* (2022) 36:1720–48. doi: 10.1038/s41375-022-01620-2

4. Rogers AM, Brammer JE. Hematopoietic cell transplantation and adoptive cell therapy in peripheral T cell lymphoma. *Curr Hematol Malig Rep* (2020) 15:316–32. doi: 10.1007/s11899-020-00590-5

5. Vose J., Armitage J., Weisenburger D. International peripheral T-cell and natural killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26:4124–30. doi: 10.1200/JCO.2008.16.4558

6. Jantunen E, Wiklund T, Juvonen E, Putkonen M, Lehtinen T, Kuittinen O, et al. Autologous stem cell transplantation in adult patients with peripheral T-cell lymphoma: a nation-wide survey. *Bone Marrow Transplant* (2004) 33:405–10. doi: 10.1038/sj.bmt.1704367

7. Yam C, Landsburg DJ, Nead KT, Lin X, Mato AR, Svoboda J, et al. Autologous stem cell transplantation in first complete remission may not extend progression-free survival in patients with peripheral T cell lymphomas. *Am J Hematol* (2016) 91:672–6. doi: 10.1002/ajh.24372

8. Reimer P, Rüdiger T, Geissinger E, Weissinger F, Nerl C, Schmitz N, et al. Autologous stem-cell transplantation as first-line therapy in peripheral t-cell lymphomas: Results of a prospective multicenter study. *J Clin Oncol* (2009) 27:106–13. doi: 10.1200/JCO.2008.17.4870

9. Corradini P, Tarella C, Zallio F, Dodero A, Zanni M, Valagussa P, et al. Long-term follow-up of patients with peripheral T-cell lymphomas treated up-front with high-dose chemotherapy followed by autologous stem cell transplantation. *Leukemia* (2006) 20:1533–8. doi: 10.1038/sj.leu.2404306

10. Rodríguez J, Conde E, Gutiérrez A, Arranz R, León Á., Marín J, et al. Frontline autologous stem cell transplantation in high-risk peripheral T-cell lymphoma: a prospective study from The Gel-Tamo Study Group. *Eur J Haematol* (2007) 79:32–8. doi: 10.1111/j.1600-0609.2007.00856.x

11. Mercadal S, Briones J, Xicoy B, Pedro C, Escoda L, Estany C, et al. Intensive chemotherapy (high-dose CHOP/ESHAP regimen) followed by autologous stem-cell transplantation in previously untreated patients with peripheral T-cell lymphoma. *Ann Oncol* (2008) 19:958–63. doi: 10.1093/annonc/mdn022

12. D'Amore F, Relander T, Lauritzsen GF, Jantunen E, Hagberg H, Anderson H, et al. Up-front autologous stem-cell transplantation in peripheral T-cell lymphoma: NLG-T-01. *J Clin Oncol* (2012) 30:3093–9. doi: 10.1200/JCO.2011.40.2719

13. Wilhelm M, Smetak M, Reimer P, Geissinger E, Ruediger T, Metzner B, et al. First-line therapy of peripheral T-cell lymphoma: extension and long-term follow-up of a study investigating the role of autologous stem cell transplantation. *Blood Cancer J* (2016) 6:e452–2. doi: 10.1038/bcj.2016.63

14. Rodríguez J, Caballero MD, Gutiérrez A, Marín J, Lahuerta JJ, Sureda A, et al. High-dose chemotherapy and autologous stem cell transplantation in peripheral T-cell lymphoma: the GEL-TAMO experience. *Ann Oncol* (2003) 14:1768–75. doi: 10.1093/annonc/mdg459

15. Park SI, Horwitz SM, Foss FM, Pinter-Brown LC, Carson KR, Rosen ST, et al. The role of autologous stem cell transplantation in patients with nodal peripheral T-cell lymphomas in first complete remission: Report from COMPLETE, a prospective, multicenter cohort study. *Cancer* (2019) 125:1507–17. doi: 10.1002/cncr.31861

16. Chen AI, McMillan A, Negrin RS, Horning SJ, Laport GG. Long-term results of autologous hematopoietic cell transplantation for peripheral T cell lymphoma: the stanford experience. *Biol Blood Marrow Transplant* (2008) 14:741–7. doi: 10.1016/j.bbmt.2008.04.004

17. Huang H, Jiang Y, Wang Q, Guo L, Jin Z, Fu Z, et al. Outcome of allogeneic and autologous hematopoietic cell transplantation for high-risk peripheral T cell lymphomas: a retrospective analysis from a Chinese center. *Biol Blood Marrow Transplant* (2017) 23:1393–7. doi: 10.1016/j.bbmt.2017.04.021

18. Rohlfing S, Dietrich S, Witzens-Harig M, Hegenbart U, Schönland S, Luft T, et al. The impact of stem cell transplantation on the natural course of peripheral T-cell lymphoma: a real-world experience. *Ann Hematol* (2018) 97:1241–50. doi: 10.1007/s00277-018-3288-7

19. Smith SM, Burns LJ, van Besien K, LeRademacher J, He W, Fenske TS, et al. Hematopoietic cell transplantation for systemic mature T-cell non-Hodgkin lymphoma. *J Clin Oncol* (2013) 31:3100–9. doi: 10.1200/JCO.2012.46.0188

20. Czajczynska A, Günther A, Repp R, Humpe A, Schub N, Raff T, et al. Allogeneic stem cell transplantation with BEAM and alemtuzumab conditioning immediately after remission induction has curative potential in advanced T-cell non-Hodgkin's lymphoma. *Biol Blood Marrow Transplant* (2013) 19:1632–7. doi: 10.1016/j.bbmt.2013.07.003

21. Wulf G, Hasenkamp J, Jung W, Wilhelm C, Held G, Nickelsen M, et al. Allogeneic stem cell transplantation for patients with relapsed or refractory T-cell lymphoma: efficacy of lymphoma-directed conditioning against advanced disease. *Bone Marrow Transplant* (2019) 54:877–84. doi: 10.1038/s41409-018-0360-9

22. Shustov AR, Gooley TA, Sandmaier BM, Shizuru J, Sorror ML, Sahebi F, et al. Allogeneic hematopoietic cell transplantation after nonmyeloablative conditioning in patients with T-cell and natural killer-cell lymphomas. *Br J Haematol* (2010) 150:170–8. doi: 10.1111/j.1365-2141.2010.08210.x

23. Jacobsen ED, Kim HT, Ho VT, Cutler CS, Koreth J, Fisher DC, et al. A large single-center experience with allogeneic stem-cell transplantation for peripheral T-cell non-Hodgkin lymphoma and advanced mycosis fungoides/Sezary syndrome. *Ann Oncol* (2011) 22:1608–13. doi: 10.1093/annonc/mdq698

24. Corradini P, Dodero A, Zallio F, Caracciolo D, Casini M, Bregni M, et al. Graft-versus-lymphoma effect in relapsed peripheral T-cell non-Hodgkin's lymphomas after reduced-intensity conditioning followed by allogeneic transplantation of hematopoietic cells. *J Clin Oncol* (2004) 22:2172–6. doi: 10.1200/JCO.2004.12.050

25. El-Asmar J, Reljic T, Ayala E, Hamadani M, Nishihori T, Kumar A, et al. Efficacy of high-dose therapy and autologous hematopoietic cell transplantation in peripheral T cell lymphomas as front-line consolidation or in the relapsed/refractory setting: A systematic review/meta-analysis. *Biol Blood Marrow Transplant* (2016) 22:802–14. doi: 10.1016/j.bbmt.2015.12.004

26. Sieniawski M, Angamuthu N, Boyd K, Chasty R, Davies J, Forsyth P, et al. Evaluation of enteropathy-associated T-cell lymphoma comparing standard therapies with a novel regimen including autologous stem cell transplantation. *Blood* (2010) 115:3664–70. doi: 10.1182/blood-2009-07-231324

27. Phillips EH, Lannon MM, Lopes A, Chadwick H, Jones G, Sieniawski M, et al. High-dose chemotherapy and autologous stem cell transplantation in enteropathy-associated and other aggressive T-cell lymphomas: a UK NCRI/Cancer Research UK Phase II Study. *Bone Marrow Transplant* (2019) 54:60–3. doi: 10.1038/s41409-018-0294-2

28. Jantunen E, Boumendil A, Finel H, Luan JJ, Johnson P, Rambaldi A, et al. Autologous stem cell transplantation for enteropathy-associated T-cell lymphoma: A retrospective study by the EBMT. *Blood* (2013) 121:2529–32. doi: 10.1182/blood-2012-11-466839

29. Nijeboer P, de Baaij LR, Visser O, Witte BI, Cillessen SAGM, Mulder CJ, et al. Treatment response in enteropathy associated T-cell lymphoma; survival in a large multicenter cohort. *Am J Hematol* (2015) 90:493–8. doi: 10.1002/ajh.23992

30. Rashidi A, Cashen AF. Outcomes of allogeneic stem cell transplantation in hepatosplenic T-cell lymphoma. *Blood Cancer J* (2015) 5:e318. doi: 10.1038/bcj.2015.43

31. Foss FM, Horwitz SM, Civallero M, Bellei M, Marcheselli L, Kim WS, et al. Incidence and outcomes of rare T cell lymphomas from the T Cell Project: hepatosplenic, enteropathy associated and peripheral gamma delta T cell lymphomas. *Am J Hematol* (2020) 95:151–5. doi: 10.1002/ajh.25674

32. Al-Mansour Z, Li H, Cook JR, Constine LS, Couban S, Stewart DA, et al. Autologous transplantation as consolidation for high risk aggressive T-cell non-Hodgkin lymphoma: a SWOG 9704 intergroup trial subgroup analysis. *Leukemia Lymphoma 60* (2019) 8:1934–194. doi: 10.1080/10428194.2018.1563691

33. Savage KJ, Horwitz SM, Advani R, Christensen JH, Domingo-Domenech E, Rossi G, et al. Role of stem cell transplant in CD30+ PTCL following frontline brentuximab vedotin plus CHP or CHOP in ECHELON-2. *Blood Adv* (2022) 6:5550–5. doi: 10.1182/bloodadvances.2020003971

34. Schmitz N, Truemper LH, Bouabdallah K, Ziepert M, Leclerc M, Cartron G, et al. A randomized phase 3 trial of auto vs. allo transplantation as part of first-line therapy in poor-risk peripheral T-NHL. *Blood* (2020) 19:2646–56. doi: 10.1182/blood.2020008825

35. Kharfan-Dabaja MA, Kumar A, Ayala E, Hamadani M, Reimer P, Gisselbrecht C, et al. Clinical practice recommendations on indication and timing of hematopoietic cell transplantation in mature T cell and NK/T cell lymphomas: an international collaborative effort on behalf of the guidelines committee of the American society for blood and ma. *Biol Blood Marrow Transplant* (2017) 23:1826–38. doi: 10.1016/j.bbmt.2017.07.027

36. Lansigan F, Horwitz SM, Pinter-Brown LC, Rosen ST, Pro B, Hsi ED, et al. Outcomes for relapsed and refractory peripheral T-cell lymphoma patients after front-line therapy from the COMPLETE registry. *Acta Haematol* (2020) 143:40–50. doi: 10.1159/000500666

37. Zhang JY, Briski R, Devata S, Kaminski MS, Phillips TJ, Mayer TL, et al. Survival following salvage therapy for primary refractory peripheral T-cell lymphomas (PTCL). *Am J Hematol* (2018) 93:394–400. doi: 10.1002/ajh.24992

38. Delioukina M, Zain J, Palmer JM, Tsai N, Thomas S, Forman S. Reduced-intensity allogeneic hematopoietic cell transplantation using fludarabine–melphalan conditioning for treatment of mature T-cell lymphomas. *Bone Marrow Transplant* (2012) 47:65–72. doi: 10.1038/bmt.2011.16

39. Dodero A, Spina F, Narni F, Patriarca F, Cavattoni I, Benedetti F, et al. Allogeneic transplantation following a reduced-intensity conditioning regimen in relapsed/refractory peripheral T-cell lymphomas: long-term remissions and response to donor lymphocyte infusions support the role of a graft-versus-lymphoma effect. *Leukemia* (2012) 26:520–6. doi: 10.1038/leu.2011.240

40. Epperla N, Ahn KW, Litovich C, Ahmed S, Battiwalla M, Cohen JB, et al. Allogeneic hematopoietic cell transplantation provides effective salvage despite refractory disease or failed prior autologous transplant in angioimmunoblastic T-cell lymphoma: A CIBMTR analysis. *J Hematol Oncol* (2019) 12:1–11. doi: 10.1186/s13045-018-0696-z

41. Le Gouill S, Milpied N, Buzyn A, De Latour RP, Vernant JP, Mohty M, et al. Graft-versus-lymphoma effect for aggressive T-cell lymphomas in adults: A study by the Société Française de Greffe de Moëlle et de Thérapie Cellulaire. *J Clin Oncol* (2008) 26:2264–71. doi: 10.1200/JCO.2007.14.1366

42. Du J, Yu D, Han X, Zhu L, Huang Z. Comparison of allogeneic stem cell transplant and autologous stem cell transplant in refractory or relapsed peripheral T-cell lymphoma. *JAMA Netw Open* (2021) 4:e219807. doi: 10.1001/jamanetworkopen.2021.9807

43. Firor AE, Jares A, Ma Y. From humble beginnings to success in the clinic: Chimeric antigen receptor-modified T-cells and implications for immunotherapy. *Exp Biol Med* (2015) 240:1087–98. doi: 10.1177/1535370215584936

44. Beatty GL, Haas AR, Maus MV, Torigian DA, Soulen MC, Plesa G, et al. Mesothelin-specific chimeric antigen receptor mRNA-engineered T cells induce anti-tumor activity in solid Malignancies. *Cancer Immunol Res* (2014) 2:112–20. doi: 10.1158/2326-6066.CIR-13-0170

45. Picanço-Castro V, Moço PD, Mizukami A, Vaz LD, de Souza Fernandes Pereira M, Silvestre RN, et al. Establishment of a simple and efficient platform for car-t cell

generation and expansion: from lentiviral production to in *vivo* studies. *Hematol Transfus Cell Ther* (2020) 42:150–8. doi: 10.1016/j.htct.2019.06.007

46. Svoboda J, Rheingold SR, Gill SI, Grupp SA, Lacey SF, Kulikovskaya I, et al. *Brief Report CLINICAL TRIALS AND OBSERVATIONS Nonviral RNA chimeric antigen receptor-modified T cells in patients with Hodgkin lymphoma* (2018). Available at: http://ashpublications.org/blood/article-pdf/132/10/1022/1406589/blood837609.pdf.

47. Birkholz K, Hombach A, Krug C, Reuter S, Kershaw M, Kämpgen E, et al. Transfer of mRNA encoding recombinant immunoreceptors reprograms CD4+ and CD8+ T cells for use in the adoptive immunotherapy of cancer. *Gene Ther* (2009) 16:596–604. doi: 10.1038/gt.2008.189

48. Barrett DM, Zhao Y, Liu X, Jiang S, Carpenito C, Kalos M, et al. Treatment of advanced leukemia in mice with mRNA engineered T cells. *Hum Gene Ther* (2011) 22:1575–86. doi: 10.1089/hum.2011.070

49. Stasi A, Tey S-K, Dotti G, Fujita Y, Kennedy-Nasser A, Martinez C, et al. Inducible Apoptosis as a Safety Switch for Adoptive Cell Therapy A bs t r ac t. *N Eng J Med* (2011) 365:1673-83. doi: 10.1056/NEJMoa1106152

50. Gonzalez BR, Zain J, Rosen ST, Querfeld C. Tumor microenvironment in mycosis fungoides and Sézary syndrome. *Curr Opin Oncol* (2016) 28:88–96. doi: 10.1097/CCO.0000000000000243

51. Querfeld C, Leung S, Myskowski PL, Curran SA, Goldman DA, Heller G, et al. Primary t cells from cutaneous t-cell lymphoma skin explants display an exhausted immune checkpoint profile. *Cancer Immunol Res* (2018) 6:900–9. doi: 10.1158/2326-6066.CIR-17-0270

52. Birnbaum ME, Berry R, Hsiao YS, Chen Z, Shingu-Vazquez MA, Yu X, et al. Molecular architecture of the αβ T cell receptor-CD3 complex. *Proc Natl Acad Sci U.S.A.* (2014) 111:17576–81. doi: 10.1073/pnas.1420936111

53. Gomes-Silva D, Srinivasan M, Sharma S, Lee CM, Wagner DL, Davis TH, et al. CD7-edited T cells expressing a CD7-specific CAR for the therapy of T-cell Malignancies. *Blood* (2017) 130:285–96. doi: 10.1182/blood-2017-01-761320

54. Mamonkin M, Rouce RH, Tashiro H, Brenner MK. A T-cell-directed chimeric antigen receptor for the selective treatment of T-cell Malignancies. *Blood* (2015) 126:983–92. doi: 10.1182/blood-2015-02-629527

55. Cooper ML, Choi J, Staser K, Ritchey JK, Devenport JM, Eckardt K, et al. An "off-the-shelf" fratricide-resistant CAR-T for the treatment of T cell hematologic Malignancies. *Leukemia* (2018) 32:1970–83. doi: 10.1038/s41375-018-0065-5

56. Fathi E, Farahzadi R, Sheervalilou R, Sanaat Z, Vietor I. A general view of CD33 + leukemic stem cells and CAR-T cells as interesting targets in acute myeloblatsic leukemia therapy. *Blood Res* (2020) 55:10–6. doi: 10.5045/br.2020.55.1.10

57. Jamal S, Picker LJ, Aquino DB, McKenna RW, Brian Dawson D, Kroft SH. *Immunophenotypic Analysis of Peripheral T-Cell Neoplasms A Multiparameter Flow Cytometric Approach* (2001). Available at: https://academic.oup.com/ajcp/article/116/4/512/1758178.

58. Maciocia PM, Wawrzyniecka PA, Philip B, Ricciardelli I, Akarca AU, Onuoha SC, et al. Targeting the T cell receptor β-chain constant region for immunotherapy of T cell Malignancies. *Nat Med* (2017) 23:1416–23. doi: 10.1038/nm.4444

59. Weiner DM, Durgin JS, Wysocka M, Rook AH. The immunopathogenesis and immunotherapy of cutaneous T cell lymphoma: Current and future approaches. *J Am Acad Dermatol* (2021) 84:597–604. doi: 10.1016/j.jaad.2020.12.026

60. Shen K, Liu Y, Cao X, Zhou D, Li J. Successful treatment of refractory Sezary syndrome by anti-PD-1 antibody (nivolumab). *Ann Hematol* (2017) 96:687–8. doi: 10.1007/s00277-017-2929-6

61. Pinz KG, Yakaboski E, Jares A, Liu H, Firor AE, Chen KH, et al. *Targeting T-cell Malignancies using anti-CD4 CAR NK-92 cells* (2017). Available at: www.impactjournals.com/oncotarget.

62. Perera LP, Zhang M, Nakagawa M, Petrus MN, Maeda M, Kadin ME, et al. Chimeric antigen receptor modified T cells that target chemokine receptor CCR4 as a therapeutic modality for T-cell Malignancies. *Am J Hematol* (2017) 92:892–901. doi: 10.1002/ajh.24794

63. Pan J, Tan Y, Wang G, Deng B, Ling Z, Song W, et al. Donor-derived CD7 chimeric antigen receptor T cells for T-cell acute lymphoblastic leukemia: first-in-human, phase I trial. *J Clin Oncol* (2021) 39:3340–51. doi: 10.1200/JCO.21.00389

64. Tan Y, Shan L, Zhao L, Deng B, Ling Z, Zhang Y, et al. Long-term follow-up of donor-derived CD7 CAR T-cell therapy in patients with T-cell acute lymphoblastic leukemia. *J Hematol Oncol* (2023) 16:1–15. doi: 10.1186/s13045-023-01427-3

65. Zhang Y, Li C, Du M, Jiang H, Luo W, Tang L, et al. Allogenic and autologous anti-CD7 CAR-T cell therapies in relapsed or refractory T-cell Malignancies. *Blood Cancer J* (2023) 13:1–10. doi: 10.1038/s41408-023-00822-w

66. Lu P, Liu Y, Yang J, Zhang X, Yang X, Wang H, et al. Naturally selected CD7 CAR-T therapy without genetic manipulations for T-ALL/LBL: first-in-human phase 1 clinical trial. *Blood* (2022) 140:321–34. doi: 10.1182/blood.2021014498

67. Rasaiyaah J, Georgiadis C, Preece R, Mock U, Qasim W. TCRαβ/CD3 disruption enables CD3-specific antileukemic T cell immunotherapy. *JCI Insight* (2018) 3. doi: 10.1172/jci.insight.99442

68. Morton LT, Reijmers RM, Wouters AK, Kweekel C, Remst DFG, Pothast CR, et al. Simultaneous deletion of endogenous TCRαβ for TCR gene therapy creates an improved and safe cellular therapeutic. *Mol Ther* (2020) 28:64–74. doi: 10.1016/j.ymthe.2019.10.001

69. Raikar SS, Fleischer LC, Moot R, Fedanov A, Paik NY, Knight KA, et al. Development of chimeric antigen receptors targeting T-cell Malignancies using two structurally different anti-CD5 antigen binding domains in NK and CRISPR-edited T cell lines. *Oncoimmunology* (2018) 7. doi: 10.1080/2162402X.2017.1407898

70. Hu Y, Zhou Y, Zhang M, Zhao H, Wei G, Ge W, et al. Genetically modified CD7-targeting allogeneic CAR-T cell therapy with enhanced efficacy for relapsed/refractory CD7-positive hematological Malignancies: a phase I clinical study. *Cell Res* (2022) 32:995–1007. doi: 10.1038/s41422-022-00721-y

71. Chiesa R, Georgiadis C, Syed F, Zhan H, Etuk A, Gkazi SA, et al. Base-edited CAR7 T cells for relapsed T-cell acute lymphoblastic leukemia. *New Engl J Med* (2023), 1–12. doi: 10.1056/nejmoa2300709

72. Kruschinski A, Moosmann A, Poschke I, Norell H, Chmielewski M, Seliger B, et al. *Engineering antigen-specific primary human NK cells against HER-2 positive carcinomas* (2008). Available at: www.pnas.org/cgi/content/full/.

73. Altvater B, Landmeier S, Pscherer S, Temme J, Schweer K, Kailayangiri S, et al. 2B4 (CD244) signaling by recombinant antigen-specific chimeric receptors costimulates natural killer cell activation to leukemia and neuroblastoma cells. *Clin Cancer Res* (2009) 15:4857–66. doi: 10.1158/1078-0432.CCR-08-2810

74. Shimasaki N, Fujisaki H, Cho D, Masselli M, Lockey T, Eldridge P, et al. A clinically adaptable method to enhance the cytotoxicity of natural killer cells against B-cell Malignancies. *Cytotherapy* (2012) 14:830–40. doi: 10.3109/14653249.2012.671519

75. Liu E, Tong Y, Dotti G, Shaim H, Savoldo B, Mukherjee M, et al. Cord blood NK cells engineered to express IL-15 and a CD19-targeted CAR show long-term persistence and potent antitumor activity. *Leukemia* (2018) 32:520–31. doi: 10.1038/leu.2017.226

76. Liu E, Marin D, Banerjee P, Macapinlac HA, Thompson P, Basar R, et al. Use of CAR-transduced natural killer cells in CD19-positive lymphoid tumors. *New Engl J Med* (2020) 382:545–53. doi: 10.1056/nejmoa1910607

77. Bednarski JJ, Zimmerman C, Berrien-Elliott MM, Foltz JA, Becker-Hapak M, Neal CC, et al. Donor memory-like NK cells persist and induce remissions in pediatric patients with relapsed AML after transplant. *Blood* (2022) 139:1670–83. doi: 10.1182/blood.2021013972

78. Glienke W, Esser R, Priesner C, Suerth JD, Schambach A, Wels WS, et al. Advantages and applications of CAR-expressing natural killer cells. *Front Pharmacol* (2015) 6:21. doi: 10.3389/fphar.2015.00021

79. Simonetta F, Alvarez M, Negrin RS. Natural killer cells in graft-versus-host-disease after allogeneic hematopoietic cell transplantation. *Front Immunol* (2017) 8:465. doi: 10.3389/fimmu.2017.00465

80. Prager I, Watzl C. Mechanisms of natural killer cell-mediated cellular cytotoxicity. *J Leukoc Biol* (2019) 105:1319–29. doi: 10.1002/JLB.MR0718-269R

81. Daher M, Rezvani K. Outlook for new car-based therapies with a focus on car nk cells: What lies beyond car-engineered t cells in the race against cancer. *Cancer Discovery* (2021) 11:45–58. doi: 10.1158/2159-8290.CD-20-0556

82. Tonn T, Schwabe D, Klingemann HG, Becker S, Esser R, Koehl U, et al. Treatment of patients with advanced cancer with the natural killer cell line NK-92. *Cytotherapy* (2013) 15:1563–70. doi: 10.1016/j.jcyt.2013.06.017

83. Chu J, Deng Y, Benson DM, He S, Hughes T, Zhang J, et al. CS1-specific chimeric antigen receptor (CAR)-engineered natural killer cells enhance in *vitro* and in *vivo* antitumor activity against human multiple myeloma. *Leukemia* (2014) 28:917–27. doi: 10.1038/leu.2013.279

84. Schönfeld K, Sahm C, Zhang C, Naundorf S, Brendel C, Odendahl M, et al. Selective inhibition of tumor growth by clonal NK cells expressing an ErbB2/HER2-specific chimeric antigen receptor. *Mol Ther* (2015) 23:330–8. doi: 10.1038/mt.2014.219

85. Chen KH, Wada M, Pinz KG, Liu H, Lin KW, Jares A, et al. Preclinical targeting of aggressive T-cell Malignancies using anti-CD5 chimeric antigen receptor. *Leukemia* (2017) 31:2151–60. doi: 10.1038/leu.2017.8

86. Chen KH, Wada M, Firor AE, Pinz KG, Jares A, Liu H, et al. *Novel anti-CD3 chimeric antigen receptor targeting of aggressive T cell Malignancies* (2016). Available at: www.impactjournals.com/oncotarget.

87. Xu Y, Liu Q, Zhong M, Wang Z, Chen Z, Zhang Y, et al. 2B4 costimulatory domain enhancing cytotoxic ability of anti-CD5 chimeric antigen receptor engineered natural killer cells against T cell Malignancies. *J Hematol Oncol* (2019) 12. doi: 10.1186/s13045-019-0732-7

88. Nielsen MM, Witherden DA, Havran WL. γδ T cells in homeostasis and host defence of epithelial barrier tissues. *Nat Rev Immunol* (2017) 17:733–45. doi: 10.1038/nri.2017.101

89. McCarthy NE, Eberl M. Human γδ T-cell control of mucosal immunity and inflammation. *Front Immunol* (2018) 9:985. doi: 10.3389/fimmu.2018.00985

90. Harrer DC, Simon B, Fujii S, Shimizu K, Uslu U, Schuler G, et al. RNA-transfection of γ/δ T cells with a chimeric antigen receptor or an α/β T-cell receptor: A safer alternative to genetically engineered α/β T cells for the immunotherapy of melanoma. *BMC Cancer* (2017) 17. doi: 10.1186/s12885-017-3539-3

91. Capsomidis A, Benthall G, Van Acker HH, Fisher J, Kramer AM, Abeln Z, et al. Chimeric antigen receptor-engineered human gamma delta T cells: enhanced cytotoxicity with retention of cross presentation. *Mol Ther* (2018) 26:354–65. doi: 10.1016/j.ymthe.2017.12.001

92. Papadopoulou A, Gerdemann U, Katari UL, Tzannou I, Liu H, Martinez C, et al. Activity of broad-spectrum T cells as treatment for AdV, EBV, CMV, BKV, and HHV6

infections after HSCT. *Sci Transl Med* (2014) 6(242):242ra83. doi: 10.1126/scitranslmed.3008825

93. Houghtelin A, Bollard CM. Virus-specific T cells for the immunocompromised patient. *Front Immunol* (2017) 8:1272. doi: 10.3389/fimmu.2017.01272

94. Melenhorst J, Leen AM, Bollard CM, Quigley MF, Price DA, Rooney CM, et al. Allogeneic virus-specific T cells with HLA alloreactivity do not produce GVHD in human subjects. *Blood* (2010) 116:4700–2. doi: 10.1182/blood-2010-06-289991

95. Godfrey DI, Kronenberg M. Going both ways: immune regulation *via* D1d-dependent NKT cells. *J Clin Invest* (2004) 114:1379–88. doi: 10.1172/JCI23594

96. Dellabona P, Padovan E, Casorati G, Brockhaus M, Lanzavecchia A. An invariant Va24-JaQ/Vb11 T Cell Receptor is expressed in all individuals in clonally expanded CD4-8- T cells. *J Exp Med* (1994) 180:1171–6. doi: 10.1084/jem.180.3.1171

97. Rowan AG, Ponnusamy K, Ren H, Taylor GP, Cook LBM, Karadimitris A. CAR-iNKT cells targeting clonal TCRVb chains as a precise strategy to treat T cell lymphoma. *Front Immunol* (2023). doi: 10.3389/fimmu.2023.1118681

98. Heczey A, Courtney AN, Montalbano A, Robinson S, Liu K, Li M, et al. AntiGD2 CAR-NKT cells in patients with relapsed or refractory neuroblastoma: An interim analysis. *Nat Med* (2020) 26:1686–90. doi: 10.1038/s41591-020-1074-2

99. Chen Y, Zhao H, Luo J, Liao Y, Dan X, Hu G, et al. A phase I dose-escalation study of neoantigen-activated haploidentical T cell therapy for the treatment of relapsed or refractory peripheral T-cell lymphoma. *Front Oncol* (2022) 12:944511. doi: 10.3389/fonc.2022.944511

100. O'Connor OA, Pro B, Pinter-Brown L, Bartlett N, Popplewell L, Coiffier B, et al. Pralatrexate in patients with relapsed or refractory peripheral T-cell lymphoma: Results from the pivotal PROPEL study. *J Clin Oncol* (2011) 29:1182–9. doi: 10.1200/JCO.2010.29.9024

101. Weisenburger DD, Savage KJ, Harris NL, Gascoyne RD, Jaffe ES, Maclennan KA, et al. Peripheral T-cell lymphoma, not otherwise specified: a report of 340 cases from the International Peripheral T-cell Lymphoma Project. *Blood* (2011) 117 :3402–8. doi: 10.1182/blood-2010-09

102. O'Connor OA, Horwitz S, Masszi T, Van Hoof A, Brown P, Doorduijn J, et al. Belinostat in patients with relapsed or refractory peripheral T-cell lymphoma: Results of the pivotal phase II BELIEF (CLN-19) study. *J Clin Oncol* (2015) 33:2492–9. doi: 10.1200/JCO.2014.59.2782

103. Matsushita H, Vesely MD, Koboldt DC, Rickert CG, Uppaluri R, Magrini VJ, et al. Cancer exome analysis reveals a T-cell-dependent mechanism of cancer immunoediting. *Nature* (2012) 482:400–4. doi: 10.1038/nature10755

104. Dagogo-Jack I, Shaw AT. Tumor heterogeneity and resistance to cancer therapies. *Nat Rev Clin Oncol* (2018) 15:81–94. doi: 10.1038/nrclinonc.2017.166

105. Horwitz S, O'Connor OA, Pro B, Trümper L, Iyer S, Advani R, et al. The ECHELON-2 Trial: 5-year results of a randomized, phase III study of brentuximab vedotin with chemotherapy for CD30-positive peripheral T-cell lymphoma. *Ann Oncol* (2022) 33:288–98. doi: 10.1016/j.annonc.2021.12.002

106. Abeyakoon C, van der Weyden C, Harrop S, Khot A, Dickinson M, Yannakou CK, et al. Role of hematopoietic stem cell transplantation in peripheral T-cell lymphoma. *Cancers (Basel)* (2020) 12:3125. doi: 10.3390/cancers12113125

107. Ramos RN, Couto SCF, Oliveira TGM, Klinger P, Braga TT, Rego EM, et al. Myeloid immune cells CARrying a new weapon against cancer. *Front Cell Dev Biol* (2021) 9:784421. doi: 10.3389/fcell.2021.784421

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 14 September 2023
DOI 10.3389/fonc.2023.1240359

OPEN ACCESS

EDITED BY
Sylvain Choquet,
Hôpitaux Universitaires Pitié Salpêtrière, France

REVIEWED BY
Patrice Morand,
Université Grenoble Alpes, France
Janos Minarovits,
University of Szeged, Hungary

*CORRESPONDENCE
Mario Henrique M. Barros
mariohenrique.barros@gmail.com

RECEIVED 14 June 2023
ACCEPTED 24 August 2023
PUBLISHED 14 September 2023

CITATION
Barros MHM and Alves PDS (2023) Contribution of the Epstein-Barr virus to the oncogenesis of mature T-cell lymphoproliferative neoplasms.
Front. Oncol. 13:1240359.
doi: 10.3389/fonc.2023.1240359

Contribution of the Epstein-Barr virus to the oncogenesis of mature T-cell lymphoproliferative neoplasms

Mario Henrique M. Barros[1*] and Paula Daniela S. Alves[2]

[1]Institute of Pathology, Klinikum Chemnitz, Chemnitz, Germany, [2]Oncovirology Laboratory, Bone Marrow Transplantation Center, Instituto Nacional de Câncer (INCA), Rio de Janeiro, RJ, Brazil

EBV is a lymphotropic virus, member of the *Herpesviridae* family that asymptomatically infects more than 90% of the human population, establishing a latent infection in memory B cells. EBV exhibits complex survival and persistence dynamics, replicating its genome through the proliferation of infected B cells or production of the lytic virions. Many studies have documented the infection of T/NK cells by EBV in healthy individuals during and after primary infection. This feature has been confirmed in humanized mouse models. Together these results have challenged the hypothesis that the infection of T/NK cells *per se* by EBV could be a triggering event for lymphomagenesis. Extranodal NK/T-cell lymphoma (ENKTCL) and Epstein-Barr virus (EBV)-positive nodal T- and NK-cell lymphoma (NKTCL) are two EBV-associated lymphomas of T/NK cells. These two lymphomas display different clinical, histological and molecular features. However, they share two intriguing characteristics: the association with EBV and a geographical prevalence in East Asia and Latin America. In this review we will discuss the genetic characteristics of EBV in order to understand the possible role of this virus in the oncogenesis of ENKTCL and NKTCL. In addition, the main immunohistological, molecular, cytogenetic and epigenetic differences between ENKTCL and NKTCL will be discussed, as well as EBV differences in latency patterns and other viral molecular characteristics.

KEYWORDS

Epstein-Barr virus, T-cell lymphoma, EBV-positive nodal T-and NK-cell lymphoma, extranodal NK/T-cell lymphoma, infectious mononucleosis, LMP1, HLA

Introduction

EBV is a lymphotropic virus, member of the *Herpesviridae* family that asymptomatically infects more than 90% of the human population (1). EBV enters the organism mainly via the oropharyngeal epithelium and infects circulating B cells to establish itself in a state of latency in the memory B cells. Its primoinfection occurs at variable ages, depending on the socioeconomic conditions of the populations (1, 2).

Although the primary infection is almost always subclinically controlled, it may lead to the clinical syndrome of infectious mononucleosis (IM) (1, 3) when it occurs in adolescents and young adults.

IM is an EBV-driven proliferation of B lymphocytes that is controlled by humoral and cellular immune responses (1, 3). Characteristically there is a florid T cell response mainly consisting of activated CD8+ cytotoxic T cells specific for lytic, and to a lesser extent, latent viral antigens expressed on EBV-infected B cells (3, 4).

In an immunocompetent population, EBV remains latent for most of the host's life. However, the viral load secreted in saliva can fluctuate over time and virions can be continuously released into saliva due to the viral reactivation process, which characterizes the switch from the latent to the lytic cycle of the virus (5). Additionally, a series of associations between terminal cell differentiation and EBV reactivation has been established (6), demonstrating that viral reactivation occurs mainly when the infected memory B cell is induced to differentiate into a plasma cell (7). Further, the transcription factors responsible for maintaining the memory B cell differentiation stage are described as repressors of lytic activation (8).

EBV is a ubiquitous herpesvirus and well-adapted to the human species, where most infections tend to converge to benign clinical outcomes. Although not being part of its natural replicative cycle, EBV is etiologically associated with the development of several neoplasms, highlighted by strong epidemiological and molecular evidences (9).

Solid and lymphoid neoplasms can be EBV-associated, such as nasopharyngeal carcinoma, gastric carcinoma, classical Hodgkin lymphoma, Burkitt lymphoma, post-transplant lymphoproliferative disease, extranodal NK/T-cell lymphoma (ENKTCL) and EBV-positive nodal T- and NK-cell lymphoma (NKTCL) (1, 10–12). It is estimated that approximately 200,000 new cases of EBV-associated tumours are diagnosed globally each year, which has led the International Agency for Research on Cancer (IARC) to consider it as group 1 carcinogen (9). The epidemiology of EBV-associated neoplasms is complex and may depend on age, sex, socioeconomic status, ethnographic customs, as well as host and viral genetic background (1, 6, 10, 13–15).

An established causal relation between EBV-infection of B cells and development of B cell lymphomas such as classical Hodgkin lymphoma and Burkitt lymphoma is well described (1, 12–14, 16, 17).

Much progress has been made in understanding how EBV transforms B cells and can contribute to their oncogenesis (1, 16–18). Meanwhile, since the first description of EBV association with the lethal midline granuloma (later renamed as ENKTCL), considerably less information has been published about its role in T and NK cell lymphomagenesis (19).

In lymphoid proliferations of T and NK cells, EBV is associated with ENKTCL, NKTCL, angioimmunoblastic- and follicular-type of nodal T-follicular helper cell lymphoma, systemic EBV-positive T-cell lymphoma of childhood as well as EBV-positive T- and NK-cell lymphoid proliferations (severe mosquito bite allergy, hydroa vacciniforme lymphoproliferative disorder and systemic chronic active EBV disease) (11, 18). Specially in the angioimmunoblastic- and follicular-type of nodal T-follicular helper cell lymphoma, EBV is detected mostly in the non-neoplastic B cells, while occasionally EBV-associated T cells, presumably reactive T cells, can be found in some cases (11, 18, 20, 21).

Due to a high incidence of ENKTCL and NKTCL in East Asian and Latin American populations a possible genetic predisposition has been suggested (18).

In this review we will discuss the genetic characteristics of EBV, its possible role in the oncogenesis of ENKTCL and NKTCL, two lymphomas of mature T and NK cells, as well as the main differences between these two lymphomas.

The Epstein-Barr virus

EBV is a gammaherpesvirus and was the first human candidate oncovirus in 1964 (22). Subsequently it was identified as a ubiquitous herpesvirus worldwide and it took many years to establish its etiological role in several human cancers (9). Nowadays EBV is causally associated with lymphoproliferations of B- or T- cell origins and carcinomas, which could reflect the EBV cell tropism (23, 24).

Viral latency

EBV exhibits complex survival and persistence dynamics, replicating its genome through the proliferation of infected B cells or production of the lytic virions. For this, the EBV expresses its genome differentially, characterizing the latency patterns (III, IIb, IIa, I and 0). The dynamics of EBV latencies were initially described in studies using B lymphoblastoid cell lineages and proposed as a germinal center model (25, 26). This model reflects mechanisms of viral adaptation through the host infection and it is characterized by successive downregulation of potentially immunogenic oncoproteins of EBV during the different stages of infection.

During the infection of immature B cells, the EBV genome, which is linear in the virion, circularizes through the fusion of its direct terminal repeats regions (TR), establishing itself in the episomal form (27, 28). In an initial moment, the virus begins to express a set of lytic genes in a transitory way, in a phase known as pre-latent abortive lytic cycle. In this phase there is no virion production or there is limited production of viral particles, which seems to occur due to the absence of methylation in the viral genome during this phase (29). Its known that the main lytic regulator of EBV, the Zta protein, preferentially binds to methylated DNA motifs, causing the transcription of lytic genes. During this initial phase that does not occur efficiently (30). Moreover this pre-latent abortive lytic phase proved to be essential for the establishment of latency in B cells (29).

Subsequently, the virus begins to express all of its latent genes. This phase of the infection is known as the growth program or the latency III profile and is characterized by the expression of six nuclear antigens (EBNA-1, -2, -3A, -3B, -3C and leader protein), three latent membrane protein (LMP-1, -2A and -2B), two

untranslatable RNAs (EBER1 and EBER2, herein referred to as EBERs) and two clusters of microRNAs (BART and BHRF), which gather more than 40 sequences of miRNAs (26). The latency III pattern can be detected during the primary EBV infection in IM, lymphoblastoid cell lines and post-transplant lymphoproliferative diseases (31, 32).

The latency IIb may be observed during the transitional phase between latency III and IIa, and it is characterized by the expression of *EBNA1*, *EBNA2*, *EBNA3A*, *EBNA3C*, *EBERs*, BART miRNAs and BHRF miRNAs. This latency is observed in IM and post-transplant lymphoproliferative diseases too (31, 33, 34).

In the latency IIa expression of *EBNA1*, *LMPs*, EBERs and BART miRNAs takes place. This latency program is responsible for the survival of EBV-infected B cells in the germinal centre reaction, allowing them the possibility to differentiate into memory B cells. This latency can be found in classic Hodgkin lymphoma and nasopharyngeal carcinoma (31).

After differentiation into a memory B cell, the EBV-infected B cell in asymptomatic individuals exhibits the latency 0 which is characterized by the presence of only EBERs (31).

Latency I is detected during homeostatic proliferation of EBV-infected memory B cells, ensuring the viral persistence in the progeny. In this latency only *EBNA1*, EBERs and BART miRNAs are detected. Beyond EBV-infected memory B cells in cell division, the latency I is identified in Burkitt lymphoma (31). After cell division, the expression program will return to latency 0.

Although it varies between individuals, the frequency of infected cells in peripheral blood remains relatively stable, with a constant absolute number over several years in healthy carriers (35). Furthermore, despite this apparent stability in the number of infected B cells in the host, the viral load in saliva fluctuates over time, as a consequence of the viral reactivation process (36).

The mechanisms that lead to viral reactivation are not fully understood, but evidence suggests that it possibly occurs when the infected memory B cells are stimulated to differentiate in plasma cells, for example by stimuli such as cognate antigen recognition and interaction with T cells (37). Due to the transcription factors associated with plasma cell differentiation, such as XPB-1 (X-box binding protein 1) and BLIMP1 (B-lymphocyte-induced maturation protein 1), *BZLF1*, the main regulator of the switch from the latent cycle to the lytic cycle, is activated inducing the expression of other lytic genes and resulting in the formation of new viral particles (38).

The initial activation of the *BZLF1*, which encodes the lytic transactivator Zta, leads to the expression of another important gene at the beginning of the lytic phase, the *BRLF1* gene, which encodes the Rta transcription factor (39). Together, these two transcription factors drive the expression of a series of viral genes that enable and direct the amplification of viral DNA, as well as enzymes necessary for replication (40, 41). This process results in the production of new viral particles by EBV-infected plasma cells, which migrate mainly to the Waldeyer ring region. The virus is released and it is able to infect new immature B cells, restarting the replicative cycle and maintaining the B cell compartment of infected memory B cells. In addition the new viral particles can infect local epithelial cells, where the virus will replicate, and posteriorly be released in saliva with the potential to infect other hosts (42).

Immune response

Taking into account the anti-EBV immune response, the viral proteins display a hierarchical immunodominance for the CD8+ T cell response. The strongest responses are induced by the proteins EBNA3A, EBNA3B, and EBNA3C, presenting in the latency III program (43, 44). A similar hierarchical response pattern is observed for the lytic cycle antigens, to which the strongest responses are observed against the immediate early antigens BZLF1, BRLF1 and BMRF1, also observed in the latency III (1, 43–45). These features allow the control of cells expressing the latency III program, consequently decreasing the deleterious potential offered by those proteins, which display high transforming power (43, 46).

The description of latency in the normal life cycle of EBV is only well characterized in B cells (32). Specially in IM, it is observed that different latency patterns can be found at the same time during the disease, possibly reflecting the dynamics of viral survival where the highly immunogenic latency pattern III is gradually replaced by the non-immunogenic latency pattern 0 (31, 47).

The molecular characteristics of EBV

EBV is also known as human gammaherpesvirus 4 (or HHV-4), and like all human herpesviruses, is considered a biological agent with stable genetic material. However, the long period of coevolution with the host led to the development of viral adaptation control against the conditions imposed by the host's antiviral defence (48, 49).

EBV possesses a complex double-strand DNA genome of approximately 172 kb, with the potential to translate more than 80 proteins and 46 functional small untranslated RNAs (EBERs, BART and BHRF1 miRNAs) (50). The majority of EBV's transcripts are expressed only during the lytic cycle and eleven of them are transcribed during latency, of which only nine are translated. Moreover, EBV genome has several internal direct repeats, which are found in latency promoters and in short and long sequences throughout the genome, as well as TR at both ends of the genome (50). The presence of clonal TR from EBV in EBV-associated neoplasms suggests that the virus was present from an early stage, before the oncogenesis (50).

EBV harbours genes with high conservation among herpesviruses, such as genes encoding lytic cycle proteins involved in viral DNA replication, viral particle structure and viral DNA packaging. However, other genes are shared only among the *Gammaherpesvirinae* subfamily, for example those that encode immediate initial controllers of the lytic cycle, as *BZLF1* and *BRLF1*, and the latent proteins LMP1 and LMP2. Furthermore, some genes have similarities with the host, such as: *BZLF1*, *BHRF1* and *BCRF1* which are similar to the *c-FOS*, *BCL-2* and *IL-10* of the host, respectively (51, 52).

As the latency genes of EBV may be related with the development of some neoplasms, they have been used to characterize the viral diversity. The main objective behind this approach is to try to differentiate if restricted strains are truly associated with a neoplasia or if it reflects only a geographical restriction, prevailing in a specific population (53–60).

The EBV can be separated in two different genotypes (type 1 and type 2), based on the differences in the sequences of *EBNA-2* and *EBNA-3* (61). The main functional difference between these genotypes is that the type 1 is more efficient in establishing lymphoblastoid cell lineages *in vitro* when compared to type 2 (50). Despite the functional differences, it is observed that the EBV type 1 is highly prevalent worldwide when compared to type 2 (53, 62–64). The types are associated with geographic restriction due to immunocompetence of the population and/or group studied, rather than disease association (53, 62–64). Although the EBV types are not commonly associated with neoplasms *per se*, previous epidemiological studies associated the haplotype Type1+V3 with tumours in Southeast Asia and AIDS-associated lymphomas (65, 66). The V3 polymorphism is located in the promoter zone (Zp) of the lytic transactivator *BZLF1* gene, which is responsible for switch from latent to lytic cycle (65, 66). Recently it was demonstrated that this haplotype confers a functional increase in viral lytic reactivation, which could favour tumour development, since viral reactivation is a known risk for EBV-associated neoplasms (67, 68). Therefore, EBV type 1 together with other genetic viral factors may contribute to the development of EBV-associated malignancies.

Early efforts to describe EBV diversity led to a genetic characterization of various latent proteins (69). From these, LMP1 is a well-characterized latent protein with the ability to transform and immortalize not only B cells but also epithelial cells *in vitro* (64, 70). LMP1 protein has 386 amino acids, as well as three domains with different characteristics and functions, during the viral replicative cycle and cellular transformation. Moreover *LMP1* is one of the most variable EBV genes, displays a high intra-host variability, has a high genetic diversity and is geographically restricted, reflecting human migration over the past few centuries (54, 57, 60, 71, 72).

In the 1990s, the *LMP1* variant called CAO was characterized, harbouring specific polymorphisms such as 30 bp deletion (del30) located at the 3'end of the C-terminal domain, compared to the prototype (59, 73, 74). This CAO variant, in a model of overexpression, was able to induce neoplasia *in vivo* (73, 74). Since then, *LMP1* polymorphisms have been extensively studied in several groups of EBV-associated malignancies worldwide (54, 59, 60, 75, 76). Several polymorphisms and mutation hotspots, such as 15 bp insertion (ins15) that encodes a Janus Kinase 3 (JAK3) motif and the number of 33 bp repeats were associated to specific variants, as well as B cell lymphomas and AIDS-associated B cell lymphomas in specific populations (54, 59, 60, 75, 76).

EBV-infection of T and NK cells

We previously described that although B cells are the main compartment of EBV infection in IM, T and dendritic cells are also infected during the primary infection, however to a minor amount. In the same study we were able to show a predominance of EBV-infected CD8+ T cells over CD4+ T cells (47). Like T cells, NK cells can also be infected by EBV *in vivo* (77, 78). Moreover we showed that the EBV-infected T cells expressed EBNA1 and EBNA2 proteins, but not BZLF1, suggesting absence of lytic cycle (47). It is unknown whether these EBV-infected T/NK cells survive as a viral reservoir, go into apoptosis or are being destroyed by the immune system. The results described by Coleman et al, which EBV-infected T cells can be found in healthy Kenyan children at 12 months of age with persistence through 24 months of age, suggest that the EBV-infected T cells may survive (79). *In vitro* studies and studies using humanized mouse models have confirmed these observations (80, 81). Taking into account these studies, it is very likely that the infection of "healthy" T/NK cells by EBV *per se* is not sufficient to trigger the lymphomagenesis in these cells. Additional events may be necessary to the establishment of a fully malignant phenotype and consequently development of ENKTCL and NKTCL (Figure 1).

ENKTCL

ENKTCL is an extranodal EBV-associated lymphoma of either NK or T cell lineage, which can affect nasal mucosa, skin, testis, kidney, gastrointestinal tract and salivary glands. As this lymphoma displays high prevalence in Asia and South America, it is hypothesized that population genetic characteristics, which influence the host immune response against EBV, may be related to lymphomagenesis (18, 82).

ENKTCL is characterized by a diffuse infiltrate of atypical lymphocytes of variable size, ranging from small to large. The nuclei are irregularly folded and hyperchromatic. In the small and medium atypical cells, the chromatin is granular, whereas in the large cells the nucleus tends to be vesicular. The nucleoli are usually small or inconspicuous. The amount of cytoplasm is moderate and frequently it appears pale to clear. Mitoses and apoptotic bodies are common. In most of the cases an angiocentric arrangement of tumour cells, together with angiodestruction and coagulative necrosis, is observed. Small lymphocytes, plasma cells, histiocytes and eosinophils are found in the background. Large areas of ulceration are noted in cases of mucosal presentation (11, 83).

Immunohistochemically the neoplastic cells are typically CD2+, CD56+, NKG2D+, NKG2A+, Tia1+, granzyme B+, perforin +, CD3-, CD4-, CD5-, CD8-, TCRαβ- and TCRγδ-, characterising the NK cell lineage (11, 84). CD7 shows a variable expression (11).

In the cases of true T cell lineage, the neoplastic cells are immunohistochemically CD2+, CD3+, CD5+, CD8+, Tia1+, granzyme B+ and perforin +. CD56 is normally negative, however can be positive. In addition there is the expression of TCRαβ or TCRγδ (11).

Cytogenetically ENKTCL are characterized by gains in 1p13, 2q33, 2q5, 3p14, 3q26, 6p21, 6p22, 7q34, 8q24, 9p24, 10q3, 13q4, 14q32, 17q21 and 22q11 as well as losses in chromosomes 1p4, 3q26, 5p13, 6q21-6q25, 8p22, 9p21, 12q3, 14q11, 14q21, 15q24, 17p13, 17p4, 18q22, 19q13 and 22q11 (85–88). Losses of 3q26 affected 50% of ENKTCL in one study, however abnormalities in this region are frequent in many neoplasms, including NKTCL and peripheral T cell lymphoma NOS (89, 90). Losses of 6q21-6q25 and the tumour suppressor genes present

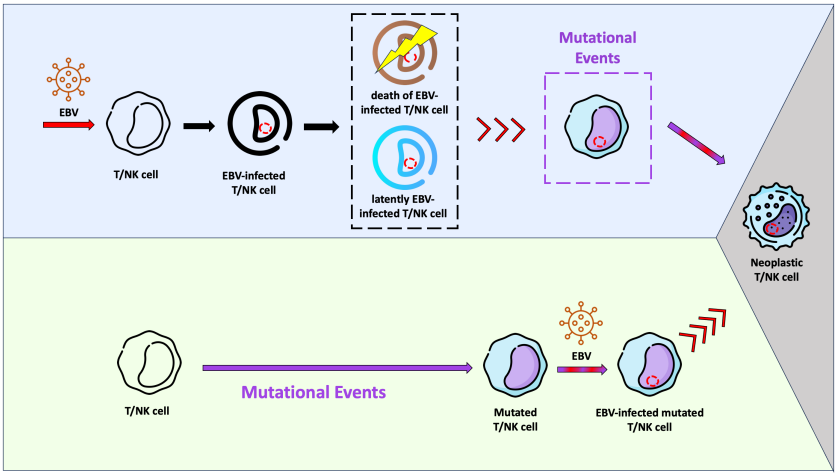


FIGURE 1
Schematic representation of two hypotheses related with the Epstein-Barr virus (EBV) infection and lymphomagenesis in T/NK cells. In the light purple background, a possible scenario is shown, where EBV infects "healthy" T/NK cells and stablishes a latent infection (cell in blue). It is possible that some of these infected cells are eliminated by the immune system or go into apoptosis (cell in brown). Viral reactivation phases with virus entry into the lytic cycle may occur in infected T/NK cells (red arrows), just as they happen in infected B cells. Mutational events in one EBV-infected T/NK cell may take place later, resulting in the fully malignant phenotype (grey background). In this context, EBV would act as an initiating agent. In the light green background, another hypothesis is presented. The EBV-infection occurs in a previously mutated T/NK cell (initiated cell) and the viral machinery would trigger the fully malignant phenotype (grey background). In this context, EBV would serve as a promoting agent.

in that region, as *PODC3*, *PREP*, *PRDM1*, *ATG5*, *AIM1* and *HACE1*, are found in circa 20-41% of ENKTCL (86–89).

Regarding the mutational profile, the ENTKTCL displays frequent mutations in *JAK3*, *STAT3* and *STAT5b* (genes from JAK-STAT signalling pathway); in *EPHA1*, *GNAQ*, *NOTCH3*, *PTPRK* and *PTPRQ* (genes from RAS-MAPK signalling pathway); in *ARID1A, ASXL1*, *BCOR1, EP300, KMT2D* and *MLL2* (epigenetic modifiers); in *DDX3X* (a RNA helicase gene); in *MGA* and *TP53* (tumour suppressor genes); as well as in *FAS* (gene related to apoptosis) (85, 91–99).

Many overexpressed genes have been described in ENTKTCL and some of them are suspected to be involved in the pathogenesis of this lymphoma. *EZH2* may be overexpressed in the majority of ENKTCL. This gene has a dual function; it can act as a histone methyltransferase, inhibiting the protective role of tumour suppressor genes or it can activate genes involved in oncogenic pathways (100). In ENKTCL *EZH2* directly promotes cyclin D1 expression and this is related to MYC-mediated repression of miRNAs, such as miR26 and miR101, that normally target and inhibit *EZH2* expression (100). It is important to note that *MYC* is upregulated in ENKTCL and is a transcriptional target of EBNA1, EBNA2 and LMP1 (31). *RUNX3* is also overexpressed in ENKTCL, due to transcriptional action of MYC, inducing cell proliferation and reduced apoptosis (101). Other overexpressed genes are *AURKA* (occasioning cell proliferation) *PDGFRA* and *PD-L1* (contributing to immune scape) (87, 102, 103).

Additionally, promoter methylation may be a frequent event in ENKTCL with consequent silencing of tumour suppressor genes (such *ASNA*, *BIM*, *DAPK1*, *SOCS6*, *SHP1* and *TET2*) and regulators of cell cycle (such *CDKN1A*, *CDKN2A* and *CDKN2B*) (104, 105).

Recently Xiong et al. suggested that ENKTCL can be molecularly classified in three different molecular subtypes, named TSIM (from Tumour Suppressor and Immune Modulation), MB (from *MGA* mutation and LOH in the *BRDT* locus) and HEA (from mutations in *HDAC1*, *EP300* and *ARID1A*) (85).

The TSIM subtype is characterized by high expression of NK cell genes (*GZMB*, *KIR2DL1/2/4*, *KLRC1/2/3*, *KLRD1*, *KLRK1* and *NCR1/3*), mutations in genes of JAK-STAT pathway and in *TP53*, as well as amp9p24.1/*PD-L1/2* locus and del6q21. In this subgroup, an upregulation of *PD-L1/2* is found (85), which makes this group of patients feasible for checkpoint inhibitor treatment, at least from a theoretical point of view. Prospective studies are needed to confirm this hypothesis.

The MB subtype is distinguished by *MGA* mutation and 1p22.1/*BRDT* LOH. Both *MGA* and *BRDT* dysfunctions are associated with *MYC* amplification and clinically with tumour dissemination (85).

High expression of T cells genes (*CD3D/G*, *CD8A/B*, CD28, *ICOS* and VAV2/3), as well as mutation in *HDAC9*, *EP300* and *ARID1A* characterize the HEA subtype. Aberrant histone acetylation is the hallmark of this subgroup (85).

NKTCL

NKTCL is a nodal EBV-associated lymphoma predominantly of T cells and more rarely of NK cells, which affects the lymph nodes (with or without extranodal involvement) and lacks nasal involvement (11). Clinically the patients are elderly or immunocompromised and display B symptoms as well as advanced stages (11). In the past, this lymphoma was known as EBV+ peripheral T cell lymphoma, not otherwise specified, and was part of the peripheral T cell lymphoma group. Now NKTCL is recognized as a distinct entity in the new WHO Classification (11).

Morphologically a diffuse and monomorphic proliferation of atypical cells of medium to large size with hyperchromatic nuclei

and enlarged nucleoli, sometimes reminiscent of centroblasts is observed. Unlike ENKTCL, there is no coagulative necrosis and no angioinvasion (11).

Immunohistochemically the neoplastic cells are typically CD2+, CD3+ CD8+, Tia1+, granzyme B+, perforin + and CD56-, characterizing the T cell lineage. CD4 expression is unusual. The expression of TCRαβ or TCRγδ can be observed (11, 89, 106).

Cases of NK cell lineage are described and characterized by the expression of CD56, Tia1, granzyme B, perforin, as well as CD4-negativity. A small proportion of cases can co-express CD8 and CD56 (CD8+CD56+). In this situation, a clonality analysis should be performed to distinguish between T and NK cell origin (11, 89, 106).

Genetically, gains were found only in a small proportion of cases in the regions 1p13, 2q33, 3p14, 6p21, 6p22, 8q24, 14q32 and 22q11. Chromosomal loss seems to be a more recurrent lesion in this disease and it includes losses in 3q26, 6q24, 8p22, 9p21, 14q11, 17p13 and 22q11 (89, 106).

Regarding the mutational profile, NKTCL is characterized by frequent mutation in *TET2* followed by *PIK3CD, STAT3, DDX3X* and *PTPRD* (89). Mutation in *ATM, SETD2, JAK3, IRF4, STAT5B, DMXL2, MGA, FYN, LRP1B, FBXW7, FAT3, NOTCH3, LTK, CIC, FGFR2, MITF, KIT, SDHA, FANCD2, TNKS2, TOP2A, SLIT2, AXIN2, SYK, RAD54L, HSD3B1, MAPK2K4, GRIN2A, RBM10, FAT1* and *KDR* were also described by Wai et al, however in a very low frequency (89). Furthermore, mutations in *RHOA*, besides in *TET2*, were recently described in two cases of NKTCL with a T follicular helper cell phenotype (107).

Considering the gene expression profile, NKTCL is characterized by overexpression of many genes, including T-related genes (*CD2, CD8, CD3G, CD3D, TRAC, LEF1*), NF-κB-related genes (*BIRC3, NF−κB1, TLR8 and CD27*), *PD-L1*, *CD68* as well as downregulation of *CD56* (89, 106).

Moreover, the IL6/JAK/STAT3 signalling axis may be aberrantly hyperactivated in NKTCL, contributing to proliferation, survival, invasiveness and dissemination of neoplastic cells, as well as suppression of the antitumour immune response (89, 107, 108). The *PD-L1* upregulation in NKTCL may be associated to hyperactivation of IL6/JAK/STAT3 signalling, IFN−γ, as well as NF −κB pathway, and not related to amplification of 9p24 (89, 107). Considering these characteristics and the fact that IL6/JAK/STAT3 signalling axis is already therapeutically targetable, patients with NKTCL could benefit from targeted therapy (108).

The landscape of epigenetic alterations associated with NKTCL is still unknown (18, 89, 108, 109).

A possible role of EBV in the oncogenesis of ENKTCL and NKTCL

Different EBV gene expression profiles reflect regulatory programs related to the lineage of infected host cells. Albeit EBV is detected in few T and NK cells in IM (47, 77, 78), the mechanism of infection in these cells type have not yet been completely elucidated. Some studies suggested that the infection of T and NK cells could occur through the immunological synapse, in an attempt by these cells to kill the EBV-infected B cells (23). We previously showed that EBV-infected T cells were mostly in contact with EBV-infected B cells in IM and the amount of EBV-infected T cells was directly related with the numbers of EBV-infected cells expressing PD-L1 (47). In addition, the molecule of HLA class II present on NK cells may interact with glycoproteins gp42 and gp85 from EBV, already described as fundamental in the internalization of EBV in HLA class II positive cells (110).

Recently it was demonstrated that CD21 cellular protein together with glycoprotein gp350 play an important role in the infection of the NK and/or mature T cells via trogocytosis, a mechanism that allows different cells to exchange pieces of their plasma membranes and suggested to occur in the interaction of mature T cells with malignant cells (23, 24).

Therefore, it is possible to think that the mechanism of EBV infection may be dependent on the cell type and the models so far established, using B cells, may not reflect what happens in other cell types. In consonance, EBV-infected T cells demonstrate *in vitro* a potential oncogenic distinct from those observed in LCLs, including different viral gene expression profile (111). Furthermore, previous studies *in vitro* demonstrated that T cells are possibly more permissive for the expression of immediate early viral genes than EBV-infected B cells (112, 113), with some evidence of this in EBV-associated neoplasia of T cell origin (114).

Although EBV has a dsDNA genome considered stable, genetic mechanisms that contribute to variability occur, such as point mutation, deletion, duplication, and intra/interstrain homologous recombination, as observed in *LMP1* (71, 72, 115). The del30 polymorphism of *LMP1* has been associated with diverse lymphomas worldwide (54, 61, 75). Moreover *LMP1* harbouring del30 may have a lower capacity to stimulate pro-inflammatory cytokine production, suggesting an immune escape ability, when compared to the prototype and del69 (116).

EBV in ENKTCL

EBV is present in virtually almost all cases of ENKTCL and should be detected by EBER-ISH (11). Despite a relative consensus that this lymphoma seems to show latency I/II, a bone find characterization of latency pattern in this disease is still missing. The published studies have determined the viral latency with molecular-based methodology (as gene expression profile and RNA-Seq) (85, 89, 117, 118), which does not make it possible to assess which viral genes are being transcribed at the same time in the same cell.

A proportion of cases seems to exhibit the latency I, based on the identification of high levels of transcripts from EBNA1 only (85, 117). Another fraction of cases seems to show the latency II on account of the identification of high levels of transcripts from *EBNA1, LMP1, LMP2A, LMP2B, BNRF1, BILF1, BALF2, BALF3, BALF4, BALF5* and *BNLF2b* (85, 117). A minor number of cases seems to be in latency III due to the identification of high levels of transcripts from *EBNA1, LMP1, LMP2A* and *EBNA2* (89).

Interestingly, high level of antibodies against the proteins EBNA3A, BZLF1, BALF2, BMRF1, BVRF and BPLF1 (but not against EBNA1) have been detected in patients with ENKTCL

(119), suggesting some degree of viral replication in these patients. The usefulness of these findings in the clinical management of ENKTCL is not yet established.

A precise *in situ* characterization of the viral latency in this disease, using multi colour immunohistochemistry or fluorescence *in situ* hybridization, as described for infectious mononucleosis (47), is still lacking. Furthermore, it remains to be determined whether all neoplastic cells show the same pattern of viral latency and which proportion of neoplastic cells could possibly show viral replication.

An apparent association of ENKTCL with EBV type 1 has been described (85, 120). However, EBV types *per se* are associated with specific populations than with diseases. EBV, specially type 1, is known to be highly frequent worldwide (121) and that association could represent an observational bias. Future studies focusing on the molecular characteristics of EBV and on host characteristics related to the anti-EBV immune response are needed to confirm that association. This notion is supported by some studies suggesting that few genetic regions of EBV may not be sufficient to understand the extent of EBV variation and its subsequent contribution to development of EBV-associated neoplasms (61, 122). The evaluation of different viral haplotypes needs to be included in further studies for a better understanding of EBV variants and their implications to the lymphomagenesis (61, 122).

Regarding the gene expression profile of EBV, this lymphoma subtype is enriched in the latent genes *LMP1/2A/2B*, *EBER1/2* and *EBNA1*, as well as lytic genes *BNRF1*, *BILF1*, *BALF5/4/3/2* and *BNLF2b* (85). Furthermore, single nucleotide variations in *BALF3* (G421R and T127A) may be prevalent in this lymphoma in relation to other EBV-associated diseases (85).

By considering the mutational profile of EBV, the del30 of *LMP1* is variable and depends on the population studied (120). Moreover small deletions in *EBNA2*, *EBNA3s* and *BLLF1/2* are common in ENKTCL (117). Frequent intragenic deletions affecting several BART micro-RNA clusters may be prevalent in ENKTCL (123). These deletions could impact the lytic cycle activation by eliciting the upregulation of *BZLF1* and *BRLF1*, which are downregulated by one of the BARTs miRNA.

Interestingly Xiong et al. found a correlation among EBV transcripts and their proposed molecular classification of ENKTCL (85). The TSIM subtype was associated with latency II pattern and high levels of *BALF3*; the MB subtype exhibited the lower levels of *LMP1* (as it is observed in the latency I pattern) and the HEA subtype correlated with latency II pattern and high levels of *BNRF1* (a protein necessary to latent infection) (85). In the same study the authors revealed that *BALF3* overexpression may cause DNA damage and contribute to genomic instability (85).

EBV in NKTCL

EBV is also present in virtually all cases of NKTCL and should be detected by EBER-ISH (11). The EBV latency pattern in this lymphoma is not firmly established and only few studies have dealt with this, using only molecular techniques (as RT-PCR) (89, 114).

Apparently, the majority of cases displays latency II with expression of *EBNA1*, *BART*, *LMP1*, *LMP2A* and *LMP2B* (89, 114). A small proportion of cases exhibits latency III with additional expression of *EBNA2* (89, 114). One study demonstrated that the expression of the early lytic genes *BZLF1* was not accompanied by the expression of the late lytic genes *BHRF1* and *BLLF1* (114), suggesting an abortive lytic cycle.

Like in ENKTCL a precise *in situ* characterization of the viral latency in NKTCL, using multi colour immunohistochemistry or fluorescence *in situ* hybridization is also lacking. It is unknown in how far all neoplastic cells display the same viral latency pattern and which proportion of neoplastic cells may be in lytic cycle.

In a small cohort of patients from Hong Kong, it was demonstrated that all cases presented the del30 of *LMP1* and the majority of cases carried type 1 EBV (114). Results like this need to be interpreted with caution. As discussed previously, the EBV type 1 is more prevalent worldwide and currently it is impossible to stablish an unbiased association between this subtype and NKTCL. Moreover, it is unclear if the association between the del30 of *LMP1* and this lymphoma reflects a role of tumour cells in the origin and selection of this variant or if this is an observational bias, due to a possible higher prevalence of del30 of *LMP1* in healthy individuals from Asia. For example, in a small study including individuals from different regions from Thailand, a prevalence of del30 of *LMP1* in the Southern region was observed (124). Large studies including healthy individuals from Asia and/or other parts of the world, evaluating the prevalence of del30 of *LMP1*, are still lacking.

Compared to ENKTCL, NKTCL may exhibit lower expression of EBV miRNA (89). The exact meaning of this observation needs to be clarified.

Although NKTCL is strongly associated with EBV, both genomic and transcription profiles of the virus, as well as the characterization of the humoral immune response against EBV, have not been robustly explored in NKTCL.

Viral proteins as candidates for target therapy

Although the therapeutic options for ENKTCL and NKTCL are not part of the objectives of this review in this research topic "Challenges in Peripheral T-Cell Lymphomas: from Biological Advances to Clinical Applicability" (125), we shall mention that epitopes derived of EBV proteins provide targets for immunotherapy in ENKTCL and NKTCL.

Adoptive immunotherapy with antigen-specific cytotoxic T cells (CTL) has been tested since the early 2000s for EBV-associated tumours and has been demonstrated to be safe (126–128).

As discussed previously, a hierarchical immunodominance for the CD8+ T cell response is observed against epitopes derived from proteins of the *EBNA3* family, *BZLF*, *BRLF1* and *BMRF1* (1, 43–45). Albeit less immunogenic, epitopes derived from *LMP1* and *LMP2* can be also used as potential targets for CD8+ T cells (128, 129). Considering the immune response of CD4+ T cells, the epitopes derived from *EBNA1* are the most immunodominant (43, 44, 128, 130, 131). The results published so far, regarding the characterization of latency pattern in ENKTCL and NKTCL, favour *LMP1*- and *LMP2*-derived epitopes as the best targets for adoptive immunotherapy with CTL.

Few studies using adoptive immunotherapy with CTL in ENKTCL have been published to date. All were phase 1 or 2; used *LMP1*- and/or *LMP2*-derived epitopes as target for the CTL (exception for the most recent, which included also *BARF1*- and *EBNA1*-derived epitopes); demonstrated no severe toxicity and exhibited objective responses in most of the cases, characterized by disease stability or remission during the follow-up time of the studies (132–135). These results are encouraging and point to the necessity of optimising this therapeutic option.

Currently there are no published studies on the use of CTL in NKTCL.

EBV viral load

The detection and quantification of circulating EBV DNA have been used in the diagnosis and management of EBV-associated neoplasms (136). Plasma and whole blood can be used to this quantification, with a good correlation between them. However, the optimal source of viral DNA remains uncertain, due to the limited number of studies comparing the two methodologies (136). *EBNA1*, *BamHI* and *LMP2* are commonly used as target for the viral load evaluation in real-time PCR-based assays (137–141).

Specially in ENKTCL, assessing the viral load in plasma seems to be more useful, since this methodology appears to reflect the tumour burden (137–139, 142). Moreover, high levels of EBV DNA load have shown a close correlation with a worse clinical outcome and prediction of early relapse (137–139).

The impact of EBV viral load on the clinical management of NKTCL is so far unknown.

Human leukocyte antigens in ENKTCL and NKTCL

Besides the EBV *per se*, the genetic background of the host, related to the anti-EBV and/or anti-tumour immune response, may also have an influence on the development of ENKTCL and NKTCL. It is well established that the anti-viral immune response is dependent on major histocompatibility complex (MHC) presentation of viral antigens (143–145).

During the primary EBV infection in patients with IM, the immune response is mainly characterized by a large expansion of EBV-specific CD8 + T cells. Response against immediate early and early lytic EBV epitopes may constitute half of the CD8+ T cells population (146). A directed immune response to late proteins is less frequent and in small amount, which directly impacts any future viral reactivation (146). Different levels of immunodominance may reflect the time which different epitopes are presented on the surface of infected cells. As the lytic cycle progresses the cell's antigen-processing capacity may be increasingly impaired by the set of viral immune evasion proteins (146). In this way, it is suggested that the main immune response against EBV is driven by direct CD8+ T cell contact with lytically infected cells, and this interaction is depending on MHC characteristics and EBV epitopes (44, 46).

In humans, the MHC is known as human leukocyte antigen (HLA), and it exhibits high polymorphism. Many studies have shown that different HLA haplotypes are associated with different types of diseases, such as autoimmune diseases and neoplasms (145, 147–153).

Regarding ENKTCL, the haplotype 47F-67I from *HLA-DRB1* may be associated with reduced risk of lymphoma development, while the haplotype 47Y-67L may be associated with increased risk in patients from various countries of East Asia. In the same study, the authors suggested that *HLA-DPB1* and *HLA-DRB1* are the two major genes independently conferring individual risk to ENKTCL (82). The β chain, which forms HLA-DR heterodimers with α chain, is encoded by *HLA-DRB1* (147). The different antigen binding affinities of the HLA-DR complex impact its ability to present extracellular antigens to CD4 T cell lymphocytes, influencing the immune response against EBV and/or tumour antigens (82, 147).

The association between variants of *HLA-DRB1* and ENKTCL must be confirmed by other robust studies. Furthermore, specific studies comparing the HLA subtypes between cases of NKTCL and controls are still lacking.

Conclusions

Although molecular, clinical and immunohistological differences are well established between ENKTCL and NKTCL, allowing the diagnostic difference between these neoplasms in clinical practice, the etiopathogenic role of EBV in these lymphomas has not yet been elucidated. In the light of current knowledge, there are more questions than answers.

The ability of EBV to infect T/NK cells during the primary infection is well characterised, as discussed before. Considering the studies published so far, it is very likely that this infection in T/NK cells *per se* does not trigger the lymphomagenesis *in vivo*, in the same way that it does not for EBV-infected B cells either. In other words, the EBV would act as an initiating agent, which can transform the cells it infects, however subsequent additional cellular events may be required for the fully malignant phenotype (Figure 1) (1, 154). Nonetheless, it is not possible to exclude a promoting role of EBV in the lymphomagenesis, as a consequence of its infection in previously mutated T/NK cells (Figure 1). The scenario in which ENKTCL and NKTCL can develop is still unknown.

May EBV-infected T/NK cells with persistent latency pattern II or III be the most susceptible to the oncogenic events? For example, ENKTCL is enriched in transcripts of latent EBV genes such as *LMP1/2A/2B*, as well as transcripts of lytic genes, including *BNRF1*, *BILF1*, *BALF5/4/3/2* and *BNLF2b* (85), which possess the ability to interfere with the host cell machinery. May recombination and/or mutation events of the EBV genome in some T/NK cell be responsible for the oncogenesis (or at least the initial event)? Specific T cell epitope mutations of EBV, favouring the immune evasion, have been described in ENKTCL (117). Furthermore, it is still unknown whether the molecular profile of EBV in neoplastic cells of ENKTCL and NKTCL is the same or not as that present in non-neoplastic EBV-infected cells of the same host.

No less important are the genetic characteristics of the host, specially those related to the anti-EBV immune response, which still need better characterisation in ENKTCL and NKTCL. Is it possible that the immune inability to recognise and destroy any EBV-infected T/NK cells in

persistent latency pattern other than 0 is a major factor in the lymphomagenesis? Some grade of immunodeficiency (as immunesenescence), for example, is present at diagnosis of NKTCL (11).

Nearly sixty years after the discovery of EBV, this virus still remains intriguing.

Author contributions

MHMB: conceptualization, writing, review and editing. PDSA: writing and review. All authors contributed to the article and approved the submitted version.

Funding

PDSA is postdoctoral researcher, supported by the Oncology Research Fellowship Program of the Instituto Nacional de Câncer (INCA) of the Ministry of Health of Brazil.

Acknowledgments

Dr. Stephan Föhse for editing of the manuscript.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Damania B, Kenney SC, Raab-Traub N. Epstein-Barr virus: Biology and clinical disease. *Cell* (2022) 185(20):3652–70. doi: 10.1016/j.cell.2022.08.026

2. Figueira-Silva CM, Pereira FEL. Prevalence of Epstein-Barr virus antibodies in healthy children and adolescents in Vitória, State of Espírito Santo, Brazil. *Rev da Sociedade Bras Medicina Tropical* (2004) 37(5):409–12. doi: 10.1590/S0037-86822004000500008

3. Vetsika EK, Callan M. Infectious mononucleosis and Epstein-Barr virus. *Expert Rev Mol Med* (2004) 6(23):1–16. doi: 10.1017/S1462399404008440

4. Callan M, Steven N, Krausa P, Wilson J, Moss P, Gillsepie G, et al. Large clonal expansion of CD8+ T cells in acute infectious mononucleosis. *Nat Med* (1996) 2 (8):906–11. doi: 10.1038/nm0896-906

5. Golden HD, Chang RS, Prescott W, Simpson E, Cooper TY. Leukocyte-transforming agent: prolonged excretion by patients with mononucleosis and excretion by normal individuals. *J Infect Diseases* (1973) 127(4):471–3. doi: 10.1093/infdis/127.4.471

6. Liang X, Collins CM, Mendel JB, Iwakoshi NN, Speck SH. Gammaherpesvirus-driven plasma cell differentiation regulates virus reactivation from latently infected B lymphocytes. *PloS Pathog* (2009) 5(11):e1000677. doi: 10.1371/journal.ppat.1000677

7. Sun CC, Thorley-Lawson DA. Plasma cell-specific transcription factor XBP-1s binds to and transactivates the epstein-barr virus BZLF1 promoter. *J Virol* (2007) 81 (24):13566–77. doi: 10.1128/JVI.01055-07

8. Raver RM, Panfil AR, Hagemeier SR, Kenney SC. The B-cell-specific transcription factor and master regulator pax5 promotes epstein-barr virus latency by negatively regulating the viral immediate early protein BZLF1. *J Virol* (2013) 87(14):8053–63. doi: 10.1128/JVI.00546-13

9. Centre international de recherche sur le cancer. *Epstein-Barr virus and Kaposi's sarcoma herpevirus: human herpesvirus 8*. Lyon: IARC (1997). IARC monographs on the evaluation of carcinogenic risks to humans.

10. Barros MHM, Hassan R, Niedobitek G. Disease patterns in pediatric classical Hodgkin lymphoma: a report from a developing area in Brazil. *Hematological Oncol* (2011) 29(4):190–5. doi: 10.1002/hon.984

11. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBDO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

12. Sausen DG, Basith A, Muqeemuddin S. EBV and lymphomagenesis. *Cancers* (2023) 15(7):2133. doi: 10.3390/cancers15072133

13. Chabay PA, Barros MHM, Matteo ED, Rey G, Carrico K, Renault IZ, et al. Pediatric hodgkin lymphoma in 2 South American series : A distinctive epidemiologic pattern and lack of association of epstein-barr virus with clinical outcome. *Journal of Pediatric Hematology/Oncology* (2008) 30(4):285–91. doi: 10.1097/MPH.0b013e3181647bc3

14. Hassan R, Klumb CE, Felisbino FE, Guiretti DM, White LR, Stefanoff CG, et al. Clinical and demographic characteristics of Epstein-Barr virus-associated childhood Burkitt's lymphoma in Southeastern Brazil: epidemiological insights from an intermediate risk region. *Haematologica* (2008) 93(5):780–3. doi: 10.3324/haematol.12424

15. Chabay P, Lens D, Hassan R, Rodríguez Pinilla SM, Valvert Gamboa F, Rivera I, et al. KSHV and HTLV in latin America: epidemiology and associated Malignancies. A literature-based study by the RIAL-CYTED. *Cancers* (2020) 12(8):2166. doi: 10.3390/cancers12082166

16. Marques-Piubelli ML, Salas YI, Pachas C, Becker-Hecker R, Vega F, MIranda RN. Epstein–Barr virus-associated B-cell lymphoproliferative disorders and lymphomas: a review. *Pathology* (2020) 52(1):40–52. doi: 10.1016/j.pathol.2019.09.006

17. Barros MHM, Scheliga A, De Matteo E, Minnicelli C, Soares Fa, Zalcberg IR, et al. Cell cycle characteristics and Epstein-Barr virus are differentially associated with aggressive and non-aggressive subsets of Hodgkin lymphoma in pediatric patients. *Leukemia lymphoma* (2010) 51(8):1513–22. doi: 10.3109/10428194.2010.489243

18. Shannon-Lowe C, Rickinson A. The global landscape of EBV-associated tumors. *Front Oncol* (2019) 9:713. doi: 10.3389/fonc.2019.00713

19. Harabuchi Y, Yamanaka N, Kataura A, Imai S, Kinoshita T, Osato T. Epstein-Barr virus in nasal T-cell lymphomas in patients with lethal midline granuloma. *Lancet* (1990) 335(8682):128–30. doi: 10.1016/0140-6736(90)90002-M

20. Weiss LM, Jaffe ES, Liu XF, Chen YY, Shibata D, Medeiros LJ. Detection and localization of Epstein-Barr viral genomes in angioimmunoblastic lymphadenopathy and angioimmunoblastic lymphadenopathy-like lymphoma. *Blood* (1992) 79(7):1789–95. doi: 10.1182/blood.V79.7.1789.1789

21. Jain S, Mallick S, Ramteke P, Gogia A. Neoplasms of follicular helper T-cells: an insight into the pathobiology. *Am J Blood Res* (2022) 12(3):64–81.

22. Epstein MA, Achong BG, Barr YM. Virus particles in cultured lymphoblasts from burkitt's lymphoma. *Lancet* (1964) 283(7335):702–3. doi: 10.1016/S0140-6736(64)91524-7

23. Tabiasco J, Vercellone A, Meggetto F, Hudrisier D, Brousset P, Fournié JJ. Acquisition of viral receptor by NK cells through immunological synapse. *J Immunol* (2003) 170(12):5993–8. doi: 10.4049/jimmunol.170.12.5993

24. Smith NA, Coleman CB, Gewurz BE, Rochford R. CD21 (Complement receptor 2) is the receptor for epstein-barr virus entry into T cells. *Longnecker RM editor J Virol* (2020) 94(11):e00428–20. doi: 10.1128/JVI.00428-20

25. Thorley-Lawson DA. EBV Persistence—Introducing the Virus. In: Münz C, editor. *Epstein Barr Virus*, vol. 1 . Cham: Springer International Publishing (2015). p. 151–209. (Current Topics in Microbiology and Immunology; vol. 390). doi: 10.1007/978-3-319-22822-8_8

26. Kang MS, Kieff E. Epstein–Barr virus latent genes. *Exp Mol Med* (2015) 47(1): e131–1. doi: 10.1038/emm.2014.84

27. Hurley EA, Thorley-Lawson DA. B cell activation and the establishment of Epstein-Barr virus latency. *J Exp Med* (1988) 168(6):2059–75. doi: 10.1084/jem.168.6.2059

28. Zimmermann J, Hammerschmidt W. Structure and role of the terminal repeats of Epstein-Barr virus in processing and packaging of virion DNA. *J Virol* (1995) 69 (5):3147–55. doi: 10.1128/jvi.69.5.3147-3155.1995

29. Inagaki T, Sato Y, Ito J, Takaki M, Okuno Y, Yaguchi M, et al. Direct evidence of abortive lytic infection-mediated establishment of epstein-barr virus latency during B-cell infection. *Front Microbiol* (2021) 11:575255. doi: 10.3389/fmicb.2020.575255

30. Kalla M, Schmeinck A, Bergbauer M, Pich D, Hammerschmidt W. AP-1 homolog BZLF1 of Epstein–Barr virus has two essential functions dependent on the epigenetic state of the viral genome. *Proc Natl Acad Sci USA* (2010) 107(2):850–5. doi: 10.1073/pnas.0911948107

31. Münz C. Latency and lytic replication in Epstein–Barr virus-associated oncogenesis. *Nat Rev Microbiol* (2019) 17(11):691–700. doi: 10.1038/s41579-019-0249-7

32. Babcock GJ, Hochberg D, Thorley-Lawson DA. The expression pattern of epstein-barr virus latent genes *in vivo* is dependent upon the differentiation stage of the infected B cell. *Immunity* (2000) 13(4):497–506. doi: 10.1016/S1074-7613(00)00049-2

33. Price AM, Luftig MA. To be or not IIb: A multi-step process for epstein-barr virus latency establishment and consequences for B cell tumorigenesis. *PloS Pathogens* (2015) 11(3):e1004656–e1004656. doi: 10.1371/journal.ppat.1004656

34. Klein E, Nagy N, Rasul AE. EBV genome carrying B lymphocytes that express the nuclear protein EBNA-2 but not LMP-1: Type IIb latency. *OncoImmunology* (2013) 2(2):e23035. doi: 10.4161/onci.23035

35. Khan G, Miyashita EM, Yang B, Babcock GJ, Thorley-Lawson DA. Is EBV persistence in vivo a model for B cell homeostasis? *Immunity* (1996) 5(2):173–9. doi: 10.1016/S1074-7613(00)80493-8

36. Hadinoto V, Shapiro M, Sun CC, Thorley-Lawson DA. The dynamics of EBV shedding implicate a central role for epithelial cells in amplifying viral output. *PloS Pathog* (2009) 5(7):e1000496. doi: 10.1371/journal.ppat.1000496

37. Laichalk LL, Thorley-Lawson DA. Terminal differentiation into plasma cells initiates the replicative cycle of epstein-barr virus in vivo. *J Virol* (2005) 79(2):1296–307. doi: 10.1128/JVI.79.2.1296-1307.2005

38. Kenney SC, Mertz JE. Regulation of the latent-lytic switch in Epstein–Barr virus. *Semin Cancer Biol* (2014) 26:60–8. doi: 10.1016/j.semcancer.2014.01.002

39. Chiu YF, Sugden B. Epstein-barr virus: the path from latent to productive infection. *Annu Rev Virol* (2016) 3(1):359–72. doi: 10.1146/annurev-virology-110615-042358

40. Fixman ED, Hayward GS, Hayward SD. Replication of Epstein-Barr virus oriLyt: lack of a dedicated virally encoded origin-binding protein and dependence on Zta in cotransfection assays. *J Virol* (1995) 69(5):2998–3006. doi: 10.1128/jvi.69.5.2998-3006.1995

41. Daikoku T, Kudoh A, Fujita M, Sugaya Y, Isomura H, Shirata N, et al. Architecture of replication compartments formed during epstein-barr virus lytic replication. *J Virol* (2005) 79(6):3409–18. doi: 10.1128/JVI.79.6.3409-3418.2005

42. Borza CM, Hutt-Fletcher LM. Alternate replication in B cells and epithelial cells switches tropism of Epstein–Barr virus. *Nat Med* (2002) 8(6):594–9. doi: 10.1038/nm0602-594

43. Hislop AD, Taylor GS, Sauce D, Rickinson AB. Cellular responses to viral infection in humans: lessons from Epstein-Barr virus. *Annu Rev Immunol* (2007) 25 (1):587–617. doi: 10.1146/annurev.immunol.25.022106.141553

44. Rickinson AB, Moss DJ. Human cytotoxic T lymphocyte responses to Epstein-Barr virus infection. *Annu Rev Immunol* (1997) 15(1):405–31. doi: 10.1146/annurev.immunol.15.1.405

45. Taylor GS, Long HM, Brooks JM, Rickinson AB, Hislop AD. The immunology of epstein-barr virus–induced disease. *Annu Rev Immunol* (2015) 33(1):787–821. doi: 10.1146/annurev-immunol-032414-112326

46. Thorley-Lawson DA. Epstein-Barr virus: exploiting the immune system. *Nat Rev Immunol* (2001) 1(1):75–82. doi: 10.1038/35095584

47. Barros MHM, Vera-Lozada G, Segges P, Hassan R, Niedobitek G. Revisiting the tissue microenvironment of infectious mononucleosis: identification of EBV infection in T cells and deep characterization of immune profiles. *Front Immunol* (2019) 10:146. doi: 10.3389/fimmu.2019.00146

48. McGeoch DJ, Rixon FJ, Davison AJ. Topics in herpesvirus genomics and evolution. *Virus Res* (2006) 117(1):90–104. doi: 10.1016/j.virusres.2006.01.002

49. Ehlers B, Spiess K, Leendertz F, Peeters M, Boesch C, Gatherer D, et al. Lymphocryptovirus phylogeny and the origins of Epstein-Barr virus. *J Gen Virol* (2010) 91(3):630–42. doi: 10.1099/vir.0.017251-0

50. Farrell PJ. Epstein–barr virus and cancer. *Annu Rev Pathol Mech Dis* (2019) 14 (1):29–53. doi: 10.1146/annurev-pathmechdis-012418-013023

51. Pearson GR, Luka J, Petti L, Sample J, Birkenbach M, Braun D, et al. Identification of an Epstein-Barr virus early gene encoding a second component of the restricted early antigen complex. *Virology* (1987) 160(1):151–61. doi: 10.1016/0042-6822(87)90055-9

52. Farrell PJ, Rowe DT, Rooney CM, Kouzarides T. Epstein-Barr virus BZLF1 trans-activator specifically binds to a consensus AP-1 site and is related to c-fos. *EMBO J* (1989) 8(1):127–32. doi: 10.1002/j.1460-2075.1989.tb03356.x

53. Young LS, Yap LF, Murray PG. Epstein–Barr virus: more than 50 years old and still providing surprises. *Nat Rev Cancer* (2016) 16(12):789–802. doi: 10.1038/nrc.2016.92

54. Guiretti DM, Chabay PA, Valva P, Stefanoff CG, Barros MHM, De Matteo E, et al. Structural variability of the carboxy-terminus of Epstein-Barr virus encoded latent membrane protein 1 gene in Hodgkin's lymphomas. *J Med Virol* (2007) 79(11):1730–22. doi: 10.1002/jmv.21020

55. Bristol JA, Djavadian R, Albright ER, Coleman CB, Ohashi M, Hayes M, et al. A cancer-associated Epstein-Barr virus BZLF1 promoter variant enhances lytic infection. *PloS Pathog* (2018) 14(7):e1007179. doi: 10.1371/journal.ppat.1007179

56. Liao HM, Liu H, Chin PJ, Li B, Hung GC, Tsai S, et al. Epstein-barr virus in Burkitt lymphoma in Africa reveals a limited set of whole genome and LMP-1 sequence patterns: analysis of archival datasets and field samples from Uganda, Tanzania, and Kenya. *Front Oncol* (2022) 12:812224. doi: 10.3389/fonc.2022.812224

57. Edwards RH, Seillier-Moiseiwitsch F, Raab-Traub N. Signature amino acid changes in latent membrane protein 1 distinguish epstein–barr virus strains. *Virology* (1999) 261(1):79–95. doi: 10.1006/viro.1999.9855

58. Lorenzetti MA, Gantuz M, Altcheh J, De Matteo E, Chabay PA, Preciado MV. Epstein–Barr virus BZLF1 gene polymorphisms: Malignancy related or geographically distributed variants? *Clin Microbiol Infection* (2014) 20(11):O861–9. doi: 10.1111/1469-0691.12631

59. Zuercher E, Butticaz C, WyNiger J, Martinez R, Battegay M, Boffi El Amari E, et al. Genetic diversity of EBV-encoded LMP1 in the swiss HIV cohort study and implication for NF-κb activation. *PloS One* (2012) 7(2):e32168. doi: 10.1371/journal.pone.0032168

60. Alves P, Larrate M, Garcia-Costa A, Rohan P, Gama BE, Abdelhay E, et al. Spatial dispersal of epstein–barr virus in South America reveals an African American variant in Brazilian lymphomas. *Viruses* (2022) 14(8):1762. doi: 10.3390/v14081762

61. Palser AL, Grayson NE, White RE, Corton C, Correia S, Ba Abdullah MM, et al. Genome diversity of epstein-barr virus from multiple tumor types and normal infection. *J Virol* (2015) 89(10):5222–37. doi: 10.1128/JVI.03614-14

62. Kwok H, Chan KW, Chan KH, Chiang AKS. Distribution, persistence and interchange of epstein-barr virus strains among PBMC, plasma and saliva of primary infection subjects. *PloS One* (2015) 10(3):e0120710. doi: 10.1371/journal.pone.0120710

63. Macsween KF, Crawford DH. Epstein-Barr virus—recent advances. *Lancet Infect Diseases* (2003) 3(3):131–40. doi: 10.1016/S1473-3099(03)00543-7

64. Young LS, Rickinson AB. Epstein-Barr virus: 40 years on. *Nat Rev Cancer* (2004) 4(10):757–68. doi: 10.1038/nrc1452

65. Tong JHM, Lo KW, Au FWL, Huang DP, To KF. Re: discrete alterations in the BZLF1 promoter in tumor and non-tumor-associated epstein-barr virus. *JNCI J Natl Cancer Institute* (2003) 95(13):1008–9. doi: 10.1093/jnci/95.13.1008

66. Martini M, Capello D, Serraino D, Navarra A, Pierconti F, Cenci T, et al. Characterization of variants in the promoter of EBV gene BZLF1 in normal donors, HIV-positive patients and in AIDS-related lymphomas. *J Infection* (2007) 54(3):298–306. doi: 10.1016/j.jinf.2006.04.015

67. Geser A, De Thé G, Lenoir G, Day NE, Williams EH. Final case reporting from the Ugandan prospective study of the relationship between ebv and burkitt's lymphoma. *Int J Cancer* (1982) 29(4):397–400. doi: 10.1002/ijc.2910290406

68. Wu CC, Fang CY, Huang SY, Chiu SH, Lee CH, Chen JY. Perspective: contribution of epstein–barr virus (EBV) reactivation to the carcinogenicity of nasopharyngeal cancer cells. *Cancers* (2018) 10(4):120. doi: 10.3390/cancers10040120

69. Kanda T, Yajima M, Ikuta K. Epstein-Barr virus strain variation and cancer. *Cancer Sci* (2019) 110(4):1132–9. doi: 10.1111/cas.13954

70. Kaye KM, Izumi KM, Kieff E. Epstein-Barr virus latent membrane protein 1 is essential for B-lymphocyte growth transformation. *Proc Natl Acad Sci USA* (1993) 90 (19):9150–4. doi: 10.1073/pnas.90.19.9150

71. Berenstein AJ, Lorenzetti MA, Preciado MV. Recombination rates along the entire Epstein Barr virus genome display a highly heterogeneous landscape. *Infection Genet Evolution* (2018) 65:96–103. doi: 10.1016/j.meegid.2018.07.022

72. Walling DM, Shebib N, Weaver SC, Nichols CM, Flaitz CM, Webster-Cyriaque J. The molecular epidemiology and evolution of epstein-barr virus: sequence variation and genetic recombination in the latent membrane protein-1 gene. *J Infect Dis* (1999) 179(4):763–74. doi: 10.1086/314672

73. Chen ML, Tsai CN, Liang CL, Shu CH, Huang CR, Sulitzeanu D, et al. Cloning and characterization of the latent membrane protein (LMP) of a specific Epstein-Barr virus variant derived from the nasopharyngeal carcinoma in the Taiwanese population. *Oncogene* (1992) 7(11):2131–40.

74. Hu LF, Chen F, Zheng X, Ernberg I, Cao SL, Christensson B, et al. Clonability and tumorigenicity of human epithelial cells expressing the EBV encoded membrane protein LMP1. *Oncogene* (1993) 8(6):1575–83.

75. Lorenzetti MA, Gantuz M, Altcheh J, De Matteo E, Chabay PA, Preciado MV. Distinctive epstein-barr virus variants associated with benign and Malignant pediatric pathologies: LMP1 sequence characterization and linkage with other viral gene polymorphisms. *J Clin Microbiol* (2012) 50(3):609–18. doi: 10.1128/JCM.05778-11

76. Gantuz M, Lorenzetti MA, Chabay PA, Preciado MV. A novel recombinant variant of latent membrane protein 1 from Epstein Barr virus in Argentina denotes phylogeographical association. *PloS One* (2017) 12(3):e0174221. doi: 10.1371/journal.pone.0174221

77. Anagnostopoulos I, Hummel M, Kreschel C, Stein H. Morphology, immunophenotype, and distribution of latently and/or productively Epstein-Barr virus-infected cells in acute infectious mononucleosis: implications for the interindividual infection route of Epstein-Barr virus. *Blood* (1995) 85(3):744–50. doi: 10.1182/blood.V85.3.744.bloodjournal853744

78. Trempat P, Tabiasco J, Andre P, Faumont N, Meggetto F, Delsol G, et al. Evidence for early infection of nonneoplastic natural killer cells by Epstein-Barr virus. *J virol* (2002) 76(21):11139–42. doi: 10.1128/JVI.76.21.11139-11142.2002

79. Coleman CB, Daud II, Ogolla SO, Ritchie JA, Smith NA, Sumba PO, et al. Epstein-barr virus type 2 infects T cells in healthy Kenyan children. *J Infect Diseases* (2017) 216(6):670–7. doi: 10.1093/infdis/jix363

80. Coleman CB, Wohlford EM, Smith NA, King CA, Ritchie JA, Baresel PC, et al. Epstein-barr virus type 2 latently infects T cells, inducing an atypical activation characterized by expression of lymphotactic cytokines. *J Virol* (2015) 89(4):2301–12. doi: 10.1128/JVI.03001-14

81. Coleman CB, Lang J, Sweet LA, Smith NA, Freed BM, Pan Z, et al. Epstein-barr virus type 2 infects T cells and induces B cell lymphomagenesis in humanized mice. *J Virol* (2018) 92(21):e00813–18. doi: 10.1128/JVI.00813-18

82. Lin GW, Xu C, Chen K, Huang HQ, Chen J, Song B, et al. Genetic risk of extranodal natural killer T-cell lymphoma: a genome-wide association study in multiple populations. *Lancet Oncol* (2020) 21(2):306–16. doi: 10.1016/S1470-2045(19)30799-5

83. Jhuang JY, Chang ST, Weng SF, Pan ST, Chu PY, Hsieh PP, et al. Extranodal natural killer/T-cell lymphoma, nasal type in Taiwan: a relatively higher frequency of T-cell lineage and poor survival for extranasal tumors. *Hum Pathol* (2015) 46(2):313–21. doi: 10.1016/j.humpath.2014.11.008

84. Haedicke W, Ho FC, Chott A, Moretta L, Rüdiger T, Ott G, et al. Expression of CD94/NKG2A and killer immunoglobulin-like receptors in NK cells and a subset of extranodal cytotoxic T-cell lymphomas. *Blood* (2000) 95(11):3628–30. doi: 10.1182/blood.V95.11.3628

85. Xiong J, Cui BW, Wang N, Dai YT, Zhang H, Wang CF, et al. Genomic and transcriptomic characterization of natural killer T cell lymphoma. *Cancer Cell* (2020) 37(3):403–419.e6. doi: 10.1016/j.ccell.2020.02.005

86. Huang Y, de Reyniès A, de Leval L, Ghazi B, Martin-Garcia N, Travert M, et al. Gene expression profiling identifies emerging oncogenic pathways operating in extranodal NK/T-cell lymphoma, nasal type. *Blood* (2010) 115(6):1226–37. doi: 10.1182/blood-2009-05-221275

87. Iqbal J, Weisenburger DD, Chowdhury A, Tsai MY, Srivastava G, Greiner TC, et al. Natural killer cell lymphoma shares strikingly similar molecular features with a group of non-hepatosplenic γδ T-cell lymphoma and is highly sensitive to a novel aurora kinase A inhibitor in *vitro*. *Leukemia* (2011) 25(2):348–58. doi: 10.1038/leu.2010.255

88. Karube K, Nakagawa M, Tsuzuki S, Takeuchi I, Honma K, Nakashima Y, et al. Identification of FOXO3 and PRDM1 as tumor-suppressor gene candidates in NK-cell neoplasms by genomic and functional analyses. *Blood* (2011) 118(12):3195–204. doi: 10.1182/blood-2011-04-346890

89. Wai CMM, Chen S, Phyu T, Fan S, Leong SM, Zheng W, et al. Immune pathway upregulation and lower genomic instability distinguish EBV-positive nodal T/NK-cell lymphoma from ENKTL and PTCL-NOS. *haematol* (2022) 107(8):1864–79. doi: 10.3324/haematol.2021.280003

90. Fields AP, Justilien V, Murray NR. The chromosome 3q26 OncCassette: A multigenic driver of human cancer. *Adv Biol Regulation* (2016) 60:47–63. doi: 10.1016/j.jbior.2015.10.009

91. Küçük C, Jiang B, Hu X, Zhang W, Chan JKC, Xiao W, et al. Activating mutations of STAT5B and STAT3 in lymphomas derived from γδ-T or NK cells. *Nat Commun* (2015) 6(1):6025. doi: 10.1038/ncomms7025

92. Song TL, Nairismägi ML, Laurensia Y, Lim JQ, Tan J, Li ZM, et al. Oncogenic activation of the STAT3 pathway drives PD-L1 expression in natural killer/T-cell lymphoma. *Blood* (2018) 132(11):1146–58. doi: 10.1182/blood-2018-01-829424

93. Sim SH, Kim S, Kim TM, Jeon YK, Nam SJ, Ahn YO, et al. Novel JAK3-activating mutations in extranodal NK/T-cell lymphoma, nasal type. *Am J Pathol* (2017) 187(5):980–6. doi: 10.1016/j.ajpath.2017.01.004

94. Li Z, Zhang X, Xue W, Zhang Y, Li C, Song Y, et al. Recurrent GNAQ mutation encoding T96S in natural killer/T cell lymphoma. *Nat Commun* (2019) 10(1):4209. doi: 10.1038/s41467-019-12032-9

95. de Mel S, Hue SSS, Jeyasekharan AD, Chng WJ, Ng SB. Molecular pathogenic pathways in extranodal NK/T cell lymphoma. *J Hematol Oncol* (2019) 12(1):33. doi: 10.1186/s13045-019-0716-7

96. Jiang L, Gu ZH, Yan ZX, Zhao X, Xie YY, Zhang ZG, et al. Exome sequencing identifies somatic mutations of DDX3X in natural killer/T-cell lymphoma. *Nat Genet* (2015) 47(9):1061–6. doi: 10.1038/ng.3358

97. Quintanilla-Martinez L, Kremer M, Keller G, Nathrath M, Gamboa-Dominguez A, Meneses A, et al. p53 mutations in nasal natural killer/T-cell lymphoma from Mexico. *Am J Pathol* (2001) 159(6):2095–105. doi: 10.1016/S0002-9440(10)63061-1

98. Takakuwa T, Dong Z, Nakatsuka S, Kojya S, Harabuchi Y, Yang WI, et al. Frequent mutations of Fas gene in nasal NK/T cell lymphoma. *Oncogene* (2002) 21(30):4702–5. doi: 10.1038/sj.onc.1205571

99. Shen L, Liang ACT, Lu L, Au WY, Kwong YL, Liang RHS, et al. Frequent deletion of fas gene sequences encoding death and transmembrane domains in nasal natural killer/T-cell lymphoma. *Am J Pathol* (2002) 161(6):2123–31. doi: 10.1016/S0002-9440(10)64490-2

100. Yan J, Ng SB, Tay JLS, Lin B, Koh TL, Tan J, et al. EZH2 overexpression in natural killer/T-cell lymphoma confers growth advantage independently of histone methyltransferase activity. *Blood* (2013) 121(22):4512–20. doi: 10.1182/blood-2012-08-450494

101. Selvarajan V, Osato M, Nah GSS, Yan J, Chung TH, Voon DCC, et al. RUNX3 is oncogenic in natural killer/T-cell lymphoma and is transcriptionally regulated by MYC. *Leukemia* (2017) 31(10):2219–27. doi: 10.1038/leu.2017.40

102. Chen YP, Chang KC, Su WC, Chen TY. The expression and prognostic significance of platelet-derived growth factor receptor alpha in mature T- and natural killer-cell lymphomas. *Ann Hematol* (2008) 87(12):985–90. doi: 10.1007/s00277-008-0539-z

103. Bi X-w, Wang H, Zhang W-w, Wang J-h, Liu W-j, Xia Z-j, et al. PD-L1 is upregulated by EBV-driven LMP1 through NF-κB pathway and correlates with poor prognosis in natural killer/T-cell lymphoma. *J Hematol Oncol* (2016) 9(1):109. doi: 10.1186/s13045-016-0341-7

104. Kawamata N, Inagaki N, Mizumura S, Sugimoto K-j, Sakajiri S, Ohyanagi-Hara M, et al. Methylation status analysis of cell cycle regulatory genes (p16INK4A, p15INK4B, p21Waf1/Cip1, p27Kip1 and p73) in natural killer cell disorders. *Eur J Haematol* (2005) 74(5):424–9. doi: 10.1111/j.1600-0609.2005.00417.x

105. Küçük C, Hu X, Jiang B, Klinkebiel D, Geng H, Gong Q, et al. Global promoter methylation analysis reveals novel candidate tumor suppressor genes in natural killer cell lymphoma. *Clin Cancer Res* (2015) 21(7):1699–711. doi: 10.1158/1078-0432.CCR-14-1216

106. Ng SB, Chung TH, Kato S, Nakamura S, Takahashi E, Ko YH, et al. Epstein-Barr virus-associated primary nodal T/NK-cell lymphoma shows a distinct molecular signature and copy number changes. *Haematologica* (2018) 103(2):278–87. doi: 10.3324/haematol.2017.180430

107. Zhang Z, Wang W, Li Y, Xu Z, Wu H, Wang Z. A nodal EBV-positive T cell lymphoma with a T follicular helper cell phenotype. *Histopathology* (2023) 1:14919. doi: 10.1111/his.14919

108. Johnson DE, O'Keefe RA, Grandis JR. Targeting the IL-6/JAK/STAT3 signalling axis in cancer. *Nat Rev Clin Oncol* (2018) 15(4):234–48. doi: 10.1038/nrclinonc.2018.8

109. Kato S, Yamashita D, Nakamura S. Nodal EBV+ cytotoxic T-cell lymphoma: A literature review based on the 2017 WHO classification. *JCEH* (2020) 60(2):30–6. doi: 10.3960/jslrt.20001

110. Isobe Y, Sugimoto K, Yang L, Tamayose K, Egashira M, Kaneko T, et al. Epstein-barr virus infection of human natural killer cell lines and peripheral blood natural killer cells. *Cancer Res* (2004) 64(6):2167–74. doi: 10.1158/0008-5472.CAN-03-1562

111. Kelleher C. EBV infection of T cells: potential role in Malignant transformation. *Semin Cancer Biol* (1996) 7(4):197–207. doi: 10.1006/scbi.1996.0027

112. Yoshiyama H, Shimizu N, Takada K. Persistent Epstein-Barr virus infection in a human T-cell line: unique program of latent virus expression. *EMBO J* (1995) 14(15):3706–11. doi: 10.1002/j.1460-2075.1995.tb00040.x

113. Nakamura H, Iwakiri D, Ono Y, Fujiwara S. Epstein-Barr-virus-infected human T-cell line with a unique pattern of viral-gene expression. *Int J Cancer* (1998) 76(4):587–94. doi: 10.1002/(SICI)1097-0215(19980518)76:4<587::AID-IJC23>3.0.CO;2-3

114. Ho JWY, Li L, Wong KY, Srivastava G, Tao Q. Comprehensive profiling of EBV gene expression and promoter methylation reveals latency II viral infection and sporadic abortive lytic activation in peripheral T-cell lymphomas. *Viruses* (2023) 15(2):423. doi: 10.3390/v15020423

115. Brown JC. The role of DNA repair in herpesvirus pathogenesis. *Genomics* (2014) 104(4):287–94. doi: 10.1016/j.ygeno.2014.08.005

116. Sueur C, Lupo J, Mas P, Morand P, Boyer V. Difference in cytokine production and cell cycle progression induced by Epstein-Barr virus Lmp1 deletion variants in Kmh2, a Hodgkin lymphoma cell line. *Virol J* (2014) 11(1):94. doi: 10.1186/1743-422X-11-94

117. Peng RJ, Han BW, Cai QQ, Zuo XY, Xia T, Chen JR, et al. Genomic and transcriptomic landscapes of Epstein-Barr virus in extranodal natural killer T-cell lymphoma. *Leukemia* (2019) 33(6):1451–62. doi: 10.1038/s41375-018-0324-5

118. Takahashi E, Asano N, Li C, Tanaka T, Shimada K, Shimada S, et al. Nodal T/NK-cell lymphoma of nasal type: a clinicopathological study of six cases. *Histopathology* (2008) 52(5):585–96. doi: 10.1111/j.1365-2559.2008.02997.x

119. Liu Z, Sarathkumara YD, Chan JKC, Kwong YL, Lam TH, Ip DKM, et al. Characterization of the humoral immune response to the EBV proteome in extranodal NK/T-cell lymphoma. *Sci Rep* (2021) 11(1):23664. doi: 10.1038/s41598-021-02788-w

120. Montes-Mojarro IA, Fend F, Quintanilla-Martinez L. EBV and the pathogenesis of NK/T cell lymphoma. *Cancers* (2021) 13(6):1414. doi: 10.3390/cancers13061414

121. Schuster V, Ott G, Seidenspinner S, Kreth H. Common Epstein-Barr virus (EBV) type-1 variant strains in both Malignant and benign EBV-associated disorders. *Blood* (1996) 87(4):1579–85. doi: 10.1182/blood.V87.4.1579.bloodjournal8741579

122. Zanella L, Riquelme I, Buchegger K, Abanto M, Ili C, Brebi P. A reliable Epstein-Barr Virus classification based on phylogenomic and population analyses. *Sci Rep* (2019) 9(1):9829. doi: 10.1038/s41598-019-45986-3

123. Jung YJ, Choi H, Kim H, Lee SK. MicroRNA miR-BART20-5p Stabilizes Epstein-Barr Virus Latency by Directly Targeting *BZLF1* and *BRLF1*. Longnecker RM, editor. *J Virol* (2014) 88(16):9027–37. doi: 10.1128/JVI.00721-14

124. Saechan V, Mori A, Mitarnun W, Settheetham-Ishida W, Ishida T. Analysis of LMP1 variants of EBV in Southern Thailand: Evidence for strain-associated T-cell tropism and pathogenicity. *J Clin Virol* (2006) 36(2):119–25. doi: 10.1016/j.jcv.2006.01.018

125. Pereira J, Wilcox R, Lage LuísAC. *Challenges in peripheral T-cell lymphomas: from biological advances to clinical applicability*. Available at: https://www.frontiersin.org/research-topics/35284/challenges-in-peripheral-t-cell-lymphomas-from-biological-advances-to-clinical-applicability#overview.

126. Haque T, Taylor C, Wilkie GM, Murad P, Amlot PL, Beath S, et al. Complete regression of posttransplant lymphoproliferative disease using partially HLA-matched Epstein Barr virus-specific cytotoxic T cells. *Transplantation* (2001) 72(8):1399–402. doi: 10.1097/00007890-200110270-00012

127. Haque T, Wilkie GM, Jones MM, Higgins CD, Urquhart G, Wingate P, et al. Allogeneic cytotoxic T-cell therapy for EBV-positive posttransplantation lymphoproliferative disease: results of a phase 2 multicenter clinical trial. *Blood* (2007) 110(4):1123–31. doi: 10.1182/blood-2006-12-063008

128. Merlo A, Turrini R, Dolcetti R, Zanovello P, Amadori A, Rosato A. Adoptive cell therapy against EBV-related Malignancies: a survey of clinical results. *Expert Opin Biol Ther* (2008) 8(9):1265–94. doi: 10.1517/14712598.8.9.1265

129. Gottschalk S, Edwards OL, Sili U, Huls MH, Goltsova T, Davis AR, et al. Generating CTLs against the subdominant Epstein-Barr virus LMP1 antigen for the adoptive immunotherapy of EBV-associated Malignancies. *Blood* (2003) 101(5):1905–12. doi: 10.1182/blood-2002-05-1514

130. Murray RJ, Kurilla MG, Brooks JM, Thomas WA, Rowe M, Kieff E, et al. Identification of target antigens for the human cytotoxic T cell response to Epstein-Barr virus (EBV): implications for the immune control of EBV-positive Malignancies. *J Exp Med* (1992) 176(1):157–68. doi: 10.1084/jem.176.1.157

131. Adhikary D, Behrends U, Boerschmann H, Pfünder A, Burdach S, Moosmann A, et al. Immunodominance of lytic cycle antigens in epstein-barr virus-specific CD4+ T cell preparations for therapy. *PloS One* (2007) 2(7):e583. doi: 10.1371/journal.pone.0000583

132. Cho HI, Hong YS, Lee MA, Kim EK, Yoon SH, Kim CC, et al. Adoptive transfer of epstein-barr virus-specific cytotoxic T-lymphocytes for the treatment of angiocentric lymphomas. *Int J Hematol* (2006) 83(1):66–73. doi: 10.1532/IJH97.A30505

133. Cho SG, Kim N, Sohn HJ, Lee SK, Oh ST, Lee HJ, et al. Long-term outcome of extranodal NK/T cell lymphoma patients treated with postremission therapy using EBV LMP1 and LMP2a-specific CTLs. *Mol Ther* (2015) 23(8):1401–9. doi: 10.1038/mt.2015.91

134. Eom HS, Choi BK, Lee Y, Lee H, Yun T, Kim YH, et al. Phase I clinical trial of 4-1BB-based adoptive T-cell therapy for epstein-barr virus (EBV)-positive tumors. *J Immunother* (2016) 39(3):140–8. doi: 10.1097/CJI.0000000000000113

135. Kim WS, Oki Y, Kim SJ, Yoon SE, Ardeshna KM, Lin Y, et al. Autologous EBV-specific T cell treatment results in sustained responses in patients with advanced extranodal NK/T lymphoma: results of a multicenter study. *Ann Hematol* (2021) 100 (10):2529–39. doi: 10.1007/s00277-021-04558-0

136. Kimura H, Kwong YL. EBV viral loads in diagnosis, monitoring, and response assessment. *Front Oncol* (2019) 9:62. doi: 10.3389/fonc.2019.00062

137. Wang XX, Li PF, Bai B, Gao Y, Rong QX, Cai QQ, et al. Differential clinical significance of pre-, interim-, and post-treatment plasma Epstein–Barr virus DNA load in NK/T-cell lymphoma treated with P-GEMOX protocol. *Leukemia Lymphoma* (2019) 60(8):1917–25. doi: 10.1080/10428194.2018.1563690

138. Wang L, Wang H, Wang J-h, Xia Z-j, Lu Y, Huang H-q, et al. Post-treatment plasma EBV-DNA positivity predicts early relapse and poor prognosis for patients with extranodal NK/T cell lymphoma in the era of asparaginase. *Oncotarget* (2015) 6 (30):30317–26. doi: 10.18632/oncotarget.4505

139. Cho J, Kim SJ, Park S, Yoo KH, Ki CS, Ko Y, et al. Significance of circulating Epstein-Barr virus DNA monitoring after remission in patients with extranodal natural killer T cell lymphoma. *Ann Hematol* (2018) 97(8):1427–36. doi: 10.1007/s00277-018-3313-x

140. Kim HS, Kim KH, Kim KH, Chang MH, Ji SH, Lim DH, et al. Whole blood Epstein-Barr virus DNA load as a diagnostic and prognostic surrogate: extranodal natural killer/T-cell lymphoma. *Leukemia Lymphoma* (2009) 50(5):757–63. doi: 10.1080/10428190902803669

141. Gulley ML, Fan H, Elmore SH. Validation of roche lightCycler epstein-barr virus quantification reagents in a clinical laboratory setting. *J Mol Diagn* (2006) 8 (5):589–97. doi: 10.2353/jmoldx.2006.050152

142. Kanakry J, Ambinder R. The Biology and Clinical Utility of EBV Monitoring in Blood. In: Münz C, editor. *Epstein Barr Virus*, vol. 2 . Cham: Springer International Publishing (2015). p. 475–99. Current Topics in Microbiology and Immunology; vol. 391. doi: 10.1007/978-3-319-22834-1_17

143. Hewitt EW. The MHC class I antigen presentation pathway: strategies for viral immune evasion. *Immunology* (2003) 110(2):163–9. doi: 10.1046/j.1365-2567.2003.01738.x

144. Hegde N. Viral inhibition of MHC class II antigen presentation. *Trends Immunol* (2003) 24(5):278–85. doi: 10.1016/S1471-4906(03)00099-1

145. Tian C, Hromatka BS, Kiefer AK, Eriksson N, Noble SM, Tung JY, et al. Genome-wide association and HLA region fine-mapping studies identify susceptibility loci for multiple common infections. *Nat Commun* (2017) 8(1):599. doi: 10.1038/s41467-017-00257-5

146. Quinn LL, Zuo J, Abbott RJM, Shannon-Lowe C, Tierney RJ, Hislop AD, et al. Cooperation between Epstein-Barr Virus Immune Evasion Proteins Spreads Protection from CD8+ T Cell Recognition across All Three Phases of the Lytic Cycle. *PloS Pathog* (2014) 10(8):e1004322. doi: 10.1371/journal.ppat.1004322

147. Klein J, Sato A, Mackay IR, Rosen FS. The HLA System. *N Engl J Med* (2000) 343(10):702–9. doi: 10.1056/NEJM200009073431006

148. Sanchez-Mazas A. *A review of HLA allele and SNP associations with highly prevalent infectious diseases in human populations*. Swiss Med Wkly (2020) 150: w20214. Available at: https://smw.ch/index.php/smw/article/view/2757.

149. Noble JA, Valdes AM. Genetics of the HLA region in the prediction of type 1 diabetes. *Curr Diabetes Rep* (2011) 11(6):533–42. doi: 10.1007/s11892-011-0223-x

150. Chai H, Phipps M, Othman I, Tan L, Chua K. HLA variants rs9271366 and rs9275328 are associated with systemic lupus erythematosus susceptibility in Malays and Chinese. *Lupus* (2013) 22(2):198–204. doi: 10.1177/0961203312470183

151. Liu Z, Hildesheim A. Association between human leukocyte antigen class I and II diversity and non-virus-associated solid tumors. *Front Genet* (2021) 12:675860. doi: 10.3389/fgene.2021.675860

152. Ning L, Ko JMY, Yu VZ, Ng HY, Chan CKC, Tao L, et al. Nasopharyngeal carcinoma MHC region deep sequencing identifies HLA and novel non-HLA TRIM31 and TRIM39 loci. *Commun Biol* (2020) 3(1):759. doi: 10.1038/s42003-020-01487-y

153. Zhong C, Cozen W, Bolanos R, Song J, Wang SS. The role of HLA variation in lymphoma aetiology and survival. *J Intern Med* (2019) 2:12911.

154. Mesri EA, Feitelson MA, Munger K. Human viral oncogenesis: A cancer hallmarks analysis. *Cell Host Microbe* (2014) 15(3):266–82. doi: 10.1016/j.chom.2014.02.011

frontiers | Frontiers in Oncology

TYPE Review
PUBLISHED 22 September 2023
DOI 10.3389/fonc.2023.1241532

OPEN ACCESS

EDITED BY
Juliana Pereira,
University of São Paulo, Brazil

REVIEWED BY
Shih-Sung Chuang,
Chi Mei Medical Center, Taiwan
Suzanne Dawn Turner,
University of Cambridge, United Kingdom

*CORRESPONDENCE
Megan S. Lim
Limm4@mskcc.org

RECEIVED 16 June 2023
ACCEPTED 04 September 2023
PUBLISHED 22 September 2023

CITATION
Wu R and Lim MS (2023) Updates in pathobiological aspects of anaplastic large cell lymphoma.
Front. Oncol. 13:1241532.
doi: 10.3389/fonc.2023.1241532

Updates in pathobiological aspects of anaplastic large cell lymphoma

Rui Wu and Megan S. Lim*

Department of Pathology and Laboratory Medicine, Memorial Sloan Kettering Cancer Center, New York, NY, United States

Anaplastic large cell lymphomas (ALCL) encompass several distinct subtypes of mature T-cell neoplasms that are unified by the expression of CD30 and anaplastic cytomorphology. Identification of the cytogenetic abnormality t(2;5)(p23;q35) led to the subclassification of ALCLs into ALK+ ALCL and ALK- ALCL. According to the most recent World Health Organization (WHO) Classification of Haematolymphoid Tumours as well as the International Consensus Classification (ICC) of Mature Lymphoid Neoplasms, ALCLs encompass ALK+ ALCL, ALK- ALCL, and breast implant-associated ALCL (BI-ALCL). Approximately 80% of systemic ALCLs harbor rearrangement of *ALK*, with *NPM1* being the most common partner gene, although many other fusion partner genes have been identified to date. ALK- ALCLs represent a heterogeneous group of lymphomas with distinct clinical, immunophenotypic, and genetic features. A subset harbor recurrent rearrangement of genes, including *TYK2*, *DUSP22*, and *TP63*, with a proportion for which genetic aberrations have yet to be characterized. Although primary cutaneous ALCL (pc-ALCL) is currently classified as a subtype of primary cutaneous T-cell lymphoma, due to the large anaplastic and pleomorphic morphology together with CD30 expression in the malignant cells, this review also discusses the pathobiological features of this disease entity. Genomic and proteomic studies have contributed significant knowledge elucidating novel signaling pathways that are implicated in ALCL pathogenesis and represent candidate targets of therapeutic interventions. This review aims to offer perspectives on recent insights regarding the pathobiological and genetic features of ALCL.

KEYWORDS

pathogenesis, ALCL, lymphoma biology, genetic abnormalities, proteomics

1 Introduction

Anaplastic large cell lymphomas (ALCL) refer to a heterogeneous group of CD30-positive T-cell neoplasms with diverse clinical, histologic, and genetic features. The disease group comprises approximately 15% of all peripheral T-cell lymphoma and 3 to 5% of all non-Hodgkin lymphoma (1). ALCL was first recognized in 1985 based on the large size of the

neoplastic cells with uniform strong expression of CD30 (2). The recurrent chromosomal translocation t (2;5)(p23;q35), which was identified in 1994, results in a novel fusion tyrosine kinase involving the N-terminus of the nucleophosmin (*NPM1*) gene and the C-terminus of the anaplastic lymphoma kinase (*ALK*) gene (3). The chimeric protein NPM::ALK functions as an oncogenic tyrosine kinase which impacts diverse cellular signaling pathways leading to lymphoma. In addition to *NPM1*, over 20 distinct partner genes of *ALK* have been identified (Figure 1) (4). Moreover, the significantly enhanced survival of ALK+ ALCL patients and distinct genetic features rationalized its distinction from ALK- ALCLs (5, 6). Based on the recent understanding of the genetic basis of ALK- ALCL and those that occur in breast-implant ALCL, the updated 5th edition of the World Health Organization Classification of Haematolymphoid Tumours (5), and the International Consensus Classification (ICC) of Mature Lymphoid Neoplasms (7) recognize three subtypes, namely ALK+ ALCL, ALK- ALCL, breast implant-associated ALCL (BI-ALCL) (5). Further, we discuss the pathobiological features of primary cutaneous anaplastic large cell lymphoma (pc-ALCL) due to many overlapping histologic and immunophenotypic features with other ALCLs. Recent genomic studies provide an enhanced understanding of the pathobiological events in this group of intriguing neoplasms that will be summarized in this manuscript (6, 8).

2 Pathobiology of ALK+ ALCL

2.1 *ALK* rearrangements in ALK+ ALCL

ALK+ ALCLs, by definition, express the ALK protein, which functions as a strong driver oncogene. *ALK* gene rearrangements result in the expression of a novel fusion protein and constitutive activation of the ALK tyrosine kinase (9). The majority (80%) of cases express *NPM::ALK* as a result of t(2;5)(q23;35) involving the 3' segment of the *ALK* gene on chromosome 2p23 and the 5' segment of the nucleophosmin (*NPM1*) gene on chromosome 5q35. The

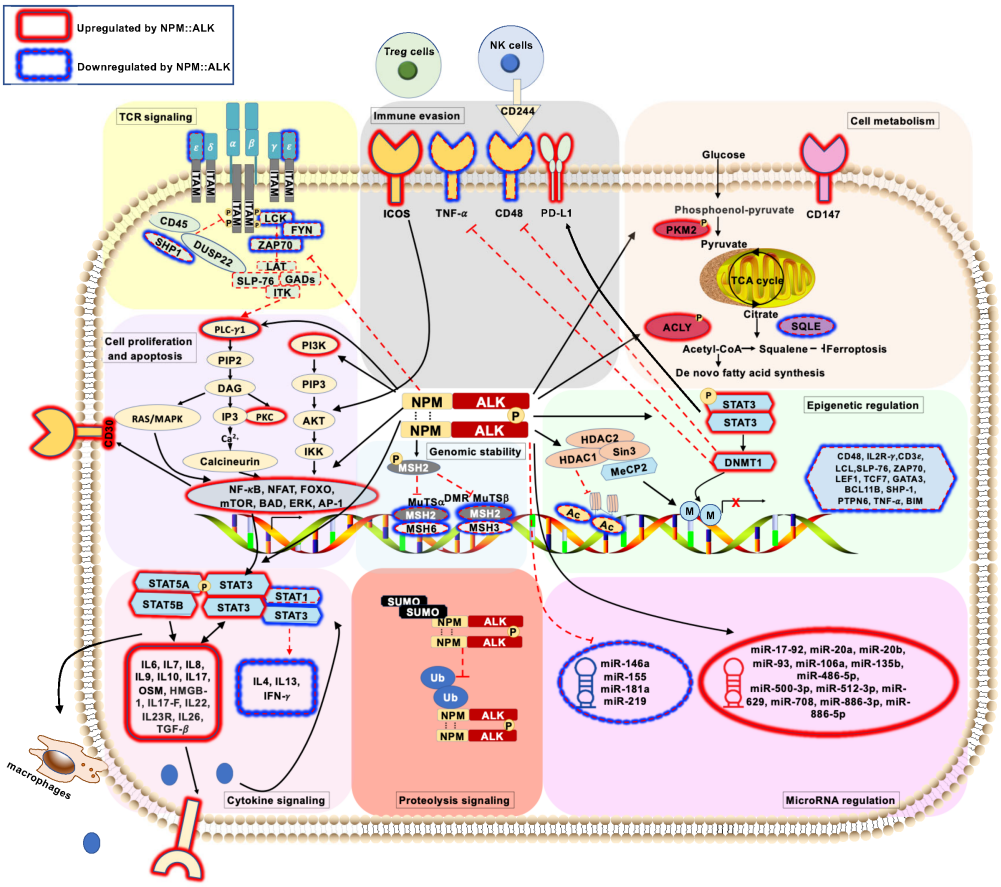


FIGURE 1
Schematic representation of pathogenic signaling pathways in ALK+ ALCL ( ) represents deregulated T-cell receptor signaling pathways mediated by *NPM::ALK*. ( ) represents deregulated cell proliferation and apoptosis signaling pathways mediated by *NPM::ALK*. ( ) represents deregulated cellular metabolism signaling pathways mediated by *NPM::ALK*. ( ) represents deregulated genomic stability signaling pathways mediated by *NPM::ALK*. ( ) represents deregulated epigenetic regulation signaling pathway mediated by *NPM::ALK*. ( ) represents deregulated miRNA repertoire by *NPM::ALK*. ( ) represents immune evasion mechanisms mediated by *NPM::ALK*. ( ) represents deregulated cytokine signaling mediated by *NPM::ALK*. ( ) represents deregulated proteolytic signaling mediated by *NPM::ALK*.

chimeric protein NPM::ALK is composed of the N-terminal oligomerization domain of NPM1 and the C-terminal tyrosine kinase domain of ALK. Due to ligand-independent oligomerization mediated by NPM1, the catalytic domain of ALK undergoes transautophosphorylation with constitutive tyrosine kinase activity, which translates into increased intracellular signaling promoting cell proliferation, resistance to apoptosis, and oncogenic transformation (10). Apart from *NPM1* as its N-terminal fusion partner, in approximately 20% of ALK+ ALCLs, *ALK* is fused with other genes, including tropomyosin 3 and 4 (*TPM3* and *TPM4*) (11), TRK-fused gene (*TFG*) (12), 5-amino-imidazole-4-carboxamide ribonucleotide formyltransferase/IMP cyclohydrolase (*ATIC*) (13), ring finger protein 213 (*RNF213*) (14), clathrin heavy chain (*CLTC*) (14), moesin (*MSN*) (15), non-muscle myosin heavy chain (*MYH9*) (15), TNF receptor-associated factor 1 (*TRAF1*) (16), eukaryotic translation elongation factor I gamma (*EEF1G*) (17) and the poly(A) binding protein cytoplasmic 1 (*PABPC1*) (18). In contrast to NPM::ALK, which is expressed in both the nucleus and cytoplasm, other ALK fusion proteins are localized in various cellular compartments (Table 1) (3, 11, 13–20). *TPM3::ALK*, the second most frequent fusion, is present in approximately 15% of ALK+ ALCLs. Despite all *ALK* rearrangements involving the same region of *ALK*, the downstream signaling pathways vary due to the different fusion partners (21). The pathogenetic mechanisms regulated by variant *ALK* fusion genes have not been explored due to the rarity of cases.

2.2 Deregulated T-cell receptor signaling pathway in ALK+ ALCL

T-cell receptor (TCR) engagement triggers various cascades of signaling pathways, including phospholipase C-γ1 (PLC-γ1)-inositol triphosphate (IP3)-Ca^{2+}-nuclear factor of activated T-cells (NFAT) pathway (22, 23), the protein kinase C (PKC)-IκB kinase (IKK)-nuclear factor (NF)-κB pathway (24), the Ras-extracellular signal-related kinase (ERK)-activator protein (AP)-1 pathway (25), as well as the Phosphoinositide 3-kinase (PI3K)-AKT-mammalian target of rapamycin (mTOR) pathway (26). These activated signaling pathways ultimately determine cell fate through cytokine production, cell survival, cell proliferation, and differentiation. Although ALK+ ALCLs express rearranged T-cell receptors, the expression of many pivotal TCR molecules, including TCF-1, TCF-1/LEF-1, LCK, ZAP-70, LAT, NFATc1, c-Jun, c-Fos, and Syk is repressed (27). Inhibition of the kinase activity of NPM::ALK or exposure to DNA methyltransferase inhibitors rescues the expression of CD3ε, ZAP70, LAT, and SLP76, suggesting that *NPM::ALK*-mediated transcriptional repression occurs via DNA methylation to downregulate components of the TCR signaling cascade in ALK+ ALCL (28). Apart from this, *NPM::ALK* further mimics TCR-induced signal transduction by directly interacting with and phosphorylating PLC-γ1, which triggers downstream signaling cascades for cell survival (29, 30). As such, NPM::ALK promotes the proliferation and survival of malignant cells by bypassing the TCR signaling pathway (Figure 1).

In addition to promoting cell proliferation and differentiation, T-cell receptor engagement also induces activation-induced T-cell death (AICD) through Fas-mediated apoptosis to prevent the accumulation of alloreactive T-cells and the development of graft-vs-host disease (31). It has been demonstrated that FLICE-like inhibitory protein (c-FLIP) prevents neoplastic cells from undergoing Fas-mediated apoptosis in ALK+ ALCL. Specifically, exposure of ALK+ ALCL cell lines that express high levels of c-FLIP, to CH-11, a CD95/FAS agonistic antibody, alone is not able to reduce the viability of malignant cells. si-RNA-mediated knockdown of *c-FLIP* together with CH-11 treatment rescues Fas-mediated apoptosis by triggering downstream caspase signaling pathways in ALK+ ALCL cells (32). Further investigation is required to determine whether the loss of TCR signaling molecules contributes to reduced AICD process in ALK+ ALCL.

TABLE 1 Summary of chromosomal rearrangements in ALK+ ALCL.

ALK fusion proteins	Translocation	Localization	Reference
NPM::ALK	t(2;5)(p23;q35)	nucleus, cytoplasm	(3)
TPM3::ALK	t(1;2)(q25; p23)	cytoplasm	(11)
TPM4::ALK	t(2;19)(p23;p13)	cytoplasm	(11)
TFG::ALK	t(2;3)(p23;q21)	cytoplasm	(12)
ATIC::ALK	inv(2)(p23q35)	cytoplasm	(13)
RNF213::ALK	t(2;17)(p23;q25)	cytoplasm	(14)
CLTC::ALK	t(2;17)(p23;q23)	cytoplasm	(14)
MSN::ALK	t(X;2)(q11; p23)	cytoplasm	(15)
MYH9::ALK	t(2;22)(p23;q11)	cytoplasm	(15)
TRAF1::ALK	t(2;19)(p23;q33)	cytoplasm	(16)
EEF1G::ALK	t(2;11)(p23;q11)	cytoplasm	(17)
PABPC1::ALK	t(2;9)(p23;q33)	cytoplasm	(18)

2.3 Deregulated cell proliferation and apoptosis in ALK+ ALCL

NPM::ALK regulates many proliferative and anti-apoptotic signaling pathways, including mitogen-activated protein (MAP) kinase (33), JAK-STAT (34, 35), PLC-γ1 (30), and PI3K-AKT (29), to promote lymphomagenesis (Figure 1). The MAP kinase signaling pathway is a major cell proliferation and survival regulator (36). NPM::ALK phosphorylates extracellular signal-regulated kinase (ERK)1/2 in a MEK1/2-dependent manner, and perturbation of MEK and ERK1/2 reduced cell proliferation and promoted cell apoptosis of ALK+ ALCL cells (37). NPM::ALK also regulates cell growth of ALK+ ALCL via activation of the PLC-γ pathway. The interaction of NPM::ALK with PLC-γ1 occurs via the tyrosine 644 residue, which is located at the C-terminus of the chimeric protein, and expression of NPM::ALK^{Y644F} abrogates PLC-γ1 phosphorylation and activation (30). The PI3K-AKT pathway has been implicated in oncogenesis for its role in cell-cycle progression. NPM::ALK constitutively activates PI3K and its downstream effector, the serine/threonine kinase AKT, and thus promotes growth and inhibits apoptosis in ALK+ALCL (38). AKT phosphorylates the Bcl2-associated death promoter (BAD), thereby suppressing apoptosis and promoting cell survival (39). Similarly, mTOR, a serine/threonine protein kinase and a key regulator of cell growth and proliferation, is also activated by NPM::ALK. Inhibition of the mTOR pathway leads to cell cycle arrest and apoptosis in ALK+ ALCL (40). Further, NPM::ALK promotes the expression of anti-apoptotic factors Bcl-xl and cell cycle-promoting cyclin-dependent kinase 4 (CDK4) and increased levels of phospho-RB to trigger cell proliferation (37). Thus, NPM::ALK regulates cell proliferation and survival while inhibiting apoptosis by orchestrating multiple signaling pathways.

2.4 Deregulated cellular metabolism in ALK+ ALCL

Neoplastic metabolic reprogramming is largely characterized by the shift from efficient energy-producing pathways to strategies for biomass production to support cell growth. In this regard, integrated analysis of the phosphoproteomic and metabolomic signature revealed that NPM::ALK signaling triggers an increase in biomass production by rerouting glycolytic intermediates (6) as well as modulation of lipid metabolism, amino acid metabolism, and nucleotide metabolism (Figure 1). Particularly, NPM::ALK-mediated phosphorylation of PKM2 at Y105, a key enzyme in aerobic glycolysis, leads to a metabolic switch promoting lymphomagenesis (6). Regarding lipid metabolism, NPM::ALK phosphorylates ATP citrate lyase (ACLY) at residue 682, which may serve as a switch to promote lipid synthesis required for cell proliferation. ACLY is a critical enzyme that catalyzes acetyl-CoA synthesis and connects vital biosynthetic pathways such as carbohydrate and lipid metabolism (41). Genetic or pharmacologic disruption in the NPM::ALK-ACLY signaling axis leads to impaired cell proliferation, impaired clonogenic potential, reduced tumor growth in an *in vivo* xenograft model, and attenuated lipid synthesis in ALK+ ALCL (42). NPM::ALK also modulates cancer metabolism through the downregulation of CD147, causing aberrant glycolysis and thus impairing the major energy source of tumor cells (43).

Further, metabolic alterations in cancer not only modulate the metabolic state of the cell but also impact cellular signaling and the epigenetic state. ALK+ ALCL-derived cell lines and primary tumors exhibit cholesterol auxotrophy due to reduced expression of a critical enzyme, squalene monooxygenase rendering the accumulation of squalene, a metabolite with antioxidant-like properties. Squalene monooxygenase oxidizes squalene to 2,3-oxidosqualene. Aggregation of squalene in cells prevents malignant cells from ferroptosis, which is induced by oxidative stress (44). Cholesterol auxotrophy of ALK+ ALCL can be a therapeutic vulnerability that can be utilized in combination with conventional therapies. In summary, NPM::ALK signaling orchestrates cellular metabolic reprogramming that favors lymphomagenesis.

2.5 Increased genomic instability in ALK+ ALCL

Under physiologic conditions, cells consistently encounter intracellular stress, such as reactive oxygen species (ROS) generated by cellular metabolism, and extracellular stress, such as UV light and carcinogenic chemicals. These stresses can disrupt genomic integrity. One of the hallmarks of cancer is genomic instability. It is generally accepted that *NPM::ALK* is required and sufficient to transform primary human T cells in a relatively short span of time. Furthermore, genetic alterations (single nucleotide variants) in ALK+ ALCL are relatively uncommon, suggesting a stable genome (45–47). In ALK+ ALCL, however, impaired DNA repair pathways (48), particularly DNA mismatch repair, may represent a mechanism by which tumor cells initiate additional genetic lesions, considering that a subset of ALK+ALCL patients develop resistance against ALK-specific treatment approaches. To initiate DNA mismatch repair, it is essential for the system to recognize DNA lesions. Two ATPase protein complexes participate in the mismatch recognition process, namely MuTSα, which identifies the base-base mismatches and small insertion/deletion, and MuTsβ, which modulates larger insertion/deletion. MuTSα is composed of MutS protein homolog 2 (MSH2) and MutS protein homolog 6 (MSH6). MuTSβ is composed of MSH2 and MutS protein homolog 3 (MSH3) (49). NPM::ALK directly binds to MSH2 and phosphorylates it at the tyrosine 238 residue. This abnormal interaction prevents MSH2 from binding to its normal partners, MSH3 and MSH6, leading to the ablation of normal MuTs complexes and consequentially impaired DNA damage repair (50). In addition, the expression of *NPM::ALK* in primary CD4+ T cells downregulates genes participating in DNA repair pathways (51). Thus, *NPM::ALK* ablates genomic stability by compromising the DNA mismatch repair process (Figure 1). Therefore, the role of *NPM::ALK* in promoting genomic instability by compromising the DNA repair process needs further study.

2.6 Epigenetic deregulation in ALK+ ALCL

Epigenetic modifications are heritable yet reversible covalent modifications in DNA or histones that alter the expression of genes without affecting DNA code. The most studied and significant modifications are the methylation of DNA at cytosine residues that function to repress gene expression (52) and the methylation or acetylation of distinct amino acids of the histone tail that dictate their repressive or activating properties (53). These modifications and their combinations dictate nucleosome positioning and local chromatin conformation that provide access to transcriptional regulators to modulate gene expression. DNA modifications direct histone modifications, and methylation of DNA causes steric hindrance to the transcriptional regulators (54). Moreover, the interaction of DNA with methyl-binding proteins, such as methyl CpG binding protein 2 (MeCP2), also prevents transcription factor binding at the locus causing repression of the target gene (52, 55).

NPM::ALK regulates the transcriptional silencing of many gene promoters and enhancer regions that encode tumor suppressors through its downstream effector transcription factors (Figure 1) (56). NPM::ALK activates the transcription factor STAT3, which upregulates DNA methyltransferase 1 (DNMT1) to methylate target genes for repression (57). As an example, *IL-2Rγ* promoter methylation is induced by *NPM::ALK*. *NPM::ALK* promotes STAT3 binding to the *IL-2Rγ* promoter, which then recruits DNMT1 to its promoter for its silencing (58). Notably, deleting *DNMT1* abrogates lymphomagenesis, suggesting a therapeutic opportunity for targeting DNMT1 in patients who develop resistance to ALK inhibitor treatment (59). ALK+ALCL also exhibits CpG Island methylation at *STAT5A*, a tumor suppressor that reciprocally suppresses *NPM::ALK* gene expression by binding to its enhancer (60). These results show that silencing of tumor suppressor genes by DNA methylation may contribute to the neoplastic transformation of ALK+ ALCL.

In addition to DNA methylation, gene expression is regulated by the chromatin remodeling machinery, which modulates accessibility of the chromatin. Deregulation of chromatin remodelers has been demonstrated to participate in cancer development as well as lymphomagenesis (61, 62). Among them, SWI/SNF is a multi-subunit chromatin remodeling complex that uses the energy generated by ATP hydrolysis to displace or evict nucleosomes and further regulates local chromatin conformation (63). Expression of BRM-Related Gene1 (BRG1), a core component of the human SWI/SNF complex (64), is mediated by NPM::ALK. Further, the expression of BRG1 is dependent on the kinase activity of NPM::ALK. Knockdown of *BRG1* in ALK+ ALCL cells results in a decrease in cell viability compared to scramble shRNA control (65). The role of other chromatin remodelers in the pathogenesis of ALK+ ALCL needs further investigation, and the relationship between the chromatin remodelers and *NPM::ALK* is still largely underexplored.

Loss of cellular identity is intrinsic to neoplastic transformation. ALCLs, despite originating from T-cells, exhibit downregulation of the transcriptional program that defines its T-cell phenotype. The pharmacologic treatment combining DNA demethylation and histone acetylation was insufficient to restore the T-cell phenotype in ALK+ ALCL cells. This suggests that additional stimulus is required to repress the T-cell phenotype. However, other T-cell lymphoma cells exposed to the same treatment exhibited expression of genes characteristic of ALCL (*ID2*, *LGALS4*, *c-JUN*) as well as loss of T-cell phenotype marked by loss of *CD3*, *LCK*, and *ZAP70* expression indicating that global DNA demethylation and histone acetylation are critical for cellular reprogramming towards an ALCL-like phenotype (66).

The combinatorial pattern of DNA methylation and histone post-translational modifications (PTMs) are increasingly appreciated as epigenetic signatures of cancer subtypes. These modifications regulate cellular processes, such as cell cycle regulation, apoptosis, and DNA damage response (67–70). *BCL2L11*, also known as *BIM* (Bcl-2 interacting mediator of cell death), a Bcl-2 homology 3 (BH3)-only proapoptotic protein that belongs to the Bcl-2 family, is epigenetically silenced via the combinatorial deregulation of DNA methylation and histone acetylation in ALK+ ALCL (66, 71). Recruitments of MeCP2 and Sin3a/histone deacetylase1/2 (HDAC1/2) corepressor complex to the *BIM* promoter contributes to its silencing. Exposure of the DNA methylase inhibitor, 5-azacytidine, or the HDAC inhibitor, trichostatin, alone to ALK+ ALCL cells is not only able to rescue the expression of BIM at both mRNA and protein level but also increases apoptosis (71). This suggests that DNA methylation and histone acetylation together may contribute to the pathogenesis of ALK+ ALCL. In this regard, the pharmacological modulation of altered epigenetic machinery may represent novel therapeutic interventions.

Histone PTMs alone can also dictate disease-specific changes in the transcriptional program, and the pattern of histone PTMs can be utilized as a novel biomarker of disease subtypes (72). HDAC inhibitors have already been approved by the Food and Drug Administration (FDA) for the treatment of T-cell malignancies (73–75). However, the comprehensive landscape of histone PTMs, such as methylation and acetylation (66, 76), phosphorylation (77), ubiquitination (78), and sumoylation (79) for different classes of ALCL is yet to be determined. Since the current therapeutic approach of using HDAC inhibitors has been shown to cause nonselective toxicity, further understanding of the comprehensive epigenetic landscape of ALCL is warranted as it may lead to discoveries of novel histone modifications and their writers and erasers, which can be targeted for precision therapeutics (80). Evaluation of ALCL subtypes with highly sensitive proteomic approaches for histone modification analysis as well as single-cell proteomic approaches with enrichment and analysis of histone PTMs will add significant value to define the epigenetic signature of the disease.

2.7 Deregulated MicroRNA repertoire in ALK+ ALCL

It has been observed that nearly 90% of the human genome is transcriptionally active, yet only 1.4% of this transcriptome is constituted by protein-coding mRNA (81). The role of non-

coding RNAs (ncRNA) is underappreciated yet critical to cell physiology and diseases, including ALCL. In ALK+ ALCL, the fusion protein NPM::ALK is associated with non-coding RNAs (ncRNAs), such as microRNAs to alter the gene expression signature of ALK+ ALCL (Figure 1) (82, 83). Along with tRNA and ribosomal RNA, the non-coding transcriptome is comprised of small nuclear RNA (snRNA), long noncoding RNA (lncRNA), and microRNA (miRNA). MicroRNAs are short, usually 20-23 nt long non-coding RNA that function by activating the RNA-induced silencing complex (RISC) against specific mRNA targets (84). miRNA array based on locked nucleic acid (LNA) technology containing 636 human and 425 murine miRNA probes performed on ALK+ ALCL cell lines identified distinct miRNA clusters from ALK+, to ALK- ALCL. These clusters are cross-validated with *Npm::alk* transgenic mice and primary ALK+ and ALK- ALCL to classify the miRNA unique to each disease group. These studies demonstrated strong upregulation of the miR-17-92 cluster in ALK + ALCL and miR-155 upregulation in ALK- ALCL. Further, reduced expression of miR-101 is observed in both ALK+ ALCL and ALK- ALCL (82). Subsequent studies identified 32 miRNAs associated with ALK expression *in vitro*, presenting distinct miRNA expression profiles (85). These studies identify 7 miRNAs, of which 5 are upregulated (miR-512-3p, miR-886-5p, miR-886-3p, miR-708, miR-135b) and 2 downregulated (miR-146a, miR-155) in ALK + ALCL. Another similar study identifies a distinct profile of miRNA that are specific to ALK+ or ALK- ALCL and cross-validated earlier findings. Moreover, it also identifies that miR-181a, which participates in the regulation of T-cell differentiation and TCR signaling, is significantly downregulated in ALK+ ALCL (86).

The role of exosomal miRNA in promoting disease dissemination of ALK+ ALCL has been recently reported. RNA sequencing studies identified 12 miRNAs that are significantly differentially expressed in the plasma of 20 NPM::ALK+ ALCL patients compared to healthy donors (n=5). Among these miRNAs, the level of miR-122-5p has further been validated as highly expressed in a larger cohort of ALCL patients (n=66) compared with healthy donors. Levels of miR-122-5p are elevated in late-stage (III-IV) ALCL patients compared to those with early-stage (I-II) disease. Interestingly, the expression of miR-122-5p is barely detectable in lymph nodes and other tissues but highly enriched in the liver of ALCL patients. *In vitro* and *in vivo* experiments indicate that miR-122-5p expressed in small extracellular vesicles promotes the proliferation and progression of ALCL cells (87). These mechanisms employed by miRNA using small extracellular vesicles for the pathogenesis of ALK+ ALCL may represent opportunities for discovery of novel mechanisms of disease dissemination as well as identification of prognostic biomarkers.

2.8 Immune evasion in ALK+ ALCL

Immune evasion by cancer cells is increasingly appreciated as an emerging hallmark of cancer. ALK+ ALCL cells exploit molecular mechanisms that bypass immune recognition (Figure 1). NPM::ALK-STAT3 signaling in ALK+ ALCL induces expression of transforming growth factor beta (TGF-β), IL-10, and cell surface receptor PD-L1 (CD274, B7H1), creating an immunosuppressive tumor microenvironment (88). The NPM::ALK-STAT3-DNMT1 pathway also epigenetically downregulates CD48, an immune surveillance molecule, to prevent tumor cell recognition by natural killer cells. STAT3 directly binds and methylates the promoter of *CD48* in association with DNMT1. Pharmacologic inhibition of NPM::ALK, STAT3, or DNMT1 sensitizes ALK+ ALCL towards NK cell-mediated cytotoxicity *in vitro*. Further, expression of CD48 in ALK+ ALCL cell line increases NK cell-mediated cytotoxicity *in vitro* and in a xenograft mouse model (89). Similarly, NPM::ALK-STAT3 pathway induces the expression of *ICOS*, a member of the CD28 costimulatory receptor superfamily, by transcriptional induction, as well as suppresses the ICOS inhibitor miR-219 (90). Since ICOS engagement promotes ALK+ ALCL proliferation, it is tempting to speculate that by engaging its ligand (ICOS-L), tumor-specific ICOS subverts other critical co-stimulatory signals from immune cells, impairing cytotoxic response to tumor cells. Previous studies suggest that ALK+ ALCLs and ALK+ ALCL cell lines, do not express TNF-α as a result of promoter methylation, thus preventing its proapoptotic function on tumor cells (91). Importantly, inhibition of DNMT1 by 5'-aza-2'-deoxy-cytidine (5-ADC) rescues the expression of *TNF-α* mRNA and protein. Further, exogenous TNF-α expression inhibits the growth of ALK+ ALCL cell lines and induces the activation of apoptotic pathway intermediates, namely caspase 8 and caspase 3. Hence, inhibition of DNMT1 not only triggers the NK cell-mediated cytotoxicity but also promotes the proapoptotic signaling pathway in ALK+ ALCL, raising the possibility of DNA methyltransferase inhibitors as a therapeutic option for ALK+ ALCL. The observation that the serum titers of anti-ALK antibodies in patients are inversely proportional to stage stratification and progression of disease indicates that NPM::ALK protein is immunogenic and triggers a natural immune response that keeps a check on disease progression to some extent (92). Therefore, it will be important to comprehensively investigate NPM::ALK-mediated immune escape mechanisms. A better understanding of the immune evasion mechanism will help in developing potential alternative or combinatorial therapeutic interventions for ALK+ ALCL.

2.9 Deregulation of transcription factors in ALK+ ALCL

Various models have been proposed for the origin of malignant cells in ALK+ ALCL. The expression of CD4 or CD8 and CD30, along with clonal T-cell receptor (TCR) rearrangement, suggests that the malignant cells may originate from activated T cells (93), while the expression of FoxP3, IL10, and TGFβ suggests a regulatory T cell origin (88), and BATF and BATF3 expression is associated with a Th17/group 3 innate lymphoid cell origin (94). In addition, *NPM::ALK*-transformed CD4+ T lymphocytes and primary ALK+ ALCL biopsies share characteristics with early T cell precursors (51). Further, ALK+ ALCL cells overexpress stem cell transcription factors (*OCT4*, *SOX2*, and *NANOG*) and *HIF2A*,

which regulate hematopoietic precursor differentiation and cell growth. These findings suggest that NPM::ALK signaling may trigger dedifferentiation to early thymic progenitor-like characteristics in CD30+ mature CD4+ T cells (95). In another study utilizing the RAG2$^{-/-}$ mice model, which lacks the machinery to produce mature T or B cells (96), it was shown that *NPM::ALK* is capable of promoting thymic T cell maturation and TCR-independent tumor formation, suggesting that the initial stage of ALK+ ALCL development may occur in the thymus (97).

Further, constitutive activation of STAT3 is highly prevalent in ALK+ ALCL and contributes to its pathogenesis. *NPM::ALK* interacts and phosphorylates STAT3 leading to its activation and nuclear translocation, where it regulates the transcription of a number of genes known to be involved in apoptosis and cell cycle progression (Figure 1) (35, 98). In ALK+ ALCL, the activation of STAT3 is multifactorial. JAK3, a major physiologic activator of STAT3, is highly activated in ALK+ ALCL lines and primary tumors (34). JAK3 interacts with NPM::ALK, and its inhibition decreases the tyrosine kinase activity of NPM::ALK (99, 100). Constitutive activation of STAT3 in ALK+ ALCL is also contributed by the downregulation of SH2 domain-containing protein tyrosine phosphatase-1 (SHP1) in ALK+ ALCL (101, 102). SHP1 interacts with JAK and NPM::ALK and dephosphorylate crucial tyrosine sites and thus inhibits the kinase activity (101, 103). ALK+ ALCL from children and adult patients exhibit loss of *SHP1* at a frequency of 50% and 86%, respectively. Further, *SHP1* is methylated and thus silenced in a number of ALK+ ALCL cases (101, 102).

ALK+ ALCL cells also aberrantly express multiple members of the activator protein-1 (AP-1) family of transcription factors, which includes proteins of the Jun, Fos, ATF, and Mf subfamilies (104). AP-1 family proteins regulate a wide range of cellular and biological activities, including cell cycle and proliferation, apoptosis, autophagy, and lipid synthesis (105). They also regulate cell migration and invasion as well as inflammatory response and immune cell development and activation. Studies have shown that AP-1 proteins play a pivotal role in promoting cell survival, proliferation, and suppression of AP-1 proteins can lead to apoptosis in ALK+ ALCL (94, 106, 107). Since AP-1 family proteins regulate a myriad of signaling pathways, further investigation will be required to comprehensively understand their impact on the ALK+ ALCL pathogenesis.

In addition, C/EBPβ, CCAAT enhancer binding protein, a transcription factor that belongs to the C/ECP leucine zipper transcription factor family, is highly expressed in ALK+ ALCLs (108). The overexpression of C/EBPβ is mediated through the NPM::ALK-STAT3 axis and is dependent on the kinase activity of NPM::ALK (108, 109). Moreover, NPM::ALK also fosters stability and translation of *C/EBPβ* mRNA via enhancing binding of AU-binding protein HuR to the 3′-UTR of *C/EBPβ* transcript (110). C/EBPβ modulates gene expression and miRNA levels to promote the transformation, proliferation, and survival of the malignant cells in ALK+ ALCL (111, 112). Therefore, targeting the deregulated transcription factors and the signaling pathways regulated by them may serve as novel therapeutic interventions for ALK+ ALCL.

2.10 Deregulated cytokine signaling in ALK+ ALCL

Cytokine and cytokine receptor signaling orchestrate the immune response, hematopoiesis, cell differentiation, and cell growth (113). There is an aberrant cytokine repertoire in ALK+ ALCL (Figure 1). Integrated unbiased N-glycoproteomic and transcriptomic profiling of 32 different B cell, T cell, and NK cell lymphoma cell lines has identified many cytokine receptors, including the interleukin receptor IL-R, as well as T helper (Th) receptors, expressed by ALK+ ALCL cells (8). Similarly, the level of IL-2R, Oncostatin M (OSM), IL-6, IL-8, IL-9, IL-10, IL-17a, IL-22, and soluble CD30 is decreased in either pediatric or adult ALK+ ALCL patient samples after they reached complete remission (114–116). There is a correlation between stages of the disease, presence of the minimal disseminated disease, anti-ALK antibody titers, and risk of relapse with concentrations of cytokines including IL-6, interferon-γ (IFN-γ), IFN-γ induced protein as well as sIL-2R among ALK+ ALCL pediatric patients (114). Moreover, levels of IL-6 demonstrated an independent prognostic value with a hazard ratio of 2.9 ± 0.4.

In addition, exogenous *NPM::ALK* expression leads to significant reductions of GM-CSF, TNF, and IL2 (51). Inhibition of NPM::ALK reduces the expression of cytokine receptor proteins, including IL-1R1, IL-1R2, IL-1RAP, IL-2RA, IL-4, IL-18RA, and IL-31RB (8). These observations suggest that constitutively activated ALK signaling contributes to deregulation of cytokine signaling.

Functional studies reveal that *NPM::ALK* regulates multiple JAK-STAT pathways, including IL-2/STAT5, and IL-6/STAT3 to participate in the aberrant cytokine secretion in ALK+ ALCL (8, 117). Particularly, *NPM::ALK* induces upregulation of *STAT3* and *STAT5* expression, which upregulates IL-31RB in ALK+ ALCL (118). In addition to STATs, *NPM::ALK* also enhances cytokine production by inducing the expression of other transcription factors, such as AP-1. AP-1 binds to promotors of multiple cytokines and thus regulates *IL17F, IL22, IL26, and IL23R* genes in ALK+ ALCL (94, 119).

Besides activation, *NPM::ALK* also deregulates the cytokine signaling pathway by suppressing transcription factor function. Among normal human endothelial cells, STAT1 is one of the major modulators of IFN-γ, which can further antagonize IL-6-mediated STAT3 activation (120). During activation, STAT1 forms a homodimer. It can also bind with *STAT3* and form a heterodimer. The gene expression levels and specificities are modulated by the STAT1 homodimer vs heterodimer ratio (121). In ALK+ ALCL, *NPM::ALK* also downregulates STAT1 to antagonize STAT3 and further decrease the production of antitumor cytokine IFN-γ (122).

Further, epigenetic modulation also contributes to cytokine deregulation in ALK+ ALCL. It has been reported that *NPM::ALK* downregulates SHP1 tyrosine phosphatase, a negative modulator of multiple cytokine signaling pathways, including Epo-R, IL-4, IL-13, IL-3R, IL-2R, through STAT3-mediated upregulation of DNA methyltransferase 1 in ALK+ ALCL (102, 123, 124).

The tumor microenvironment also contributes to the formation of deregulated cytokine repertoire (125, 126). However, the

composition and cross-talk between the neoplastic cells and tumor microenvironment of ALK+ ALCL need further investigation.

2.11 Deregulated proteolysis in ALK+ ALCL

Deregulated proteolysis by ubiquitination or sumoylation contributes significantly to the sustained signaling of oncogenic proteins (127, 128). The proteasomal degradation process of target proteins requires small ubiquitin binding to the substrate (127). Similarly, SUMOylation is another post-translational modification characterized by the reversible conjugation of small ubiquitin-like modifiers (SUMOs) with the target protein. SUMOylation modification often competes with ubiquitin for substrate binding and is believed to protect candidate proteins from proteasomal degradation (128). Studies suggest that the SUMOylating of NPM::ALK antagonizes its ubiquitination and subsequent degradation prolonging its oncogenic signaling (129). Further, the removal of sumoylation by SENP1 (a sentrin-specific family of proteases) promotes NPM::ALK protein turnover and ensues a decrease in cell viability, cell proliferation, and colony formation ability. It can be surmised that targeting NPM::ALK degradation may have therapeutic benefits in ALK+ ALCL that are resistant to NPM::ALK kinase inhibitors. In this regard, several efforts are underway to develop ALK protein degraders at different levels of preclinical or clinical settings (130–133).

3 Pathobiology of ALK- ALCL

ALK- ALCL is a CD30+ large T-cell lymphoma that typically affects the older population and has variable prognosis (134, 135). Currently, ALK- ALCL is subdivided into three classes, namely systemic ALCL, breast implant-associated ALCL, and primary cutaneous ALCL. Depending upon the genetic lesions acquired, the pathogenic mechanisms and disease aggressiveness may vary.

3.1 Pathobiology of systemic ALK- ALCL

In ALK- ALCL, two gene rearrangements and identified recurrent mutations subclassify ALK- ALCL into three more categories, namely fusion involving *DUSP22::IRF4*, fusions involving *TP63* gene, and other types of ALK- ALCL (Figure 2).

3.1.1 *DUSP22*-rearranged ALK- ALCL

Rearrangement of *DUSP22* occurs near the *DUSP22::IRF4* locus on 6p25.3 (136). The FRA7H fragile site on 7q32.3 is the most affected gene in the translocation t (6;7)(p25.3,q32.3). *DUSP22* rearrangements are detected at a frequency of 30% in ALK- ALCL cases using fluorescence *in situ* hybridization (FISH). This rearrangement leads to downregulation of DUSP22, and upregulation of the microRNA miR-29 on 7q32.3 but does not affect the expression of IRF4. This subgroup lacks the expression of genes associated with JAK-STAT3 signaling, but results in overexpression of the immunogenic cancer-testis antigen, marked DNA hypomethylation, and exhibits a reduced expression of PD-L1 and high expression of CD58 and HLA class II (137). Further, a novel recurrent mutation in MSC^{E116K}, a gene encoding musculin, has been recently identified in *DUSP22* rearranged ALK- ALCL. This mutation induces the CD30-IRF4-MYC signaling axis (Figure 2) and drives cell proliferation (138). Interestingly, DUSP22 inhibits interleukin-6 (IL-6)-induced STAT3 activation, and its downregulation is another mechanism by which STAT3 signaling may be activated in ALK- ALCL (139). Notably, the 5-year

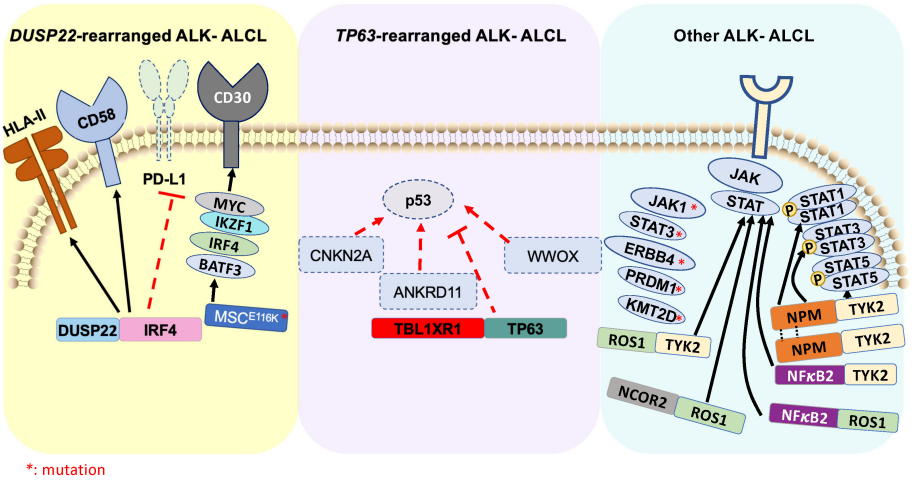


FIGURE 2
Schematic illustration of genomic rearrangements that contribute to pathogenesis of ALK- ALCL ( ) represents ALK- ALCL harboring *DUSP22*-rearrangement and associated signaling pathways. ( ) represents ALK- ALCL harboring *TP63*-rearrangement and associated signaling pathways. ( ) represents various driver oncogenic fusions involving tyrosine kinase and transcription factors, as well as high-frequency mutations that constitute an independent subgroup of ALK- ALCL.

overall survival of *DUSP22*-rearranged cases is approximately 85-90%, which is significantly higher than other ALK- ALCL (140).

3.1.2 *TP63*-rearranged ALK- ALCL

The *TP63* gene, a member of the p53 family, is expressed either as a full-length isoform carrying a transactivator domain (TP63) or as an amino-deleted isoform (ΔNp63) (141). P63 triggers multiple signaling in cancer-specific contexts, including regulation of the cell cycle, apoptosis, stemness, and tumorigenesis (142). Approximal 8% of ALK- ALCL show rearrangement of *TP63* in 3q28, frequently with *TBL1XR1* as a result of an inversion (3)(q26q28) (Figure 2) (143). Rearrangements involving *DUSP22* and *TP63* are mutually exclusive. *TP63*-rearranged ALK- ALCL exhibits the worst prognosis within the ALCL subtypes, with a 5-year overall survival rate of 17%. The biological significance of the genetic rearrangement of *TP63* in ALK- ALCL is yet to be determined.

3.1.3 Other types of ALK- ALCL

In ALK- ALCL, oncogenic mutations in *JAK1* and/or *STAT3* (Figure 2), which contribute to the consistent activation of the STAT3 signaling pathway, has been identified in nearly 20% of cases. In addition, oncogenic fusion genes involving a transcription factor and a tyrosine kinase, such as *NFκB2::ROS1*, *NCOR2::ROS1*, *NFκB2::TYK2*, and *PABPC4::TYK2* have been identified in ALK- ALCL (144). These fusion chimeras result in increased STAT3 activity and develop ALCL phenotype via STAT3 signaling, suggesting that intercepting STAT3 activation may have a therapeutic advantage (94). A recent deep-targeted next-generation sequencing of 47 ALK+ and 35 ALK- ALCL demonstrated that, on average, ALK- ALCL harbor 4.2 mutations/patient compared to 2.6 mutations/patient for ALK+ ALCL. Among all the mutations, *STAT3* and *JAK1* mutations are the most frequent (26%) in ALK- ALCL. The mutations that predicted poor prognosis of ALK- ALCL includes *TP53, STAT3, EPHA5, JAK1, PRDM1, LRP1B, and KMT2D* (46).

Approximately 25% of ALK- ALCL expresses an oncogenic truncated ERB-B2 receptor tyrosine kinase-4 (ERBB4) that is not detected in ALK+ ALCL and PTCL-NOS and may likely form another subgroup of ALK- ALCL. ERBB4 expression is mutually exclusive of *DUSP22*, *TP63*, and *ROS1* rearrangements. Pharmacologic inhibition of ERBB4 partly controls ALCL cell growth and disease progression in an ERBB4-positive patient-derived tumor graft model (145).

Hence, better understanding and targeting these rearrangements and mutation-mediated signaling pathways may serve as novel therapeutic interventions for different subtypes of ALK- ALCL.

3.2 Pathobiology of breast implant-associated ALCL

Breast implant-associated ALCL (BI-ALCL) is a distinct subtype of mature T-cell lymphoma. A persistent chronic inflammation occurring post-breast implants, particularly those with a textured outer shell, has been documented as the underlying cause of the disease (146). Cross-talk between the malignant cells and reactive cells in the microenvironment is thought to contribute to the formation of an inflammatory milieu characteristic of BI-ALCL. Elevated levels of IL-1β, IL-6, and TNF-α, the macrophage-activating cytokines, have been detected after culturing peripheral blood mononuclear cells obtained from healthy donors to the surfaces of the silicone breast implants for 4 days (147). However, no T-cell activation or specific effector cell subtype skewing has been observed. In addition, elevated expression of IL-13, IgE+ eosinophils, and mast cells in the microenvironment of primary BI-ALCL specimens suggests that allergic inflammation may contribute to the development of BI-ALCL (148).

Tumors display complex karyotypes with losses of chromosomes 1p,4q, 8p, 10p, 15, 16, 20 and gain of chromosomes 2, 9p, 12p, 19p, and 21 in BI-ALCL patients (149, 150). Targeted sequencing of 180 genes in 11 cases identified highly recurrent activating *STAT3* mutations and recurrent deletions of 1p22 involving *RPL5*, a tumor suppressor that regulates cell proliferation. In addition, abnormalities were identified in TGF-β, PKC, WNT/β-catenin pathway, and inflammasome signaling. Amplifications involving *TNFRSF11A* and *PDGFRA* were also identified (151). Genomic profiling of BI-ALCL using a variety of sequencing platforms did not detect any genomic rearrangements involving *ALK*, *DUSP22*, and *TP63*, suggesting less heterogeneity in the genetic manifestation than other subtypes of ALCL (152). Predominant JAK-STAT pathway, TP53, and DNMT3A could be molecular drivers of BI-ALCL (153). JAK1 mutations were found in 13% (3/23) of cases, with the most frequent point mutation involving G1097(D, V or S) identified in 44% (4/9) of cases (152, 154, 155). The frequency of *STAT3* mutations was 26% (6/23), with the most predominant mutations identified involving S614R (155). Apart from the JAK-STAT pathway, the second most frequent alterations in BI-ALCL were identified in epigenetic modifiers, including *TET2, TET3, ARID4B, KDM5C, KDM6A, KMT2C/D, CHD2, CREBBP* and *SMARCB* at the frequency of ~55-75% (150, 154). Currently, the first line of therapy involves surgical removal of the implant in combination with radiotherapy and standard chemotherapy. However, therapeutic targeting of JAK/STAT pathway and epigenetic deregulations may be considered as alternative therapeutic opportunities for BI-ALCL.

4 Pathobiology of primary cutaneous anaplastic large cell lymphoma

Primary cutaneous anaplastic large cell lymphoma (pc-ALCL) is a CD30+ lymphoproliferative disorder that manifests in the skin. The malignant cells exhibit large anaplastic and pleomorphic morphology with expression of CD30 in approximately 75% of cells. It has a relatively good prognosis in the absence of high-stage disease. The disease is currently classified as a subtype of primary cutaneous lymphoid proliferations and lymphomas that encompass a spectrum of other diseases, including lymphomatoid papulosis (LyP) (5). Morphologic features of pc-ALCL partly overlap with

other diseases, such as LyP (156–158) and reactive lymphoid hyperplasia (159). Therefore, genetic characterization of the disease is critical for correct diagnosis.

The majority of pc-ALCLs lack genomic rearrangements in *ALK, DUSP22*, and *TP63*. Although unusual, ALK-positive cases with only skin lesions have been identified, the frequency of these cases and ALK fusion partners are yet to be further determined (156, 160, 161). Array comparative genomic hybridization analysis of pc-ALCL demonstrates that nearly 40% of cases exhibit chromatin imbalances targeting region encompassing genes *RAF1* (3p25), *CTSB* (8p22), *FES* (15q26.1), *FGFR1* (8p11), *NRAS* (1p13.2), *MYCN* (2p24.1), and *CBFA2* (21q22.3) (162). Further, highly recurrent genomic loss of chromosomes 6q16-6q21, 6q27, and 13q34, as well as gain on the chromosome 7q31 and 17q, were also detected (163). In addition, a recurrent translocation involving *IRF4::MUM1* at chromosome 6p25.3 was identified at the frequency of approximately 20 to 25% in pc-ALCL. However, the protein expression of IRF4 and MUM1 is also detected in systemic ALCL, and therefore, examining the expression of IRF4 and MUM1 by IHC does not reliably distinguish pc-ALCL from systemic ALCL (157). Further, we identified a novel recurrent *NPM::TYK2* gene fusion in a proportion of primary cutaneous CD30+ lymphoproliferative disorders (15%), which activates STAT1/3/5 signaling and promotes cell proliferation (164). Importantly, a transgenic conditional knock-in Cd4-CreNPM::TYK2$^{fl/fl}$ mouse model demonstrates spontaneous development of CD30+ mature T-cell lymphoma with 90% penetrance (165). Hence, targeting TYK2 may serve as a therapeutic intervention for neoplasms harboring the *NPM::TYK2* rearrangement.

5 Conclusions and future perspectives

Our understanding of ALK+ ALCLs has provided opportunities for targeted therapies such as small molecular inhibitors of ALK (crizotinib, alectinib, and ceritinib) and antibody-drug conjugates targeting the tumor-specific expression of CD30. Given that a significant fraction of patients experience relapse or refractory responses, there is a continued need for the development of novel therapeutic approaches that target aberrant signaling and/or immune evasion mechanisms. ALK- ALCLs remain a genetically heterogeneous group of mature T-cell lymphoma. The identification of gene rearrangements involving *TYK2*, *DUSP22*, *TP63*, and *ERBB4*, and genetic alterations characteristic of distinct subsets of ALK- ALCLs, will facilitate improved stratification of disease outcomes. The discovery of novel gene rearrangements within the ALK- ALCL category and their functional consequences will be crucial for precision therapeutics. BI-ALCL demonstrates a predominant role of activated JAK-STAT3 signaling as the major driver of disease partly due to recurrent point mutations in *JAK1* and *STAT3*. Further, the contribution of epigenetic modifiers in conjunction with JAK-STAT3 signaling in the propagation of BI-ALCL has not been functionally explored and warrants further investigation. Moreover, an integrated approach of genetic, epigenetic, and proteomic profiling may offer an opportunity to identify novel therapeutic targets for ALCL. Despite studies that have identified the role of cytokine deregulation in ALCL, the composition of the microenvironment and its role in regulating tumor cell survival mechanisms remains largely unexplored.

Author contributions

RW and ML contributed to the ideas and structure of the manuscript. RW and ML wrote the review. All authors contributed to the article and approved the submitted version.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Vose JM, Neumann M, Harris ME. International peripheral T-cell and natural killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26(25):4124–30. doi: 10.1200/JCO.2008.16.4558

2. Falini B, Stein H, Pileri S, Canino S, Farabbi R, Martelli MF, et al. Expression of lymphoid-associated antigens on Hodgkin's and Reed-Sternberg cells of Hodgkin's disease. An immunocytochemical study on lymph node cytospins using monoclonal antibodies. *Histopathology* (1987) 11(12):1229–42. doi: 10.1111/j.1365-2559.1987.tb01869.x

3. Morris SW, Kirstein MN, Valentine MB, Dittmer KG, Shapiro DN, Saltman DL, et al. Fusion of a kinase gene, ALK, to a nucleolar protein gene, NPM, in non-Hodgkin's lymphoma. *Science* (1994) 263(5151):1281–4. doi: 10.1126/science.8122112

4. Duyster J, Bai RY, Morris SW. Translocations involving anaplastic lymphoma kinase (ALK). *Oncogene* (2001) 20(40 REV. ISS. 4):5623–37. doi: 10.1038/sj.onc.1204594

5. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IB de O, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: Lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

6. McDonnell SRP, Hwang SR, Rolland D, Murga-Zamalloa C, Basrur V, Conlon KP, et al. Integrated phosphoproteomic and metabolomic profiling reveals NPM-ALK–mediated phosphorylation of PKM2 and metabolic reprogramming in anaplastic large cell lymphoma. *Blood* (2013) 122(6):958. doi: 10.1182/blood-2013-01-482026

7. Campo E, Jaffe ES, Cook JR, Quintanilla-Martinez L, Swerdlow SH, Anderson KC, et al. The international consensus classification of mature lymphoid neoplasms: a report from the clinical advisory committee. *Blood* (2022) 140(11):1229–53. doi: 10.1182/blood.2022015851

8. Rolland DCM, Basrur V, Jeon YK, McNeil-Schwalm C, Fermin D, Conlon KP, et al. Functional proteogenomics reveals biomarkers and therapeutic targets in lymphomas. *Proc Natl Acad Sci* (2017) 114(25):6581–6586. doi: 10.1073/pnas.1701263114

9. Andraos E, Dignac J, Meggetto F. NPM-ALK: A driver of lymphoma pathogenesis and a therapeutic target. *Cancers* (2021) 13:144. doi: 10.3390/cancers13010144

10. Marzec M, Kasprzycka M, Liu X, El-Salem M, Halasa K, Raghunath PN, et al. Oncogenic tyrosine kinase NPM/ALK induces activation of the rapamycin-sensitive mTOR signaling pathway. *Oncogene* (2007) 26(38):5606–14. doi: 10.1038/sj.onc.1210346

11. Lawrence B, Perez-Atayde A, Hibbard MK, Rubin BP, Cin PD, Pinkus JL, et al. TPM3-ALK and TPM4-ALK oncogenes in inflammatory myofibroblastic tumors. *Am J Pathol* (2000) 157(2):377–84. doi: 10.1016/S0002-9440(10)64550-6

12. Hernández L, Pinyol M, Hernández S, Beà S, Pulford K, Rosenwald A, et al. TRK-fused gene (TFG) is a new partner of ALK in anaplastic large cell lymphoma producing two structurally differentTFG-ALK translocations. *Blood* (1999) 94(9):3265–8. doi: 10.1182/blood.V94.9.3265

13. Colleoni GWB, Bridge JA, Garicochea B, Liu J, Filippa DA, Ladanyi M. ATIC-ALK: A novel variant ALK gene fusion in anaplastic large cell lymphoma resulting from the recurrent cryptic chromosomal inversion, inv(2)(p23q35). *Am J Pathol* (2000) 156 (3):781–9. doi: 10.1016/S0002-9440(10)64945-0

14. Cools J, Wlodarska I, Somers R, Mentens N, Pedeutour F, Maes B, et al. Identification of novel fusion partners of ALK, the anaplastic lymphoma kinase, in anaplastic large-cell lymphoma and inflammatory myofibroblastic tumor. *Genes Chromosomes Cancer* (2002) 34(4):354–62. doi: 10.1002/gcc.10033

15. Armstrong F, Duplantier MM, Trempat P, Hieblot C, Lamant L, Espinos E, et al. Differential effects of X-ALK fusion proteins on proliferation, transformation, and invasion properties of NIH3T3 cells. *Oncogene* (2004) 23:36. doi: 10.1038/sj.onc.1207813

16. Feldman AL, Vasmatzis G, Asmann YW, Davila J, Middha S, Eckloff BW, et al. Novel TRAF1-ALK fusion identified by deep RNA sequencing of anaplastic large cell lymphoma. *Genes Chromosomes Cancer* (2013) 52(11):1097–102. doi: 10.1002/gcc.22104

17. Palacios G, Shaw TI, Li Y, Singh RK, Valentine M, Sandlund JT, et al. Novel ALK fusion in anaplastic large cell lymphoma involving EEF1G, a subunit of the eukaryotic elongation factor-1 complex. *Leukemia* (2017) 31(3):743–7. doi: 10.1038/leu.2016.331

18. Graetz D, Crews KR, Azzato EM, Singh RK, Raimondi S, Mason J, et al. Leukemic presentation of ALK-positive anaplastic large cell lymphoma with a novel partner, poly(A) binding protein cytoplasmic 1 (PABPC1), responding to single-agent crizotinib. *Haematologica* (2019) 104(5):e218. doi: 10.3324/haematol.2018.215103

19. Amano Y, Ishikawa R, Sakatani T, Ichinose J, Sunohara M, Watanabe K, et al. Oncogenic TPM3-ALK activation requires dimerization through the coiled-coil structure of TPM3. *Biochem Biophys Res Commun* (2015) 457(3):457–60. doi: 10.1016/j.bbrc.2015.01.014

20. Shi JH, Ling C, Wang TT, Zhang LN, Liu WW, Qin Y, et al. TRK-fused gene (TFG) regulates ULK1 stability via TRAF3-mediated ubiquitination and protects macrophages from LPS-induced pyroptosis. *Cell Death Dis* (2022) 13(1):93. doi: 10.1038/s41419-022-04539-9

21. Chiarle R, Voena C, Ambrogio C, Piva R, Inghirami G. The anaplastic lymphoma kinase in the pathogenesis of cancer. *Nat Rev Cancer* (2008) 8(1):11–23. doi: 10.1038/nrc2291

22. Granja C, Lin LL, Yunis EJ, Relias V, Dasgupta JD. PLC gamma 1, a possible mediator of T cell receptor function. *J Biol Chem* (1991) 266(25):16277–80. doi: 10.1016/S0021-9258(18)55290-X

23. Gonen R, Beach D, Ainey C, Yablonski D. T cell receptor-induced activation of phospholipase C-γ1 depends on a sequence-independent function of the P-I region of SLP-76. *J Biol Chem* (2005) 280(9):8364–70. doi: 10.1074/jbc.M409437200

24. Li Y, Sedwick CE, Hu J, Altman A. Role for protein kinase Ctheta (PKCtheta) in TCR/CD28-mediated signaling through the canonical but not the non-canonical pathway for NF-kappaB activation. *J Biol Chem* (2005) 280(2):1217–23. doi: 10.1074/jbc.M409492200

25. Downward J, Graves JD, Warne PH, Rayter S, Cantrell DA. Stimulation of p21ras upon T-cell activation. *Nature* (1990) 346(6286):719–23. doi: 10.1038/346719a0

26. Colombetti S, Basso V, Mueller DL, Mondino A. Prolonged TCR/CD28 engagement drives IL-2-independent T cell clonal expansion through signaling mediated by the mammalian target of rapamycin. *J Immunol* (2006) 176(5):2730–8. doi: 10.4049/jimmunol.176.5.2730

27. Geissinger E, Sadler P, Roth S, Grieb T, Puppe B, Müller N, et al. Disturbed expression of the T-cell receptor/CD3 complex and associated signaling molecules in CD30+ T-cell lymphoproliferations. *Haematologica* (2010) 95(10):1697. doi: 10.3324/haematol.2009.021428

28. Ambrogio C, Martinengo C, Voena C, Tondat F, Riera L, di Celle PF, et al. NPM-ALK oncogenic tyrosine kinase controls T-cell identity by transcriptional regulation and epigenetic silencing in lymphoma cells. *Cancer Res* (2009) 69(22):8611–9. doi: 10.1158/0008-5472.CAN-09-2655

29. Bai RY, Ouyang T, Miething C, Morris SW, Peschel C, Duyster J. Nucleophosmin–anaplastic lymphoma kinase associated with anaplastic large-cell lymphoma activates the phosphatidylinositol 3-kinase/Akt antiapoptotic signaling pathway. *Blood* (2000) 96(13):4319–27. doi: 10.1182/blood.V96.13.4319

30. Bai RY, Dieter P, Peschel C, Morris SW, Duyster J. Nucleophosmin-anaplastic lymphoma kinase of large-cell anaplastic lymphoma is a constitutively active tyrosine kinase that utilizes phospholipase C-gamma to mediate its mitogenicity. *Mol Cell Biol* (1998) 18(12):6951–61. doi: 10.1128/MCB.18.12.6951

31. Green DR, Droin N, Pinkoski M. Activation-induced cell death in T cells. *Immunol Rev* (2003) 193:70–81. doi: 10.1034/j.1600-065X.2003.00051.x

32. Oyarzo MP, Medeiros LJ, Atwell C, Feretzaki M, Leventaki V, Drakos E, et al. c-FLIP confers resistance to FAS-mediated apoptosis in anaplastic large-cell lymphoma. *Blood* (2006) 107(6):2544–7. doi: 10.1182/blood-2005-06-2601

33. Staber PB, Vesely P, Haq N, Ott RG, Funato K, Bambach I, et al. The oncoprotein NPM-ALK of anaplastic large-cell lymphoma induces JUNB transcription via ERK1/2 and JunB translation via mTOR signaling. *Blood* (2007) 110(9):3374–83. doi: 10.1182/blood-2007-02-071258

34. Lai R, Rassidakis G, Lin Q, Atwell C, Medeiros L, Amin H. Jak3 activation is significantly associated with ALK expression in anaplastic large cell lymphoma. *Hum Pathol* (2005) 36(9):939–44. doi: 10.1016/j.humpath.2005.07.011

35. Zamo A, Chiarle R, Piva R, Howes J, Fan Y, Chilosi M, et al. Anaplastic lymphoma kinase (ALK) activates Stat3 and protects hematopoietic cells from cell death. *Oncogene* (2002) 21(7):1038–47. doi: 10.1038/sj.onc.1205152

36. Wei Z, Liu HT. MAPK signal pathways in the regulation of cell proliferation in mammalian cells. *Cell Res* (2002) 12:1. doi: 10.1038/sj.cr.7290105

37. Marzec M, Kasprzycka M, Liu X, Raghunath PN, Wlodarski P, Wasik MA. Oncogenic tyrosine kinase NPM/ALK induces activation of the MEK/ERK signaling pathway independently of c-Raf. *Oncogene* (2007) 26:6. doi: 10.1038/sj.onc.1209843

38. Slupianek A, Nieborowska-Skorska M, Hoser G, Morrione A, Majewski M, Xue L, et al. Role of phosphatidylinositol 3-kinase-Akt pathway in nucleophosmin/anaplastic lymphoma kinase-mediated lymphomagenesis. *Cancer Res* (2001) 61 (5):2194–2199

39. Datta SR, Dudek H, Xu T, Masters S, Haian F, Gotoh Y, et al. Akt phosphorylation of BAD couples survival signals to the cell-intrinsic death machinery. *Cell* (1997) 91(2):231–41. doi: 10.1016/S0092-8674(00)80405-5

40. Gao J, Yin M, Zhu Y, Gu L, Zhang Y, Li Q, et al. Prognostic significance and therapeutic potential of the activation of anaplastic lymphoma kinase/protein kinase B/mammalian target of rapamycin signaling pathway in anaplastic large cell lymphoma. *BMC Cancer* (2013) 13(1):471. doi: 10.1186/1471-2407-13-471

41. Icard P, Wu Z, Fournel L, Coquerel A, Lincet H, Alifano M. ATP citrate lyase: A central metabolic enzyme in cancer. *Cancer Lett* (2020) 471:125–34. doi: 10.1016/j.canlet.2019.12.010

42. Basappa J, ElAzzouny MA, Rolland DCM, Sahasrabuddhe AA, Ma K, Bazilevsky GA, et al. Tyrosine phosphorylation is critical for ACLY activity in lipid metabolism and cancer. *bioRxiv* (2020). doi: 10.1101/2020.01.20.910752

43. Kim JY, Kim H, Suk K, Lee WH. Activation of CD147 with cyclophilin a induces the expression of IFITM1 through ERK and PI3K in THP-1 cells. *Mediators Inflammation* (2010) 2010:821940. doi: 10.1155/2010/821940

44. Garcia-Bermudez J, Baudrier L, Bayraktar EC, Shen Y, La K, Guarecuco R, et al. Squalene accumulation in cholesterol auxotrophic lymphomas prevents oxidative cell death. *Nature* (2019) 567:7746. doi: 10.1038/s41586-019-0945-5

45. Salaverria I, Beà S, Lopez-Guillermo A, Lespinet V, Pinyol M, Burkhardt B, et al. Genomic profiling reveals different genetic aberrations in systemic ALK-positive and ALK-negative anaplastic large cell lymphomas. *Br J Haematol* (2008) 140(5):516–26. doi: 10.1111/j.1365-2141.2007.06924.x

46. Lobello C, Tichy B, Bystry V, Radova L, Filip D, Mraz M, et al. STAT3 and TP53 mutations associate with poor prognosis in anaplastic large cell lymphoma. *Leukemia* (2021) 35(5):1500–5. doi: 10.1038/s41375-020-01093-1

47. Youssif C, Goldenbogen J, Hamoudi R, Carreras J, Viskaduraki M, Cui YX, et al. Genomic profiling of pediatric ALK-positive anaplastic large cell lymphoma: a Children's Cancer and Leukaemia Group Study. *Genes Chromosomes Cancer* (2009) 48(11):1018–26. doi: 10.1002/gcc.20701

48. Larose H, Prokoph N, Matthews JD, Schlederer M, Högler S, Alsulami AF, et al. Whole Exome Sequencing reveals NOTCH1 mutations in anaplastic large cell lymphoma and points to Notch both as a key pathway and a potential therapeutic target. *Haematologica* (2021) 106(6):1693–704. doi: 10.3324/haematol.2019.238766

49. Lyer RR, Pluciennik A, Burdett V, Modrich PL. DNA mismatch repair: functions and mechanisms. *Chem Rev* (2006) 106(2):302–23. doi: 10.1021/cr0404794

50. Lobello C, Bikos V, Janikova A, Pospisilova S. The role of oncogenic tyrosine kinase NPM-ALK in genomic instability. *Cancers (Basel)* (2018) 10(3):64. doi: 10.3390/cancers10030064

51. Pawlicki JM, Cookmeyer DL, Maseda D, Everett JK, Wei F, Kong H, et al. NPM-ALK-induced reprogramming of mature TCR-stimulated T cells results in dedifferentiation and Malignant transformation. *Cancer Res* (2021) 81(12):3241–54. doi: 10.1158/0008-5472.CAN-20-2297

52. Jin B, Li Y, Robertson KD. DNA methylation: superior or subordinate in the epigenetic hierarchy? *Genes Cancer* (2011) 2(6):607. doi: 10.1177/1947601910393957

53. Millán-Zambrano G, Burton A, Bannister AJ, Schneider R. Histone post-translational modifications — cause and consequence of genome function. *Nat Rev Genet* (2022) 23(9):563–80. doi: 10.1038/s41576-022-00468-7

54. Cedar H, Bergman Y. Linking DNA methylation and histone modification: patterns and paradigms. *Nat Rev Genet* (2009) 10(5):295–304. doi: 10.1038/nrg2540

55. Nan X, Campoy FJ, Bird A. MeCP2 is a transcriptional repressor with abundant binding sites in genomic chromatin. *Cell* (1997) 88(4):471–81. doi: 10.1016/S0092-8674(00)81887-5

56. Hassler MR, Pulverer W, Lakshminarasimhan R, Redl E, Hacker J, Garland GD, et al. Insights into the Pathogenesis of Anaplastic Large-Cell Lymphoma through Genome-wide DNA Methylation Profiling. *Cell Rep* (2016) 17(2):596–608. doi: 10.1016/j.celrep.2016.09.018

57. Zhang Q, Wang HY, Woetmann A, Raghunath PN, Odum N, Wasik MA. STAT3 induces transcription of the DNA methyltransferase 1 gene (DNMT1) in Malignant T lymphocytes. *Blood* (2006) 108(3):1058–64. doi: 10.1182/blood-2005-08-007377

58. Zhang Q, Wang HY, Liu X, Bhutani G, Kantekure K, Wasik M. IL-2R common gamma-chain is epigenetically silenced by nucleophosphin-anaplastic lymphoma kinase (NPM-ALK) and acts as a tumor suppressor by targeting NPM-ALK. *Proc Natl Acad Sci USA* (2011) 108(29):11977–82. doi: 10.1073/pnas.1100319108

59. Redl E, Sheibani-Tezerji R, Cardona CDJ, Hamminger P, Timelthaler G, Hassler MR, et al. Requirement of DNMT1 to orchestrate epigenomic reprogramming for NPM-ALK-driven lymphomagenesis. *Life Sci Alliance* (2020) 4(2):e202000794. doi: 10.26508/lsa.202000794

60. Zhang Q, Wang HY, Liu X, Wasik MA. STAT5A is epigenetically silenced by the tyrosine kinase NPM1-ALK and acts as a tumor suppressor by reciprocally inhibiting NPM1-ALK expression. *Nat Med* (2007) 13(11):1341–8. doi: 10.1038/nm1659

61. Wu R. Untangling the epigenetic imbalance in B cell lymphoma. *Curr Pharmacol Rep* (2020) 6(3):110–20. doi: 10.1007/s40495-020-00214-9

62. Nair SS, Kumar R. Chromatin remodeling in Cancer: A Gateway to regulate gene Transcription. *Mol Oncol* (2012) 6(6):611. doi: 10.1016/j.molonc.2012.09.005

63. Mittal P, Roberts CWM. The SWI/SNF complex in cancer — biology, biomarkers and therapy. *Nat Rev Clin Oncol* (2020) 17(7):435–48. doi: 10.1038/s41571-020-0357-3

64. Mardinian K, Adashek JJ, Botta GP, Kato S, Kurzrock R. SMARCA4: Implications of an altered chromatin-remodeling gene for cancer development and therapy. *Mol Cancer Ther* (2021) 20(12):2341–51. doi: 10.1158/1535-7163.MCT-21-0433

65. Garland GD, Ducray SP, Jahangiri L, Pucci P, Burke GAA, Monahan J, et al. Brg1 and npm-alk are co-regulated in anaplastic large-cell lymphoma; brg1 is a potential therapeutic target in alcl. *Cancers (Basel)* (2022) 14(1):151. doi: 10.3390/cancers14010151

66. Joosten M, Seitz V, Zimmermann K, Sommerfeld A, Berg E, Lenze D, et al. Histone acetylation and DNA demethylation of T cells result in an anaplastic large cell lymphoma-like phenotype. *Haematologica* (2013) 98(2):247–54. doi: 10.3324/haematol.2011.054619

67. Shilatifard A. The COMPASS family of histone H3K4 methylases: Mechanisms of regulation in development and disease pathogenesis. *Annu Rev Biochem* (2012) 81(1):65–95. doi: 10.1146/annurev-biochem-051710-134100

68. Cloos PAC, Christensen J, Agger K, Helin K. Erasing the methyl mark: histone demethylases at the center of cellular differentiation and disease. *Genes Dev* (2008) 22(9):1115–40. doi: 10.1101/gad.1652908

69. D'Oto A, Tian Q, Davidoff AM, Yang J. Histone demethylases and their roles in cancer epigenetics. *J Med Oncol Ther* (2016) 01(02):34–40. doi: 10.35841/medical-oncology.1.2.34-40

70. Haery L, Thompson RC, Gilmore TD. Histone acetyltransferases and histone deacetylases in B- and T-cell development, physiology and Malignancy. *Genes Cancer Impact Journals LLC;* (2015) 6:184–213. doi: 10.18632/genesandcancer.65

71. Piazza R, Magistroni V, Mogavero A, Andreoni F, Ambrogio C, Chiarle R, et al. Epigenetic silencing of the proapoptotic gene BIM in anaplastic large cell lymphoma through an meCP2/SIN3a deacetylating complex. *Neoplasia* (2013) 15(5):511–IN17. doi: 10.1593/neo.121784

72. Onder O, Wu R, Wysocka M, Haun PL, Kim E, Garcia BA, et al. Epiproteomic landscape and histone code of cutaneous T-cell lymphoma/sézary syndrome. *Blood* (2018) 132(Supplement 1):780. doi: 10.1182/blood-2018-99-115347

73. Suraweera A, O'Byrne KJ, Richard DJ. Combination therapy with histone deacetylase inhibitors (HDACi) for the treatment of cancer: Achieving the full therapeutic potential of HDACi. *Front Oncol* (2018) 8(MAR). doi: 10.3389/fonc.2018.00092

74. Jones PA, Issa JPJ, Baylin S. Targeting the cancer epigenome for therapy. *Nat Rev Genet* (2016) 17(10):630–41. doi: 10.1038/nrg.2016.93

75. Guha M. HDAC inhibitors still need a home run, despite recent approval. *Nat Rev Drug Discovery* (2015) 14(4):225–6. doi: 10.1038/nrd4583

76. Zhang P, Zhang M. Epigenetic alterations and advancement of treatment in peripheral T-cell lymphoma. *Clin Epigenet* (2020) 12(1):169. doi: 10.1186/s13148-020-00962-x

77. Pérez-Cadahía B, Drobic B, Davie JR. H3 phosphorylation: dual role in mitosis and interphase. *Biochem Cell Biol* (2009) 87(5):695–709. doi: 10.1139/O09-053

78. Weake VM, Workman JL. Histone ubiquitination: triggering gene activity. *Mol Cell* (2008) 29(6):653–63. doi: 10.1016/j.molcel.2008.02.014

79. Shiio Y, Eisenman RN. Histone sumoylation is associated with transcriptional repression. *Proc Natl Acad Sci USA* (2003) 100(23):13225–30. doi: 10.1073/pnas.1735528100

80. Li W, Sun Z. Mechanism of action for HDAC inhibitors-insights from omics approaches. *Int J Mol Sci* (2019) 20(7):1616. doi: 10.3390/ijms20071616

81. Baltimore D. Our genome unveiled. *Nature* (2001) 409(6822):814–6. doi: 10.1038/35057267

82. Merkel O, Hamacher F, Laimer D, Sifft E, Trajanoski Z, Scheideler M, et al. Identification of differential and functionally active miRNAs in both anaplastic lymphoma kinase (ALK)+ and ALK- anaplastic large-cell lymphoma. *Proc Natl Acad Sci USA* (2010) 107(37):16228–33. doi: 10.1073/pnas.1009719107

83. Fuchs S, Naderi J, Meggetto F. Non-coding RNA networks in ALK-positive anaplastic-large cell lymphoma. *Int J Mol Sci* (2019) 20:2150. doi: 10.3390/ijms20092150

84. Gregory RI, Chendrimada TP, Cooch N, Shiekhattar R. Human RISC couples microRNA biogenesis and posttranscriptional gene silencing. *Cell* (2005) 123(4):631–40. doi: 10.1016/j.cell.2005.10.022

85. Liu C, Iqbal J, Teruya-Feldstein J, Shen Y, Dabrowska MJ, Dybkaer K, et al. MicroRNA expression profiling identifies molecular signatures associated with anaplastic large cell lymphoma. *Blood* (2013) 122(12):2083–92. doi: 10.1182/blood-2012-08-447375

86. Steinhilber J, Bonin M, Walter M, Fend F, Bonzheim I, Quintanilla-Martinez L. Next-generation sequencing identifies deregulation of microRNAs involved in both innate and adaptive immune response in ALK+ ALCL. *PloS One* (2015) 10(2):e0117780. doi: 10.1371/journal.pone.0117780

87. Damanti CC, Ferrone L, Gaffo E, Garbin A, Tosato A, Contarini G, et al. Plasma small-extracellular vesicles enriched in miR-122-5p promote disease aggressiveness in pediatric anaplastic large-cell lymphoma. *Cancer Commun (Lond)* (2023) 43(5):630–4. doi: 10.1002/cac2.12415

88. Kasprzycka M, Marzec M, Liu X, Zhang Q, Wasik MA. Nucleophosmin/anaplastic lymphoma kinase (NPM/ALK) oncoprotein induces the T regulatory cell phenotype by activating STAT3. *Proc Natl Acad Sci* (2006) 103(26):9964–9. doi: 10.1073/pnas.0603507103

89. Wu R, Ivan E, Sahasrabuddhe AA, Shaw T, Mullighan CG, Leventaki V, et al. Epigenetic modulation of CD48 by NPM-ALK promotes immune evasion in ALK+ ALCL. *Blood* (2019) 134(Supplement_1):1510. doi: 10.1182/blood-2019-127453

90. Zhang Q, Wang H, Kantekure K, Paterson JC, Liu X, Schaffer A, et al. Oncogenic tyrosine kinase NPM-ALK induces expression of the growth-promoting receptor ICOS. *Blood* (2011) 118(11):3062–71. doi: 10.1182/blood-2011-01-332916

91. Zhang Q, Wang HY, Bhutani G, Liu X, Paessler M, Tobias JW, et al. Lack of TNFalpha expression protects anaplastic lymphoma kinase-positive T-cell lymphoma (ALK+ TCL) cells from apoptosis. *Proc Natl Acad Sci USA* (2009) 106(37):15843–8. doi: 10.1073/pnas.0907070106

92. Mussolin L, Damm-Welk C, Pillon M, Zimmermann M, Franceschetto G, Pulford K, et al. Use of minimal disseminated disease and immunity to NPM-ALK antigen to stratify ALK-positive ALCL patients with different prognosis. *Leukemia* (2013) 27(2):416–22. doi: 10.1038/leu.2012.205

93. Krenacs L, Wellmann A, Sorbara L, Himmelmann AW, Bagdi E, Jaffe ES, et al. Cytotoxic cell antigen expression in anaplastic large cell lymphomas of T- and null-cell type and hodgkin's disease: Evidence for distinct cellular origin. *Blood* (1997) 89(3):980–9. doi: 10.1182/blood.V89.3.980

94. Schleussner N, Merkel O, Costanza M, Liang HC, Hummel F, Romagnani C, et al. The AP-1-BATF and -BATF3 module is essential for growth, survival and TH17/ILC3 skewing of anaplastic large cell lymphoma. *Leukemia* (2018) 32(9):1994–2007. doi: 10.1038/s41375-018-0045-9

95. Congras A, Hoareau-Aveilla C, Caillet N, Tosolini M, Villarese P, Cieslak A, et al. ALK-transformed mature T lymphocytes restore early thymus progenitor features. *J Clin Invest* (2020) 130(12):6395–408. doi: 10.1172/JCI134990

96. Shinkai Y, Rathbun G, Lam KP, Oltz EM, Stewart V, Mendelsohn M, et al. RAG-2-deficient mice lack mature lymphocytes owing to inability to initiate V(D)J rearrangement. *Cell* (1992) 68(5):855–67. doi: 10.1016/0092-8674(92)90029-C

97. Malcolm TIM, Villarese P, Fairbairn CJ, Lamant L, Trinquand A, Hook CE, et al. Anaplastic large cell lymphoma arises in thymocytes and requires transient TCR expression for thymic egress. *Nat Commun* (2016) 7:10087. doi: 10.1038/ncomms10087

98. Bromberg J, Darnell JE. The role of STATs in transcriptional control and their impact on cellular function. *Oncogene* (2000) 19(21):2468–73. doi: 10.1038/sj.onc.1203476

99. Crockett DK, Lin Z, Elenitoba-Johnson KSJ, Lim MS. Identification of NPM-ALK interacting proteins by tandem mass spectrometry. *Oncogene* (2004) 23(15):2617–29. doi: 10.1038/sj.onc.1207398

100. Amin HM, Medeiros LJ, Ma Y, Feretzaki M, Das P, Leventaki V, et al. Inhibition of JAK3 induces apoptosis and decreases anaplastic lymphoma kinase activity in anaplastic large cell lymphoma. *Oncogene* (2003) 22(35):5399–407. doi: 10.1038/sj.onc.1206849

101. Khoury JD, Rassidakis GZ, Medeiros LJ, Amin HM, Lai R. Methylation of SHP1 gene and loss of SHP1 protein expression are frequent in systemic anaplastic large cell lymphoma. *Blood* (2004) 104(5):1580–1. doi: 10.1182/blood-2004-03-1151

102. Honorat JF, Ragab A, Lamant L, Delsol G, Ragab-Thomas J. SHP1 tyrosine phosphatase negatively regulates NPM-ALK tyrosine kinase signaling. *Blood* (2006) 107(10):4130–8. doi: 10.1182/blood-2005-06-2421

103. Klingmüller U, Lorenz U, Cantley LC, Neel BG, Lodish HF. Specific recruitment of SH-PTP1 to the erythropoietin receptor causes inactivation of JAK2 and termination of proliferative signals. *Cell* (1995) 80(5):729–38. doi: 10.1016/0092-8674(95)90351-8

104. Mathas S, Hinz M, Anagnostopoulos I, Krappmann D, Lietz A, Jundt F, et al. Aberrantly expressed c-Jun and JunB are a hallmark of Hodgkin lymphoma cells, stimulate proliferation and synergize with NF-kappa B. *EMBO J* (2002) 21(15):4104–13. doi: 10.1093/emboj/cdf389

105. Schiefer AI, Vesely P, Hassler MR, Egger G, Kenner L. The role of AP-1 and epigenetics in ALCL. *Front Biosci (Schol Ed)* (2015) 7:226–35. doi: 10.2741/s436

106. Wu Z, Nicoll M, Ingham RJ. AP-1 family transcription factors: a diverse family of proteins that regulate varied cellular activities in classical hodgkin lymphoma and ALK+ ALCL. *Exp Hematol Oncol* (2021) 10(1):4. doi: 10.1186/s40164-020-00197-9

107. Turner SD, Yeung D, Hadfield K, Cook SJ, Alexander DR. The NPM-ALK tyrosine kinase mimics TCR signalling pathways, inducing NFAT and AP-1 by RAS-dependent mechanisms. *Cell Signal* (2007) 19(4):740–7. doi: 10.1016/j.cellsig.2006.09.007

108. Quintanilla-Martinez L, Pittaluga S, Miething C, Klier M, Rudelius M, Davies-Hill T, et al. NPM-ALK-dependent expression of the transcription factor CCAAT/enhancer binding protein beta in ALK-positive anaplastic large cell lymphoma. *Blood* (2006) 108(6):2029–36. doi: 10.1182/blood-2005-10-014258

109. Anastasov N, Bonzheim I, Rudelius M, Klier M, Dau T, Angermeier D, et al. C/EBPβ expression in ALK-positive anaplastic large cell lymphomas is required for cell proliferation and is induced by the STAT3 signaling pathway. *Haematologica* (2010) 95 (5):760. doi: 10.3324/haematol.2009.014050

110. Bergalet J, Fawal M, Lopez C, Desjobert C, Lamant L, Delsol G, et al. HuR-mediated control of C/EBPbeta mRNA stability and translation in ALK-positive anaplastic large cell lymphomas. *Mol Cancer Res* (2011) 9(4):485–96. doi: 10.1158/1541-7786.MCR-10-0351

111. Bonzheim I, Irmler M, Klier-Richter M, Steinhilber J, Anastasov N, Schäfer S, et al. Identification of C/EBPβ target genes in ALK+ anaplastic large cell lymphoma (ALCL) by gene expression profiling and chromatin immunoprecipitation. *PloS One* (2013) 8(5):e64544. doi: 10.1371/journal.pone.0064544

112. Piva R, Pellegrino E, Mattioli M, Agnelli L, Lombardi L, Boccalatte F, et al. Functional validation of the anaplastic lymphoma kinase signature identifies CEBPB and BCl2A1 as critical target genes. *J Clin Invest* (2006) 116(12):3171–82. doi: 10.1172/JCI29401

113. O'Shea JJ, Gadina M, Siegel RM. Cytokines and cytokine receptors. Clinical immunology: principles and practice. *(Fifth Edition)* (2019) 127–155.e1. doi: 10.1016/B978-0-7020-6896-6.00009-0

114. Knörr F, Damm-Welk C, Ruf S, Singh VK, Zimmermann M, Reiter A, et al. Blood cytokine concentrations in pediatric patients with anaplastic lymphoma kinase-positive anaplastic large cell lymphoma. *Haematologica* (2018) 103(3):477–85. doi: 10.3324/haematol.2017.177972

115. Sueki Y, Nozaki Y, Kawashima I, Yamamoto T, Nakajima K, Mitumori T, et al. Anaplastic large cell lymphoma with paraneoplastic neutrophilia: an association between IL-17 elevation and aggressive disease progression. *Int J Hematol* (2014) 99 (6):773–6. doi: 10.1007/s12185-014-1557-3

116. Siebert S, Amos N, Williams BD, Lawson TM. Cytokine production by hepatic anaplastic large-cell lymphoma presenting as a rheumatic syndrome. *Semin Arthritis Rheum* (2007) 37(1):63–7. doi: 10.1016/j.semarthrit.2006.12.007

117. Zhang Q, Raghunath PN, Xue L, Majewski M, Carpentieri DF, Odum N, et al. Multilevel dysregulation of STAT3 activation in anaplastic lymphoma kinase-positive T/null-cell lymphoma. *J Immunol* (2002) 168(1):466–74. doi: 10.4049/jimmunol.168.1.466

118. Yoshimura A, Ichihara M, Kinjyo I, Moriyama M, Copeland NG, Gilbert DJ, et al. Mouse oncostatin M: an immediate early gene induced by multiple cytokines through the JAK-STAT5 pathway. *EMBO J* (1996) 15(5):1055–63. doi: 10.1002/j.1460-2075.1996.tb00443.x

119. Wu C, Molavi O, Zhang H, Gupta N, Alshareef A, Bone KM, et al. STAT1 is phosphorylated and downregulated by the oncogenic tyrosine kinase NPM-ALK in ALK-positive anaplastic large-cell lymphoma. *Blood* (2015) 126(3):336–45. doi: 10.1182/blood-2014-10-603738

120. Bluyssen HAR, Rastmanesh MM, Tilburgs C, Jie K, Wesseling S, Goumans MJ, et al. IFN gamma-dependent SOCS3 expression inhibits IL-6-induced STAT3 phosphorylation and differentially affects IL-6 mediated transcriptional responses in endothelial cells. *Am J Physiol Cell Physiol* (2010) 299(2):C354–62. doi: 10.1152/ajpcell.00513.2009

121. Delgoffe GM, Vignali DAA. STAT heterodimers in immunity: A mixed message or a unique signal? *JAKSTAT* (2013) 2(1):e23060. doi: 10.4161/jkst.23060

122. Gopal K, Kneteman NM, Zhang H, Lai R, Bone KM, Wu F, et al. STAT1 is phosphorylated and downregulated by the oncogenic tyrosine kinase NPM-ALK in ALK-positive anaplastic large-cell lymphoma. *Blood* (2015) 126(3):336–45. doi: 10.1182/blood-2014-10-603738

123. Haque SJ, Harbor P, Tabrizi M, Yi T, Williams BRG. Protein-tyrosine phosphatase Shp-1 is a negative regulator of IL-4- and IL-13-dependent signal transduction. *J Biol Chem* (1998) 273(51):33893–6. doi: 10.1074/jbc.273.51.33893

124. Han Y, Amin HM, Frantz C, Franko B, Lee J, Lin Q, et al. Restoration of shp1 expression by 5-AZA-2'-deoxycytidine is associated with downregulation of JAK3/STAT3 signaling in ALK-positive anaplastic large cell lymphoma. *Leukemia* (2006) 20 (9):1602–9. doi: 10.1038/sj.leu.2404323

125. Liu Y, Sattarzadeh A, Diepstra A, Visser L, Van Den Berg A. The microenvironment in classical Hodgkin lymphoma: An actively shaped and essential tumor component. *Semin Cancer Biol* (2014) 24:15–22. doi: 10.1016/j.semcancer.2013.07.002

126. Wu R, Sattarzadeh A, Rutgers B, Diepstra A, Van Den Berg A, Visser L. The microenvironment of classical Hodgkin lymphoma: heterogeneity by Epstein–Barr virus presence and location within the tumor. *Blood Cancer J* (2016) 6(5):e417–7. doi: 10.1038/bcj.2016.26

127. Popovic D, Vucic D, Dikic I. Ubiquitination in disease pathogenesis and treatment. *Nat Med* (2014) 20(11):1242–53. doi: 10.1038/nm.3739

128. Lee JS, Choi HJ, Baek SH. Sumoylation and its contribution to cancer. *Adv Exp Med Biol* (2017) 963:283–98. doi: 10.1007/978-3-319-50044-7_17

129. Vishwamitra D, Curry C v, Shi P, Alkan S, Amin HM. SUMOylation confers posttranslational stability on NPM-ALK oncogenic protein. *Neoplasia* (2015) 17 (9):742–54. doi: 10.1016/j.neo.2015.09.005

130. Zhang C, Han XR, Yang X, Jiang B, Liu J, Xiong Y, et al. Proteolysis targeting chimeras (PROTACs) of anaplastic lymphoma kinase (ALK). *Eur J Med Chem* (2018) 151:304–14. doi: 10.1016/j.ejmech.2018.03.071

131. Wang Y, Han L, Liu F, Yang F, Jiang X, Sun H, et al. Targeted degradation of anaplastic lymphoma kinase by gold nanoparticle-based multi-headed proteolysis targeting chimeras. *Colloids Surf B Biointerfaces* (2020) 188:110795. doi: 10.1016/j.colsurfb.2020.110795

132. Yan G, Zhong X, Yue L, Pu C, Shan H, Lan S, et al. Discovery of a PROTAC targeting ALK with *in vivo* activity. *Eur J Med Chem* (2021) 212:113150. doi: 10.1016/j.ejmech.2020.113150

133. Kang CH, Lee DH, Lee CO, du Ha J, Park CH, Hwang JY. Induced protein degradation of anaplastic lymphoma kinase (ALK) by proteolysis targeting chimera (PROTAC). *Biochem Biophys Res Commun* (2018) 505(2):542–7. doi: 10.1016/j.bbrc.2018.09.169

134. Pileri SA, Agostinelli C, Bacci F, Sabattini E, Sagramoso C, Falini B, et al. Pathobiology of ALK-negative anaplastic large cell lymphoma. *Pediatr Rep* (2011) 3 Suppl 2(Suppl 2):8–10. doi: 10.4081/pr.2011.s2.e5

135. Savage KJ, Harris NL, Vose JM, Ullrich F, Jaffe ES, Connors JM, et al. ALK-anaplastic large-cell lymphoma is clinically and immunophenotypically different from both ALK+ ALCL and peripheral T-cell lymphoma, not otherwise specified: report from the International Peripheral T-Cell Lymphoma Project. *Blood* (2008) 111 (12):5496–504. doi: 10.1182/blood-2008-01-134270

136. Hapgood G, Savage KJ. The biology and management of systemic anaplastic large cell lymphoma. *Blood* (2015) 126(1):17–25. doi: 10.1182/blood-2014-10-567461

137. Luchtel RA, Dasari S, Oishi N, Pedersen MB, Hu G, Rech KL, et al. Molecular profiling reveals immunogenic cues in anaplastic large cell lymphomas with DUSP22 rearrangements. *Blood* (2018) 132(13):1386–98. doi: 10.1182/blood-2018-03-838524

138. Luchtel RA, Zimmermann MT, Hu G, Dasari S, Jiang M, Oishi N, et al. Recurrent MSC E116K mutations in ALK-negative anaplastic large cell lymphoma. *Blood* (2019) 133(26):2776–89. doi: 10.1182/blood.2019000626

139. Sekine Y, Tsuji S, Ikeda O, Sato N, Aoki N, Aoyama K, et al. Regulation of STAT3-mediated signaling by LMW-DSP2. *Oncogene* (2006) 25(42):5801–6. doi: 10.1038/sj.onc.1209578

140. Parkhi M, Bal A, Das A, Kashyap D, Bhardwaj S, Prakash G, et al. ALK-negative anaplastic large cell lymphoma (ALCL): prognostic implications of molecular subtyping and JAK-STAT pathway. *Appl Immunohistochem Mol Morphol* (2021) 29 (9):648–56. doi: 10.1097/PAI.0000000000000936

141. Candi E, Dinsdale D, Rufini A, Salomoni P, Knight RA, Mueller M, et al. TAp63 and DeltaNp63 in cancer and epidermal development. *Cell Cycle* (2007) 6(3):274–84. doi: 10.4161/cc.6.3.3797

142. Melino G. p63 is a suppressor of tumorigenesis and metastasis interacting with mutant p53. *Cell Death Differ* (2011) 18(9):1487–99. doi: 10.1038/cdd.2011.81

143. Parrilla Castellar ER, Jaffe ES, Said JW, Swerdlow SH, Ketterling RP, Knudson RA, et al. ALK-negative anaplastic large cell lymphoma is a genetically heterogeneous disease with widely disparate clinical outcomes. *Blood* (2014) 124(9):1473–80. doi: 10.1182/blood-2014-04-571091

144. Crescenzo R, Abate F, Lasorsa E, Tabbo' F, Gaudiano M, Chiesa N, et al. Convergent mutations and kinase fusions lead to oncogenic STAT3 activation in anaplastic large cell lymphoma. *Cancer Cell* (2015) 27(4):516–32. doi: 10.1016/j.ccell.2015.03.006

145. Scarfò I, Pellegrino E, Mereu E, Kwee I, Agnelli L, Bergaggio E, et al. Identification of a new subclass of ALK-negative ALCL expressing aberrant levels of ERBB4 transcripts. *Blood* (2016) 127(2):221–32. doi: 10.1182/blood-2014-12-614503

146. Turner SD, Inghirami G, Miranda RN, Kadin ME. Cell of origin and immunologic events in the pathogenesis of breast implant–associated anaplastic large-cell lymphoma. *Am J Pathol* (2020) 190(1):2–10. doi: 10.1016/j.ajpath.2019.09.005

147. Cappellano G, Ploner C, Lobenwein S, Sopper S, Hoertnagl P, Mayerl C, et al. Immunophenotypic characterization of human T cells after *in vitro* exposure to different silicone breast implant surfaces. *PloS One* (2018) 13(2):e0192108. doi: 10.1371/journal.pone.0192108

148. Kadin ME, Morgan J, Xu H, Epstein AL, Sieber D, Hubbard BA, et al. IL-13 is produced by tumor cells in breast implant-associated anaplastic large cell lymphoma: implications for pathogenesis. *Hum Pathol* (2018) 78:54–62. doi: 10.1016/j.humpath.2018.04.007

149. Quesada AE, Medeiros LJ, Clemens MW, Ferrufino-Schmidt MC, Pina-Oviedo S, Miranda RN. Breast implant-associated anaplastic large cell lymphoma: a review. *Mod Pathol* (2019) 32(2):166–88. doi: 10.1038/s41379-018-0134-3

150. Laurent C, Nicolae A, Laurent C, le Bras F, Haioun C, Fataccioli V, et al. Gene alterations in epigenetic modifiers and JAK-STAT signaling are frequent in breast implant–associated ALCL. *Blood* (2020) 135(5):360–70. doi: 10.1182/blood.2019001904

151. Blombery P, Thompson E, Ryland GL, Joyce R, Byrne DJ, Khoo C, et al. Frequent activating STAT3 mutations and novel recurrent genomic abnormalities detected in breast implant-associated anaplastic large cell lymphoma. *Oncotarget* (2018) 9(90):36126–36. doi: 10.18632/oncotarget.26308

152. Oishi N, Brody GS, Ketterling RP, Viswanatha DS, He R, Dasari S, et al. Genetic subtyping of breast implant-associated anaplastic large cell lymphoma. *Blood* (2018) 132(5):544–7. doi: 10.1182/blood-2017-12-821868

153. di Napoli A, Jain P, Duranti E, Margolskee E, Arancio W, Facchetti F, et al. Targeted next generation sequencing of breast implant-associated anaplastic large cell lymphoma reveals mutations in JAK/STAT signalling pathway genes, TP53 and DNMT3A. *Br J Haematol* (2018) 180(5):741–4. doi: 10.1111/bjh.14431

154. Quesada AE, Zhang Y, Ptashkin R, Ho C, Horwitz S, Benayed R, et al. Next generation sequencing of breast implant-associated anaplastic large cell lymphomas reveals a novel STAT3-JAK2 fusion among other activating genetic alterations within the JAK-STAT pathway. *Breast J* (2021) 27(4):314–21. doi: 10.1111/tbj.14205

155. Oishi N, Miranda RN, Feldman AL. Genetics of breast implant-associated anaplastic large cell lymphoma (BIA-ALCL). *Aesthet Surg J* (2019) 39(Suppl_1):S14–20. doi: 10.1093/asj/sjy311

156. Melchers RC, Willemze R, Van De Loo M, Van Doorn R, Jansen PM, Cleven AHG, et al. Clinical, histologic, and molecular characteristics of anaplastic lymphoma kinase-positive primary cutaneous anaplastic large cell lymphoma. *Am J Surg Pathol* (2020) 44(6):776–781. doi: 10.1097/PAS.0000000000001449

157. Quintanilla-Martinez L, Jansen PM, Kinney MC, Swerdlow SH, Willemze R. Non-mycosis fungoides cutaneous T-cell lymphomas: report of the 2011 Society for Hematopathology/European Association for Haematopathology workshop. *Am J Clin Pathol* (2013) 139(4):491–514. doi: 10.1309/AJCP83AOQTMLOJTM

158. Attygalle AD, Cabeçadas J, Gaulard P, Jaffe ES, de Jong D, Ko YH, et al. Peripheral T-cell and NK-cell lymphomas and their mimics; taking a step forward - report on the lymphoma workshop of the XVIth meeting of the European Association for Haematopathology and the Society for Hematopathology. *Histopathology* (2014) 64(2):171–99. doi: 10.1111/his.12251

159. Gallardo F, Barranco C, Toll A, Pujol RM. CD30 antigen expression in cutaneous inflammatory infiltrates of scabies: a dynamic immunophenotypic pattern that should be distinguished from lymphomatoid papulosis. *J Cutan Pathol* (2002) 29(6):368–73. doi: 10.1034/j.1600-0560.2002.290608.x

160. Oschlies I, Lisfeld J, Lamant L, Nakazawa A, d'Amore ESG, Hansson U, et al. ALK-positive anaplastic large cell lymphoma limited to the skin: clinical, histopathological and molecular analysis of 6 pediatric cases. A report from the ALCL99 study. *Haematologica* (2013) 98(1):50–6. doi: 10.3324/haematol.2012.065664

161. Kadin ME, Pinkus JL, Pinkus GS, Duran IH, Fuller CE, Onciu M, et al. Primary cutaneous ALCL with phosphorylated/activated cytoplasmic ALK and novel phenotype: EMA/MUC1+, cutaneous lymphocyte antigen negative. *Am J Surg Pathol* (2008) 32(9):1421–6. doi: 10.1097/PAS.0b013e3181648d6d

162. Mao X, Orchard G, Lillington DM, Russell-Jones R, Young BD, Whittaker S. Genetic alterations in primary cutaneous CD30+ anaplastic large cell lymphoma. *Genes Chromosomes Cancer* (2003) 37(2):176–85. doi: 10.1002/gcc.10184

163. Szuhai K, van Doorn R, Tensen CP, Kester V. Array-CGH analysis of cutaneous anaplastic large cell lymphoma. *Methods Mol Biol* (2013) 973:197–212. doi: 10.1007/978-1-62703-281-0_13

164. Velusamy T, Kiel MJ, Sahasrabuddhe AA, Rolland D, Dixon CA, Bailey NG, et al. A novel recurrent NPM1-TYK2 gene fusion in cutaneous CD30-positive lymphoproliferative disorders. *Blood* (2014) 124(25):3768–71. doi: 10.1182/blood-2014-07-588434

165. Liang HC, Kapoor R, Onder O, Dennis K, Lim MS, Elenitoba-Johnson KSJ. The NPM1-TYK2 chimeric fusion promotes activation of STAT family signaling, skewing towards tfh functional subset differentiation and mature T-cell lymphomagenesis. *Blood* (2022) 140(Supplement 1):9281–2. doi: 10.1182/blood-2022-169494

frontiers | Frontiers in Oncology

TYPE Mini Review
PUBLISHED 29 September 2023
DOI 10.3389/fonc.2023.1199108

OPEN ACCESS

EDITED BY
Juliana Pereira,
University of São Paulo, Brazil

REVIEWED BY
Jean Feuillard,
University of Limoges, France
Javeed Iqbal,
University of Nebraska Medical Center, United States

*CORRESPONDENCE
Shannon A. Carty
scarty@umich.edu

RECEIVED 02 April 2023
ACCEPTED 18 September 2023
PUBLISHED 29 September 2023

CITATION
Carty SA (2023) Biological insights into the role of TET2 in T cell lymphomas. *Front. Oncol.* 13:1199108. doi: 10.3389/fonc.2023.1199108

Biological insights into the role of TET2 in T cell lymphomas

Shannon A. Carty*

Division of Hematology-Oncology, Department of Internal Medicine, University of Michigan, Ann Arbor, MI, United States

Peripheral T cell lymphomas (PTCL) are a heterogenous group of mature T cell lymphomas with an overall poor prognosis. Understanding the molecular heterogeneity in PTCL subtypes may lead to improved understanding of the underlying biological mechanisms driving these diseases. Mutations in the epigenetic regulator TET2 are among the most frequent mutations identified in PTCL, with the highest frequency in angioimmunoblastic T cell lymphomas and other nodal T follicular helper (TFH) lymphomas. This review dissects the role of TET2 in nodal TFH cell lymphomas with a focus on emerging biological insights into the molecular mechanism promoting lymphomagenesis and the potential for epigenetic therapies to improve clinical outcomes.

KEYWORDS

TET2, angioimmunoblastic T cell lymphoma, T follicular helper cell lymphoma, epigenetic therapy, peripheral T cell lymphoma (PTCL)

Introduction

Peripheral T cell lymphomas (PTCL) are a heterogenous group of aggressive lymphomas derived from mature T cells and account for 10-15% of all non-Hodgkin lymphomas (1). Among PTCL cases, the World Health Organization (WHO) classification has recently recognized a distinct entity termed nodal T follicular helper cell (TFH) lymphomas, which include the subtypes previously termed angioimmunoblastic T cell lymphoma (AITL), follicular T cell lymphoma and peripheral T cell lymphoma with a TFH phenotype (2). Nodal TFH lymphomas share phenotypic and gene expression similarities with normal T follicular helper (TFH) cells (3–5), a $CD4^+$ T cell subset that promotes germinal center B cell differentiation (6). In the International Peripheral T-cell and Natural Killer/T-cell Lymphoma study, AITL (at the time the most recognized nodal TFH lymphoma) accounted for approximately 20% of PTCL cases and thus is the second most common PTCL subtype after PTCL, not otherwise specified (1). Clinically, nodal TFH lymphomas typically present at an advanced stage with lymphadenopathy, hepatosplenomegaly and constitutional symptoms, as well as various autoimmune manifestations (7–9). PTCLs, including nodal TFH lymphomas, have an overall poor prognosis with AITL patients having an expected 5-year overall survival of ~30% (1, 10). Recurrent somatic mutations in multiple epigenetic regulators, including loss-of-function mutations in TET2, inactivating mutations in DNMT3A and neomorphic mutations in IDH2, have been strongly associated with nodal TFH lymphomas (11–16). Given the poor

prognosis of these lymphomas, understanding the underlying biology is critical to design therapies with improved efficacy. Given that TET2 is the most commonly mutated epigenetic regulator in these lymphomas, this review will focus on the mechanistic role of TET2 in the development and treatment of nodal TFH lymphomas.

TET2 function

TET2 is a member of the ten-eleven-translocation (TET) family of Fe^{2+}- and alpha-ketoglutarate-dependent methylcytosine dioxygenases. These enzymes oxidize 5-methylcytosine (5mC) to 5-hydroxymethylcytosine (5hmC) and subsequent oxidized methylcytosine intermediates to ultimately generate an unmodified cytosine (17–19). DNA methylation was long thought to be a relatively stable epigenetic mark; however, the discovery of the TET family of enzymes introduced the concept of active DNA demethylation. TET2 is broadly expressed in hematopoietic cells and TET2 loss promotes hematopoietic stem cell (HSC) and myeloid cell expansion in murine models (19–22). Studies of TET2 function in murine and human hematopoietic cells reveal that TET2 deletion or loss-of-function mutations, such as those in nodal TFH lymphomas, lead to altered DNA methylation and chromatin accessibility at regulatory enhancer regions (23–25), suggesting functional epigenetic consequences.

In the study of TET2 function in T cells, deletion of TET2 in mature T cells does not result in any appreciable alteration in late T cell development or peripheral T cell activation (26). However, in antigen-specific $CD4^+$ T cells, TET2 loss leads to an increase in TFH differentiation in a cell-intrinsic manner with hypermethylation at gene loci associated with helper T cell differentiation (27), suggesting that TET2 directly represses TFH differentiation by demethylating key regulatory loci.

TET2 and associated mutations in T cell lymphomas

Loss-of-function mutations in TET2 were first identified in myeloid malignancies (28, 29) but soon thereafter recurrent somatic mutations in TET2 were recognized in approximately 50-70% of AITL and other TFH-derived lymphomas (11–15). Subsequent gene sequencing of other T cell leukemias/lymphomas revealed TET2 mutations at much lower frequencies compared to nodal TFH lymphomas – including 17% of T-cell prolymphocytic leukemia cases (30), 14-20% of acute T-cell leukemia/lymphoma cases (31, 32) and in 4-12% cases of cutaneous T cell lymphoma and/or Sezary syndrome cases (33–35).

Furthermore, frequent mutations at isocitrate dehydrogenase 2 arginine 172 (IDH2 R172) have also been identified in AITL and other TFH-derived lymphomas (16). IDH2 is a mitochondrial enzyme that typically converts isocitrate to 2-alpha-ketoglutarate (aKG); however, the R172 mutation promotes abnormal oncometabolite production of the R-enantiomer of alpha-hydroxyglutarate (aHG) (36), which competitively inhibits aKG-dependent enzymes including the TET family (37). Thus, nodal TFH-derived lymphomas with IDH2 R172 mutations are predicted to have repressed TET activity and accordingly IDH2-mutated AITL exhibits genome-wide DNA hypermethylation compared to IDH2 wild-type AITL (38).

TET2 role in lymphomagenesis

Despite frequent TET2 mutations in a wide array of T cell lymphomas, most commonly in nodal TFH lymphomas, it was initially unclear the degree to which TET2 loss-of-function directly contributed to lymphomagenesis. TET2 deletion in murine hematopoietic stem cells (HSCs) altered early and late hematopoiesis in both myeloid and lymphoid lineages with eventual development of myeloid malignancies in the mice (14, 21, 22) but only rarely mature lymphoid malignancies (14). A murine model with a hypomorphic TET2 allele does develop TFH-like lymphomas but with a prolonged latency (39).

In several sequencing studies of nodal TFH lymphomas including AITL, multiple TET2 mutations were found in individual tumor samples implying a strong selective pressure (12, 13, 40). Additionally, in AITL cases, the majority of the cases that carried a TET2 mutation had a variant allele frequency >10% (12, 38, 40).

Since TET2 mutations were frequently found to co-occur with a glycine to valine (G17V) inactivating mutation in Rho GTPase RhoA in 50-70% of AITL cases (12, 40, 41), several groups sought to dissect the relative contribution of RhoA-G17V mutations and TET2 loss of function to T cell lymphomagenesis. Adoptive transfer of wild-type or TET2-deficient T cells retrovirally transduced to overexpress RhoA-G17V into T-cell deficient murine hosts resulted in $CD4^+$ T cell expansion, disruption of peripheral T cell homeostasis and eventually lethal inflammation but no lymphoma was noted (42). Several other groups generated transgenic mice expressing the RhoA-G17V mutation in the setting of TET2 hematopoietic deficiency. In these various murine models, RhoA-G17V overexpression in T cells promoted TFH proliferation/expansion (43, 44) and the concomitant expression in the setting of hematopoietic TET2 deficiency led to the development of TFH lymphomas with varying penetrance (43–45). On a molecular level, RhoA-G17V and TET2 loss was found to promote mammalian target of rapamycin complex 1 (mTORC1) pathway activation (43, 44) and inactivation of forkhead box O1 (FOXO1) signaling (44), suggesting potential therapeutic targets. Together these data strongly support a role for RhoA-G17V as a driving mutation in nodal TFH lymphomas but also speak to the requirement for concomitant TET2 loss in the hematopoietic compartment to promote lymphomagenesis. Targeting of downstream pathways, such as with mTOR inhibitors, may be an attractive therapeutic target to be tested in nodal TFH lymphomas, though no trials are currently underway.

TET2 in clonal hematopoiesis and tumor microenvironment

Mutations in TET2 are among the three most frequent somatic mutations in age-related clonal hematopoiesis and are associated with an increased risk of hematologic cancers as well as all-cause mortality (46–48). The extent to which TET2 mutations in the lymphoma microenvironment and responding immune cells contributes to T cell lymphomagenesis has not been fully elucidated.

It has been posited that TET2 mutations noted in nodal TFH lymphomas largely arise in the setting of clonal hematopoiesis, which is supported by the fact that TET2 mutations in T cell lymphoma patients are frequently found to co-occur in the non-neoplastic B lymphocyte, myeloid and HSC compartments as well as the neoplastic T cells (14, 49, 50). In patients with AITL, the majority of patients had TET2 mutations identified in the neoplastic T cells as well as the myeloid compartment (51). In this case series, 4 of 22 patients with TET2 mutations and available sequencing data developed myeloid neoplasms approximately 2-4 years following their lymphoma diagnosis. The myeloid neoplasms all shared multiple TET2 mutations in the myeloid clone and AITL cells but also contained additional different mutations that were not shared. Together these data support myeloid neoplasms arising from early clonal TET2-mutated hematopoietic stem cells but with divergent evolution from the neoplastic AITL cells.

The presence of TET2-mutated immune cells in AITL patients led to the question if TET2 mutations alter tumor immunity to promote T cell lymphomagenesis. TET2 is known to have pleiotropic functions in different immune cells known to play a role in tumor immunity, including macrophages/monocytes, $CD4^+$ helper T cells, T regulatory cells, $CD8^+$ T cells and B cells (52). In myeloid cells, TET2 represses inflammatory gene expression (53) with increased IL-6, IL-1β and arginase 1 in TET2-deficient macrophages (54, 55). In a murine melanoma model, TET2 deletion in myeloid cells resulted in reduced tumor burden and increased tumor-infiltrating T cells suggesting that TET2 promotes a myeloid immunosuppressive program in the tumor microenvironment (56). In $CD4^+$ T cells, TET2 inhibits cytokine production, including IFNγ, IL-17 and IL-10 (57), cytokines which can have both immunostimulatory and immunosuppressive roles. Furthermore, TET2 (in combination with either TET1 or TET3) dampens regulatory T cell immunosuppressive function (58, 59), which are a critical cellular subset known to suppress anti-tumor responses (60). In $CD8^+$ T cells, TET2 represses memory differentiation following infection (26), though less is known about the role of TET2 deficiency in $CD8^+$ T cell anti-tumor immunity and T cell exhaustion. Given these pleotropic roles TET2 may play in the tumor microenvironment, it is important to carefully analyze the tumor-intrinsic versus microenvironmental roles TET2 loss-of-function mutations play in promoting nodal TFH lymphomas.

A recent elegant study dissected T cell-intrinsic versus -extrinsic role of TET2 in lymphomagenesis using murine models with either hematopoietic or T cell specific loss of TET2 crossed to RhoA-G17V transgenic mice (61). TET2 deficiency in all hematopoietic cells accelerated the development of TFH lymphomas compared to either a wild-type hematopoietic compartment or TET2 deletion solely in T cells. To test which immune compartment contributed to TFH lymphomagenesis, the authors co-transplanted tumor cells with a variety of immune lineages into immunodeficient mice and monitored tumor development. Only when B cells were co-transplanted did donor-derived tumors develop suggesting that TET2 loss in B cells supported TFH lymphomagenesis. Subsequent analysis revealed clonal expansion of TET2-deficient germinal center B cells in the tumor-bearing mice, unique mutations in core histones developed in murine clonal B cells and that inhibition of CD40-CD40L interactions prolonged survival in mice. Correlative studies in human AITL samples demonstrated an expansion of germinal center B cells in involved lymph nodes and unique mutations (some also in core histone genes) in the tumor-associated B cells and plasma cells. These data strongly support a cooperating role for TET2-mutated B cells in the immune microenvironment to promote nodal TFH lymphoma development. Targeting these interactions could provide a novel therapeutic avenue in nodal TFH lymphoma patients, although it remains unclear if this mechanism occurs outside of TET2-mutated clonal hematopoiesis.

Treatment and prognosis implications

Since AITL and other nodal TFH lymphomas have an overall poor prognosis with currently available treatments (1, 10), novel therapeutic approaches are needed to improve patient outcomes. TET2 mutations have been noted to be associated with adverse clinical parameters (13, 62) but not associated with a change in overall survival (13). Given the frequency of TET2 and other epigenetic mutations (ie, DNMT3A) that occur in the majority of nodal TFH lymphomas, there is great interest in utilizing epigenetic therapies to target underlying biological mechanism in hopes to improve response rates and survival. PTCL has been shown to be uniquely responsive to one type of epigenetic therapy, specifically histone deacetylase (HDAC) inhibitors, with three HDAC inhibitors approved for systemic PTCL: romidepson, belinostat and chidamide (in China). In the phase II trial of romidepsin in relapsed/refractory PTCL, patients with relapsed/refractory AITL had an overall response rate of 33% compared to 25% of the overall cohort with two-thirds of the AITL responders achieving a complete remission (63). Similarly, in the phase II registration study of belinostat in relapsed/refractory PTCL, patients with AITL seemed to have improved response rates (45%) compared to response rate (26%) of the overall trial population (64). A more recent retrospective, multicenter study comparing HDAC inhibitor responses in TFH versus non-TFH PTCL patients found a significantly improved overall response rate in nodal TFH *vs.* non-TFH lymphomas (56.5% versus 29.4%) (65). Together these data support the idea that nodal TFH lymphomas may be more sensitive to epigenetic modulation than non-TFH lymphomas (summarized in Table 1), whether this sensitivity correlates with

TABLE 1 Response rates in epigenetic therapies in relapsed/refractory nodal TFH lymphomas versus overall PTCL.

Study	Type	Disease Status	Overall		TFH/AITL	
			Number	ORR	Number	ORR
Romidepsin (54)	Phase II	R/R	130	25%	27	33%
Belinostat (55)	Phase II	R/R	129	26%	22	45.5%
HDAC inhibitor (56)	Retrospective	R/R	127	45.6%	76	56.5%
Aza/Romidepsin (63)	Phase II	Tx Naïve & R/R	23	61%	15	80%

ORR, overall response rate; R/R, relapsed/refractory; Tx, treatment.

the presence of epigenetic alterations due to TET2 mutations remains unknown.

Given TET2's function in active DNA demethylation, questions naturally arise about the role of hypomethylating agents (HMAs) in nodal TFH lymphomas. Several case reports and case series suggest some clinical efficacy of single agent HMAs (5-azacitidine or decitabine) in TET2-mutated angioimmunoblastic T cell lymphoma (66–69). Preclinical studies have suggested synergy between HMAs and HDAC inhibitors in T cell lymphomas (70, 71) providing a biologic rationale for combined epigenetic targeted therapy in PTCL patients. A multicenter phase II trial examining the combination of oral 5-azacitidine and romidepsin in treatment naïve and relapsed/refractory PTCL patients found that patients with a TFH phenotype had higher overall response rate (80%) and complete response (60%) compared to the overall response rate (25%) and complete remission rate (12.5%) among patients with other subtypes (72). In this early-phase study, there were no statistical differences in response rates between patients with wild-type or mutated TET2 but this was limited by small sample size. Together these data suggest that duel epigenetic targeting therapies may be particularly effective in nodal TFH lymphomas.

Since patients with relapsed/refractory AITL have progressively shorter remissions with each subsequent line of therapy (73), the best chance to cure patients likely lies in improving first-line therapies. Based on the emerging understanding of the underlying biology and the role of epigenetic targeted therapies in nodal TFH lymphomas, several studies have been undertaken to combine epigenetic therapy with standard front-line chemotherapy (CHOP, cyclophosphamide, doxorubicin, vincristine and prednisone). A randomized phase III trial compared to romidepsin plus CHOP to CHOP alone in treatment naïve patients with PTCL. Unfortunately, there were no differences in response rates, progression free survival or overall survival and there were more treatment-related adverse advents in the Ro-CHOP arm (74). However, in an exploratory analysis, PTCL patients with a TFH phenotype had improved progression free survival after Ro-CHOP compared CHOP suggesting that nodal TFH lymphomas may derive a unique benefit and clinical trials should be further focused on PTCL subsets. Clinical trials focused on nodal TFH PTCL populations are examining the efficacy of combining azacitidine (NCT03542266) or chidamide (NCT03853044) with frontline CHOP. Recently published results of the phase II trial of oral azacitidine plus CHOP in 20 evaluable PTCL patients demonstrated a complete response in 88.2% of PTCL-TFH patients and 2-year progression free survival of 69.2% in PTCL-TFH patients. Notably, TET2 mutations were significantly associated with complete response rates and overall survival (75). The oral azacitidine plus CHOP combination is being tested in an ongoing randomized phase II trial in previously untreated patients with CD30-negative PTCL (NCT04803201).

Conclusions

From the initial identification of TET2 mutations in AITL and other nodal TFH lymphomas just over twenty years ago, significant strides have been made to advance the understanding of TET2's role in the pathogenesis of these lymphomas. Namely, TET2 loss of function in the lymphoma microenvironment, which arises in the setting of clonal hematopoiesis, likely play a critical role in supporting TFH transformation and AITL development. Additionally, emerging clinical evidence suggests that epigenetic targeted therapies may improve response rates and survival in patients with nodal TFH lymphomas. Using the evolving scientific knowledge about the underlying biology of these rare lymphomas, future clinical trials may need to tailor trial populations to discern true efficacy of these therapies.

Author contributions

The author reviewed the literature, performed the writing and revisions of the manuscript.

Funding

This work was supported by a Limpert Clinical Scholar Career Development award from the University of Michigan.

Conflict of interest

The authors declares that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Vose J, Armitage J, Weisenburger D, International TCLP. International peripheral T-cell and natural killer/T-cell lymphoma study: pathology findings and clinical outcomes. *J Clin Oncol* (2008) 26(25):4124–30. doi: 10.1200/JCO.2008.16.4558

2. Alaggio R, Amador C, Anagnostopoulos I, Attygalle AD, Araujo IBO, Berti E, et al. The 5th edition of the world health organization classification of haematolymphoid tumours: lymphoid neoplasms. *Leukemia* (2022) 36(7):1720–48. doi: 10.1038/s41375-022-01620-2

3. de Leval L, Rickman DS, Thielen C, Reynies A, Huang YL, Delsol G, et al. The gene expression profile of nodal peripheral T-cell lymphoma demonstrates a molecular link between angioimmunoblastic T-cell lymphoma (Aitl) and follicular helper T (Tfh) cells. *Blood* (2007) 109(11):4952–63. doi: 10.1182/blood-2006-10-055145

4. Grogg KL, Attygalle AD, Macon WR, Remstein ED, Kurtin PJ, Dogan A. Expression of cxcl13, a chemokine highly upregulated in germinal center T-helper cells, distinguishes angioimmunoblastic T-cell lymphoma from peripheral T-cell lymphoma, unspecified. *Mod Pathol* (2006) 19(8):1101–7. doi: 10.1038/modpathol.3800625

5. Dobay MP, Lemonnier F, Missiaglia E, Bastard C, Vallois D, Jais JP, et al. Integrative clinicopathological and molecular analyses of angioimmunoblastic T-cell lymphoma and other nodal lymphomas of follicular helper T-cell origin. *Haematologica* (2017) 102(4):e148–e51. doi: 10.3324/haematol.2016.158428

6. Crotty S. T follicular helper cell biology: A decade of discovery and diseases. *Immunity* (2019) 50(5):1132–48. doi: 10.1016/j.immuni.2019.04.011

7. Ma H, O'Connor OA, Marchi E. Management of angioimmunoblastic T-cell lymphoma (Aitl) and other T follicular helper cell lymphomas (Tfh ptcl). *Semin Hematol* (2021) 58(2):95–102. doi: 10.1053/j.seminhematol.2021.03.001

8. Federico M, Rudiger T, Bellei M, Nathwani BN, Luminari S, Coiffier B, et al. Clinicopathologic characteristics of angioimmunoblastic T-cell lymphoma: analysis of the international peripheral T-cell lymphoma project. *J Clin Oncol* (2013) 31(2):240–6. doi: 10.1200/JCO.2011.37.3647

9. Mourad N, Mounier N, Briere J, Raffoux E, Delmer A, Feller A, et al. Clinical, biologic, and pathologic features in 157 patients with angioimmunoblastic T-cell lymphoma treated within the groupe D'etude des lymphomes de L'adulte (Gela) trials. *Blood* (2008) 111(9):4463–70. doi: 10.1182/blood-2007-08-105759

10. Advani RH, Skrypets T, Civallero M, Spinner MA, Manni M, Kim WS, et al. Outcomes and prognostic factors in angioimmunoblastic T-cell lymphoma: final report from the international T-cell project. *Blood* (2021) 138(3):213–20. doi: 10.1182/blood.2020010387

11. Lemonnier F, Couronne L, Parrens M, Jais JP, Travert M, Lamant L, et al. Recurrent tet2 mutations in peripheral T-cell lymphomas correlate with tfh-like features and adverse clinical parameters. *Blood* (2012) 120(7):1466–9. doi: 10.1182/blood-2012-02-408542

12. Palomero T, Couronne L, Khiabanian H, Kim MY, Ambesi-Impiombato A, Perez-Garcia A, et al. Recurrent mutations in epigenetic regulators, rhoa and fyn kinase in peripheral T cell lymphomas. *Nat Genet* (2014) 46(2):166–70. doi: 10.1038/ng.2873

13. Odejide O, Weigert O, Lane AA, Toscano D, Lunning MA, Kopp N, et al. A targeted mutational landscape of angioimmunoblastic T-cell lymphoma. *Blood* (2014) 123(9):1293–6. doi: 10.1182/blood-2013-10-531509

14. Quivoron C, Couronne L, Della Valle V, Lopez CK, Plo I, Wagner-Ballon O, et al. Tet2 inactivation results in pleiotropic hematopoietic abnormalities in mouse and is a recurrent event during human lymphomagenesis. *Cancer Cell* (2011) 20(1):25–38. doi: 10.1016/j.ccr.2011.06.003

15. Couronne L, Bastard C, Bernard OA. Tet2 and dnmt3a mutations in human T-cell lymphoma. *N Engl J Med* (2012) 366(1):95–6. doi: 10.1056/NEJMc1111708

16. Cairns RA, Iqbal J, Lemonnier F, Kucuk C, de Leval L, Jais JP, et al. Idh2 mutations are frequent in angioimmunoblastic T-cell lymphoma. *Blood* (2012) 119(8):1901–3. doi: 10.1182/blood-2011-11-391748

17. Ito S, D'Alessio AC, Taranova OV, Hong K, Sowers LC, Zhang Y. Role of tet proteins in 5mc to 5hmc conversion, es-cell self-renewal and inner cell mass specification. *Nature* (2010) 466(7310):1129–33. doi: 10.1038/nature09303

18. Tahiliani M, Koh KP, Shen Y, Pastor WA, Bandukwala H, Brudno Y, et al. Conversion of 5-methylcytosine to 5-hydroxymethylcytosine in mammalian DNA by mll partner tet1. *Science* (2009) 324(5929):930–5. doi: 10.1126/science.1170116

19. Koh KP, Yabuuchi A, Rao S, Huang Y, Cunniff K, Nardone J, et al. Tet1 and tet2 regulate 5-hydroxymethylcytosine production and cell lineage specification in mouse embryonic stem cells. *Cell Stem Cell* (2011) 8(2):200–13. doi: 10.1016/j.stem.2011.01.008

20. Ko M, Bandukwala HS, An J, Lamperti ED, Thompson EC, Hastie R, et al. Ten-eleven-translocation 2 (Tet2) negatively regulates homeostasis and differentiation of hematopoietic stem cells in mice. *Proc Natl Acad Sci U.S.A.* (2011) 108(35):14566–71. doi: 10.1073/pnas.1112317108

21. Moran-Crusio K, Reavie L, Shih A, Abdel-Wahab O, Ndiaye-Lobry D, Lobry C, et al. Tet2 loss leads to increased hematopoietic stem cell self-renewal and myeloid transformation. *Cancer Cell* (2011) 20(1):11–24. doi: 10.1016/j.ccr.2011.06.001

22. Li Z, Cai X, Cai CL, Wang J, Zhang W, Petersen BE, et al. Deletion of tet2 in mice leads to dysregulated hematopoietic stem cells and subsequent development of myeloid Malignancies. *Blood* (2011) 118(17):4509–18. doi: 10.1182/blood-2010-12-325241

23. Rasmussen KD, Jia G, Johansen JV, Pedersen MT, Rapin N, Bagger FO, et al. Loss of tet2 in hematopoietic cells leads to DNA hypermethylation of active enhancers and induction of leukemogenesis. *Genes Dev* (2015) 29(9):910–22. doi: 10.1101/gad.260174.115

24. Izzo F, Lee SC, Poran A, Chaligne R, Gaiti F, Gross B, et al. DNA methylation disruption reshapes the hematopoietic differentiation landscape. *Nat Genet* (2020) 52(4):378–87. doi: 10.1038/s41588-020-0595-4

25. Tulstrup M, Soerensen M, Hansen JW, Gillberg L, Needhamsen M, Kaastrup K, et al. Tet2 mutations are associated with hypermethylation at key regulatory enhancers in normal and Malignant hematopoiesis. *Nat Commun* (2021) 12(1):6061. doi: 10.1038/s41467-021-26093-2

26. Carty SA, Gohil M, Banks LB, Cotton RM, Johnson ME, Stelekati E, et al. The loss of tet2 promotes cd8(+) T cell memory differentiation. *J Immunol* (2018) 200(1):82–91. doi: 10.4049/jimmunol.1700559

27. Baessler A, Novis CL, Shen Z, Perovanovic J, Wadsworth M, Thiede KA, et al. Tet2 coordinates with foxo1 and runx1 to balance T follicular helper cell and T helper 1 cell differentiation. *Sci Adv* (2022) 8(24):eabm4982. doi: 10.1126/sciadv.abm4982

28. Delhommeau F, Dupont S, Della Valle V, James C, Trannoy S, Masse A, et al. Mutation in tet2 in myeloid cancers. *N Engl J Med* (2009) 360(22):2289–301. doi: 10.1056/NEJMoa0810069

29. Tefferi A, Lim KH, Abdel-Wahab O, Lasho TL, Patel J, Patnaik MM, et al. Detection of mutant tet2 in myeloid Malignancies other than myeloproliferative neoplasms: cmml, mds, mds/mpn and aml. *Leukemia* (2009) 23(7):1343–5. doi: 10.1038/leu.2009.59

30. Lopez C, Bergmann AK, Paul U, Murga Penas EM, Nagel I, Betts MJ, et al. Genes encoding members of the jak-stat pathway or epigenetic regulators are recurrently mutated in T-cell prolymphocytic leukaemia. *Br J Haematol* (2016) 173(2):265–73. doi: 10.1111/bjh.13952

31. Shimoda K, Shide K, Kameda T, Hidaka T, Kubuki Y, Kamiunten A, et al. Tet2 mutation in adult T-cell leukemia/lymphoma. *J Clin Exp Hematop* (2015) 55(3):145–9. doi: 10.3960/jslrt.55.145

32. Marcais A, Waast L, Bruneau J, Hanssens K, Asnafi V, Gaulard P, et al. Adult T cell leukemia aggressivenness correlates with loss of both 5-hydroxymethylcytosine and tet2 expression. *Oncotarget* (2017) 8(32):52256–68. doi: 10.18632/oncotarget.13665

33. da Silva Almeida AC, Abate F, Khiabanian H, Martinez-Escala E, Guitart J, Tensen CP, et al. The mutational landscape of cutaneous T cell lymphoma and sezary syndrome. *Nat Genet* (2015) 47(12):1465–70. doi: 10.1038/ng.3442

34. Chang LW, Patrone CC, Yang W, Rabionet R, Gallardo F, Espinet B, et al. An integrated data resource for genomic analysis of cutaneous T-cell lymphoma. *J Invest Dermatol* (2018) 138(12):2681–3. doi: 10.1016/j.jid.2018.06.176

35. Park J, Yang J, Wenzel AT, Ramachandran A, Lee WJ, Daniels JC, et al. Genomic analysis of 220 ctcls identifies a novel recurrent gain-of-function alteration in rltpr (P.Q575e). *Blood* (2017) 130(12):1430–40. doi: 10.1182/blood-2017-02-768234

36. Ward PS, Patel J, Wise DR, Abdel-Wahab O, Bennett BD, Coller HA, et al. The common feature of leukemia-associated idh1 and idh2 mutations is a neomorphic enzyme activity converting alpha-ketoglutarate to 2-hydroxyglutarate. *Cancer Cell* (2010) 17(3):225–34. doi: 10.1016/j.ccr.2010.01.020

37. Xu W, Yang H, Liu Y, Yang Y, Wang P, Kim SH, et al. Oncometabolite 2-hydroxyglutarate is a competitive inhibitor of alpha-ketoglutarate-dependent dioxygenases. *Cancer Cell* (2011) 19(1):17–30. doi: 10.1016/j.ccr.2010.12.014

38. Wang C, McKeithan TW, Gong Q, Zhang W, Bouska A, Rosenwald A, et al. Idh2r172 mutations define a unique subgroup of patients with angioimmunoblastic T-cell lymphoma. *Blood* (2015) 126(15):1741–52. doi: 10.1182/blood-2015-05-644591

39. Muto H, Sakata-Yanagimoto M, Nagae G, Shiozawa Y, Miyake Y, Yoshida K, et al. Reduced tet2 function leads to T-cell lymphoma with follicular helper T-cell-like features in mice. *Blood Cancer J* (2014) 4:e264. doi: 10.1038/bcj.2014.83

40. Sakata-Yanagimoto M, Enami T, Yoshida K, Shiraishi Y, Ishii R, Miyake Y, et al. Somatic rhoa mutation in angioimmunoblastic T cell lymphoma. *Nat Genet* (2014) 46 (2):171–5. doi: 10.1038/ng.2872

41. Yoo HY, Sung MK, Lee SH, Kim S, Lee H, Park S, et al. A recurrent inactivating mutation in rhoa gtpase in angioimmunoblastic T cell lymphoma. *Nat Genet* (2014) 46 (4):371–5. doi: 10.1038/ng.2916

42. Zang S, Li J, Yang H, Zeng H, Han W, Zhang J, et al. Mutations in 5-methylcytosine oxidase tet2 and rhoa cooperatively disrupt T cell homeostasis. *J Clin Invest* (2017) 127(8):2998–3012. doi: 10.1172/JCI92026

43. Ng SY, Brown L, Stevenson K, deSouza T, Aster JC, Louissaint A Jr., et al. Rhoa G17v is sufficient to induce autoimmunity and promotes T-cell lymphomagenesis in mice. *Blood* (2018) 132(9):935–47. doi: 10.1182/blood-2017-11-818617

44. Cortes JR, Ambesi-Impiombato A, Couronne L, Quinn SA, Kim CS, da Silva Almeida AC, et al. Rhoa G17v induces T follicular helper cell specification and promotes lymphomagenesis. *Cancer Cell* (2018) 33(2):259–73 e7. doi: 10.1016/j.ccell.2018.01.001

45. Nguyen TB, Sakata-Yanagimoto M, Fujisawa M, Nuhat ST, Miyoshi H, Nannya Y, et al. Dasatinib is an effective treatment for angioimmunoblastic T-cell lymphoma. *Cancer Res* (2020) 80(9):1875–84. doi: 10.1158/0008-5472.CAN-19-2787

46. Jaiswal S, Fontanillas P, Flannick J, Manning A, Grauman PV, Mar BG, et al. Age-related clonal hematopoiesis associated with adverse outcomes. *N Engl J Med* (2014) 371(26):2488–98. doi: 10.1056/NEJMoa1408617

47. Busque L, Patel JP, Figueroa ME, Vasanthakumar A, Provost S, Hamilou Z, et al. Recurrent somatic tet2 mutations in normal elderly individuals with clonal hematopoiesis. *Nat Genet* (2012) 44(11):1179–81. doi: 10.1038/ng.2413

48. Genovese G, Kahler AK, Handsaker RE, Lindberg J, Rose SA, Bakhoum SF, et al. Clonal hematopoiesis and blood-cancer risk inferred from blood DNA sequence. *N Engl J Med* (2014) 371(26):2477–87. doi: 10.1056/NEJMoa1409405

49. Nguyen TB, Sakata-Yanagimoto M, Asabe Y, Matsubara D, Kano J, Yoshida K, et al. Identification of cell-type-specific mutations in nodal T-cell lymphomas. *Blood Cancer J* (2017) 7(1):e516. doi: 10.1038/bcj.2016.122

50. Schwartz FH, Cai Q, Fellmann E, Hartmann S, Mayranpaa MI, Karjalainen-Lindsberg ML, et al. Tet2 mutations in B cells of patients affected by angioimmunoblastic T-cell lymphoma. *J Pathol* (2017) 242(2):129–33. doi: 10.1002/path.4898

51. Lewis NE, Petrova-Drus K, Huet S, Epstein-Peterson ZD, Gao Q, Sigler AE, et al. Clonal hematopoiesis in angioimmunoblastic T-cell lymphoma with divergent evolution to myeloid neoplasms. *Blood Adv* (2020) 4(10):2261–71. doi: 10.1182/bloodadvances.2020001636

52. Tsiouplis NJ, Bailey DW, Chiou LF, Wissink FJ, Tsagaratou A. Tet-mediated epigenetic regulation in immune cell development and disease. *Front Cell Dev Biol* (2020) 8:623948. doi: 10.3389/fcell.2020.623948

53. Ko M, Huang Y, Jankowska AM, Pape UJ, Tahiliani M, Bandukwala HS, et al. Impaired hydroxylation of 5-methylcytosine in myeloid cancers with mutant tet2. *Nature* (2010) 468(7325):839–43. doi: 10.1038/nature09586

54. Zhang Q, Zhao K, Shen Q, Han Y, Gu Y, Li X, et al. Tet2 is required to resolve inflammation by recruiting hdac2 to specifically repress il-6. *Nature* (2015) 525 (7569):389–93. doi: 10.1038/nature15252

55. Cull AH, Snetsinger B, Buckstein R, Wells RA, Rauh MJ. Tet2 restrains inflammatory gene expression in macrophages. *Exp Hematol* (2017) 55:56–70 e13. doi: 10.1016/j.exphem.2017.08.001

56. Pan W, Zhu S, Qu K, Meeth K, Cheng J, He K, et al. The DNA methylcytosine dioxygenase tet2 sustains immunosuppressive function of tumor-infiltrating myeloid cells to promote melanoma progression. *Immunity* (2017) 47(2):284–97 e5. doi: 10.1016/j.immuni.2017.07.020

57. Ichiyama K, Chen T, Wang X, Yan X, Kim BS, Tanaka S, et al. The methylcytosine dioxygenase tet2 promotes DNA demethylation and activation of cytokine gene expression in T cells. *Immunity* (2015) 42(4):613–26. doi: 10.1016/j.immuni.2015.03.005

58. Yue X, Lio CJ, Samaniego-Castruita D, Li X, Rao A. Loss of tet2 and tet3 in regulatory T cells unleashes effector function. *Nat Commun* (2019) 10(1):2011. doi: 10.1038/s41467-019-09541-y

59. Yang R, Qu C, Zhou Y, Konkel JE, Shi S, Liu Y, et al. Hydrogen sulfide promotes tet1- and tet2-mediated foxp3 demethylation to drive regulatory T cell differentiation and maintain immune homeostasis. *Immunity* (2015) 43(2):251–63. doi: 10.1016/j.immuni.2015.07.017

60. Wing JB, Tanaka A, Sakaguchi S. Human foxp3(+) regulatory T cell heterogeneity and function in autoimmunity and cancer. *Immunity* (2019) 50 (2):302–16. doi: 10.1016/j.immuni.2019.01.020

61. Fujisawa M, Nguyen TB, Abe Y, Suehara Y, Fukumoto K, Suma S, et al. Clonal germinal center B cells function as a niche for T-cell lymphoma. *Blood* (2022) 140 (18):1937–50. doi: 10.1182/blood.2022015451

62. Lemonnier F, Mak TW. Angioimmunoblastic T-cell lymphoma: more than a disease of T follicular helper cells. *J Pathol* (2017) 242(4):387–90. doi: 10.1002/path.4920

63. Pro B, Horwitz SM, Prince HM, Foss FM, Sokol L, Greenwood M, et al. Romidepsin induces durable responses in patients with relapsed or refractory angioimmunoblastic T-cell lymphoma. *Hematol Oncol* (2017) 35(4):914–7. doi: 10.1002/hon.2320

64. Sawas A, Ma H, Shustov A, Hsu P, Bhat G, Acosta M, et al. Characterizing the belinostat response in patients with relapsed or refractory angioimmunoblastic T-cell lymphoma. *Leuk Lymphoma* (2020) 61(8):2003–7. doi: 10.1080/10428194.2020.1753044

65. Ghione P, Faruque P, Mehta-Shah N, Seshan V, Ozkaya N, Bhaskar S, et al. T follicular helper phenotype predicts response to histone deacetylase inhibitors in relapsed/refractory peripheral T-cell lymphoma. *Blood Adv* (2020) 4(19):4640–7. doi: 10.1182/bloodadvances.2020002396

66. Cheminant M, Bruneau J, Kosmider O, Lefrere F, Delarue R, Gaulard P, et al. Efficacy of 5-azacytidine in a tet2 mutated angioimmunoblastic T cell lymphoma. *Br J Haematol* (2015) 168(6):913–6. doi: 10.1111/bjh.13170

67. Tobiasson M, Pandzic T, Cavelier L, Sander B, Wahlin BE. Angioimmunoblastic T-cell lymphoma and myelodysplastic syndrome with mutations in tet2, dnmt3 and cux1 - azacitidine induces only lymphoma remission. *Leuk Lymphoma* (2019) 60 (13):3316–9. doi: 10.1080/10428194.2019.1627541

68. Saillard C, Guermouche H, Derrieux C, Bruneau J, Frenzel L, Couronne L, et al. Response to 5-azacytidine in a patient with tet2-mutated angioimmunoblastic T-cell lymphoma and chronic myelomonocytic leukaemia preceded by an ebv-positive large B-cell lymphoma. *Hematol Oncol* (2017) 35(4):864–8. doi: 10.1002/hon.2319

69. Yoon SE, Cho J, Kim YJ, Kim SJ, Kim WS. Real-world efficacy of 5-azacytidine as salvage chemotherapy for angioimmunoblastic T-cell lymphoma. *Clin Lymphoma Myeloma Leuk* (2022) 22(11):e972–e80. doi: 10.1016/j.clml.2022.07.009

70. Marchi E, Zullo KM, Amengual JE, Kalac M, Bongero D, McIntosh CM, et al. The combination of hypomethylating agents and histone deacetylase inhibitors produce marked synergy in preclinical models of T-cell lymphoma. *Br J Haematol* (2015) 171(2):215–26. doi: 10.1111/bjh.13566

71. Scotto L, Kinahan C, Douglass E, Deng C, Safari M, Casadei B, et al. Targeting the T-cell lymphoma epigenome induces cell death, cancer testes antigens, immune-modulatory signaling pathways. *Mol Cancer Ther* (2021) 20(8):1422–30. doi: 10.1158/1535-7163.MCT-20-0377

72. Falchi L, Ma H, Klein S, Lue JK, Montanari F, Marchi E, et al. Combined oral 5-azacytidine and romidepsin are highly effective in patients with ptcl: A multicenter phase 2 study. *Blood* (2021) 137(16):2161–70. doi: 10.1182/blood.2020009004

73. Chihara D, Fanale MA, Miranda RN, Noorani M, Westin JR, Nastoupil LJ, et al. The survival outcome of patients with relapsed/refractory peripheral T-cell lymphoma-not otherwise specified and angioimmunoblastic T-cell lymphoma. *Br J Haematol* (2017) 176(5):750–8. doi: 10.1111/bjh.14477

74. Bachy E, Camus V, Thieblemont C, Sibon D, Casasnovas RO, Ysebaert L, et al. Romidepsin plus chop versus chop in patients with previously untreated peripheral T-cell lymphoma: results of the ro-chop phase iii study (Conducted by lysa). *J Clin Oncol* (2022) 40(3):242–51. doi: 10.1200/JCO.21.01815

75. Ruan J, Moskowitz AJ, Mehta-Shah N, Sokol L, Chen Z, Kotlov N, et al. Multicenter phase 2 study of oral azacitidine (Cc-486) plus chop as initial treatment for peripheral T-cell lymphoma. *Blood* (2023) 141(18):2194–205. doi: 10.1182/blood.2022018254

Frontiers in Oncology

Advances knowledge of carcinogenesis and tumor progression for better treatment and management

The third most-cited oncology journal, which highlights research in carcinogenesis and tumor progression, bridging the gap between basic research and applications to imrpove diagnosis, therapeutics and management strategies.

Discover the latest Research Topics

See more →

Frontiers

Avenue du Tribunal-Fédéral 34
1005 Lausanne, Switzerland
frontiersin.org

Contact us

+41 (0)21 510 17 00
frontiersin.org/about/contact