



Synovial Tissue Biopsy Research

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Synovial tissue is a key structure in diarthrodial joints and is the primary target of inflammation in autoimmune arthritis. The study of synovial tissue has developed significantly in the last two decades as arthroscopic and ultrasonographic techniques have allowed visualization and access to synovial biopsy. Further progress in synovial tissue processing and analysis has improved studies of disease pathogenesis, biomarker discovery, and molecular therapeutic targeting with increasingly specialized analytical and technological approaches. In September 2018 the first course on Synovial Tissue Biopsies was convened in Brussels, in this Mini Review these approaches will be described and I will summarize how synovial tissue research advanced.

Keywords: synovial tissue biopsy, rheumatoid arthritis, single cell analysis, biomarkers, synovial tissue biomarkers

KEY POINTS

- Synovial tissue is the target tissue of rheumatoid arthritis (RA).
- Synovial biopsy, under local anesthetic, is safe and well-tolerated by patients
- Cellular and molecular analysis of the synovial tissue of RA patients might identify novel targets for therapy and specific biomarkers.
- Technological advances in single cell and molecular analysis provides new opportunities for discovery.

OPEN ACCESS

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Specialty section:

This article was submitted to
Rheumatology,
a section of the journal
Frontiers in Medicine

Received: 11 December 2018

Accepted: 22 March 2019

Published: 16 April 2019

Citation:

Veale DJ (2019) Synovial Tissue
Biopsy Research. *Front. Med.* 6:72.
doi: 10.3389/fmed.2019.00072

INTRODUCTION AND HISTORY

The main focus of synovial tissue research has been rheumatoid arthritis (RA), as the most prevalent cause of inflammatory synovitis. In the last two decades, considerable advances have been made in the diagnosis and therapy of RA (1). However, early diagnosis and precision medicine remain a challenge. In the 1970's Ralph Schumacher and Barry Bresnihan pioneered synovial biopsy research using the Parker-Pearson needle to obtain biopsies and study the cellular composition of the tissue.

SYNOVIAL JOINT

Normal synovial tissue contains specialized fibroblast-like synoviocytes (FLS) interspersed with macrophages (2). In RA the synovial tissue becomes hypervascular and hyperplastic (**Figure 1**) while microscopic analysis reveals hyperplasia of the intimal lining layer, primarily due to increased accumulation of FLS and macrophage cells in the synovial lining (3). Angiogenesis, the development of new blood vessels is probably an early event enabling infiltration of immune cells such as T cells, B cells and monocytes, and is aberrant resulting in an abnormal blood vessel pattern (4). The new blood vessels appear immature and permit increased leukocyte migration, transforming the synovial tissue into an aggressive "pannus" characterized by release of proinflammatory cytokines from macrophages, T and B cells that stimulate FLS activation and subsequent cartilage and bone destruction (5–7). Although angiogenesis leads to increased blood vessels the tissue is markedly hypoxic *in vivo* (8).

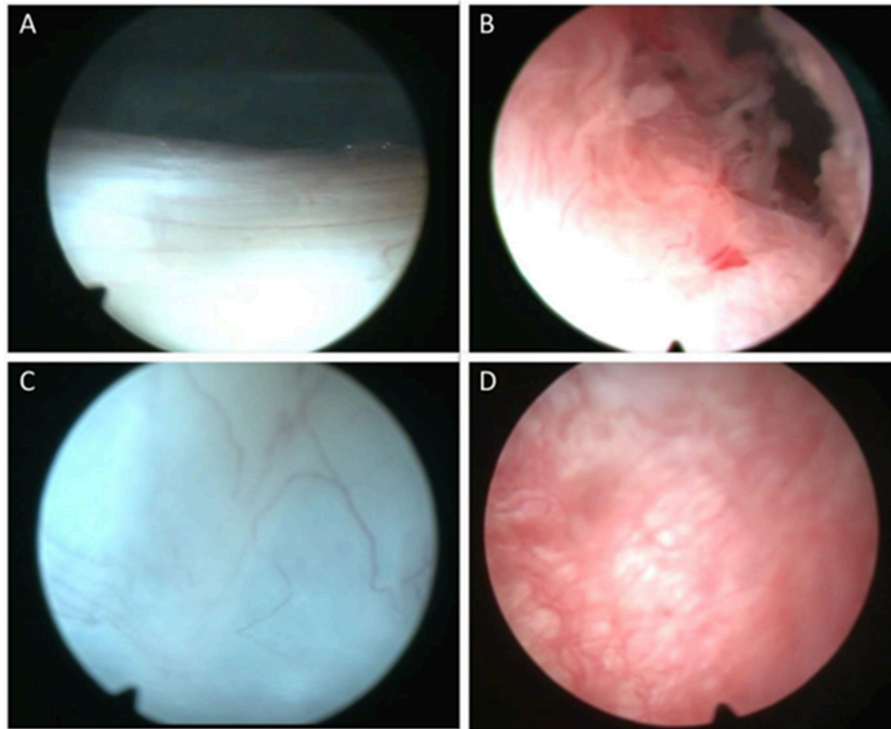


FIGURE 1 | Representative macroscopic appearance of synovial tissue. Macroscopic images of the synovial tissue demonstrating normal synovial tissue (A,C) compared to inflamed and hyperplastic synovial villi in rheumatoid arthritis (B,D).

SYNOVIAL BIOPSY

The analysis of synovial tissue biopsies has advanced our understanding of RA pathogenesis, yielded potential therapeutic targets, and allows detailed evaluation of new therapies (9, 10).

Synovial tissue biopsies have been obtained by blind needle biopsy, arthroplasty, arthroscopy, and more recently using ultrasound- (11). Arthroscopic and ultrasound-guided (USG) biopsy procedures are safe and well-tolerate, both provide good biopsy material. The main benefit of USG appears to be access to small joints, however the yield of synovial tissue is often lower (~80%) (12, 13), while in the authors experience arthroscopy provides 100% synovial tissue yields. There is a low adverse event rates of 0.9% for haemarthrosis, 0.2% for deep vein thrombosis, and 0.1% for both wound infection and joint infection (14). Similarly, a systematic review reported an overall major complication rate of 0.4% for ultrasound-guided biopsy procedures (15).

SYNOVIAL TISSUE ANALYSIS

Immunohistochemical analysis of synovial tissue has a clinical role in the differential diagnosis of arthritis (e.g., infectious, granulomatous, infiltrative, or crystal arthropathies), the benefit in studies of personalized medicine have yet to provide a substantial advance (16). Interestingly though, studies of the synovium beyond immunohistochemistry involving whole-tissue

culture, tissue digestion, homogenization, and single cell analysis with detailed molecular profiling including -omic technologies are now possible (Figure 2). Direct analysis of synovial tissue—the target of inflammation in RA—is critical to the investigation of pathogenesis in RA. Monocytes, T and B cells are expanded in the blood as well as in the synovial tissue of RA patients and this has provided the rationale for development of novel biological therapies including anti-cytokine antibodies, abatacept, and rituximab.

PREDICTORS OF ARTHRITIS AND RESPONSE TO THERAPY

In the last 15 years, synovial tissue analysis has impacted the treatment of early RA using clinical, pathological, and -omic data analysis. A link between circulating ACPAs and the development of RA in subjects with arthralgia, and bone damage has been described in patients with early arthritis (17, 18). The predictive value of a positive ACPA status in RA patients has been reported, however, in those with arthralgia it is highly variable with 30–70% subsequently developing RA on follow-up (19). One study has identified a highly expanded, T cell clone in RA synovial tissue early in the disease underlining the importance of T cells at this stage (20). Epigenetic changes in synovial tissue FLS might also define the different stages of RA after clinical onset (21).

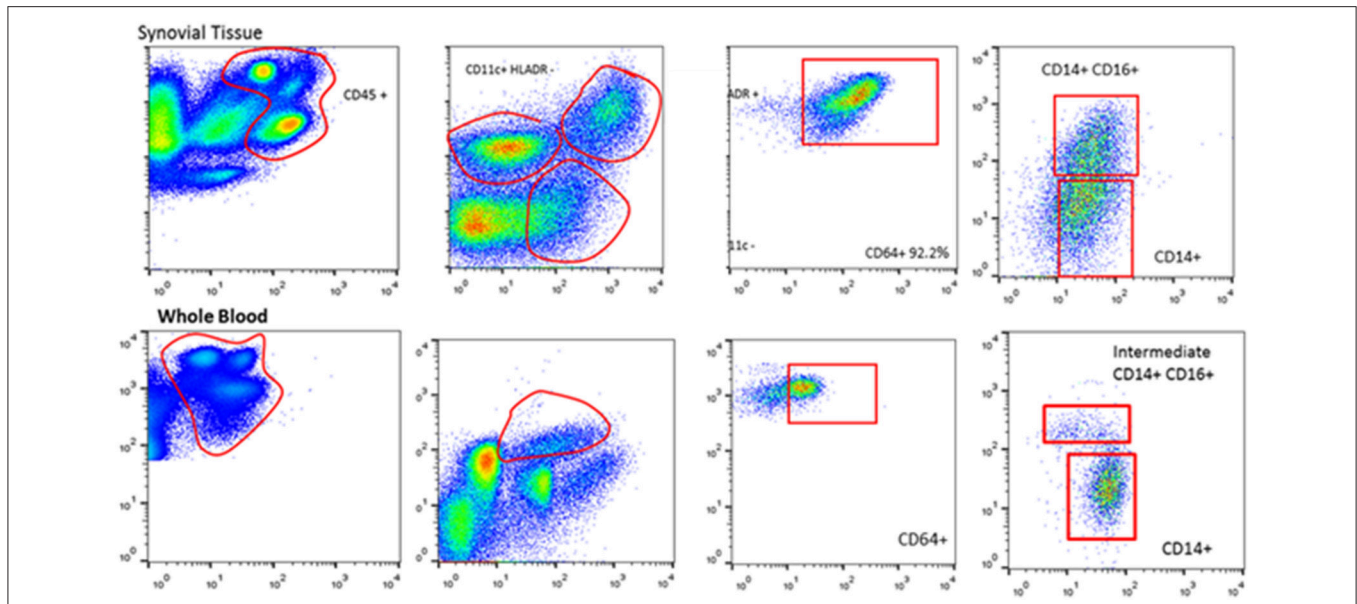


FIGURE 2 | Synovial tissue and whole blood cell isolations with flow cytometry. Flow cytometric analysis of paired peripheral blood and synovial tissue derived cells illustrating the ability to isolate and identify single cell populations of key immune cells.

Inflammatory genes overexpressed in pre-treatment biopsies might predict those RA patients most likely to respond to TNF inhibitor therapy. Another study of synovial tissue RNA suggested that transcripts associated with lymphocyte aggregates predicted response to infliximab therapy (22). The role of macrophages and T cells as biomarkers of response is also supported by gene-expression analyses of paired RA synovial biopsies before and after rituximab treatment, that showed clinical response was greater in those with high expression of macrophage and T cell associated synovial genes (23).

CHALLENGES IN BIOMARKER DISCOVERY

Recent advances in -omic techniques are allowing deeper molecular analysis of synovial tissue, however several challenges remain. The new technologies have become faster, better value and provide a more detailed analyses of genes, proteins, and epigenetic modifications. However, a number of commonly used microarray platforms have yielded poor reproducibility causing some problems with interpretation of data. In addition, the results of initial whole tissue transcriptional profiling await more detailed analysis. Therefore, in the last 2 years, we have developed laboratory techniques to dissociate the synovial biopsies into viable single cell subsets allowing specific analysis of the genes, proteins, and functions of the cell subsets that comprise the population in the actively inflamed synovial joint tissue (24).

CONCLUSIONS

RA is characterized by inflammation of the synovial tissue, which therefore represents the target tissue of autoimmune arthritis. Various methods of sampling synovial tissue have now been validated as safe, well-tolerated by patients and minimally invasive thus they have become more widely practiced. In this Mini review I have focused on the development of synovial tissue biopsy studies including the current technological advances in analysis that allow detailed cellular and molecular experiments that define the functions of immune cells in the RA synovial tissue. These studies might allow greater understanding of the pathogenesis of RA and development of a “precision medicine” approach with improved therapy, patient stratification, development of new therapeutic targets, and development of specific biomarkers of response.

AUTHOR CONTRIBUTIONS

The author confirms being the sole contributor of this work and has approved it for publication.

ACKNOWLEDGMENTS

I wish to thank all my colleagues in the European Synovitis Study Group and in the OMERACT group who have supported the development of synovial tissue research in the last 35 years.

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