



# Natural Killer Cell Response to Chemotherapy-Stressed Cancer Cells: Role in Tumor Immunosurveillance

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Natural killer (NK) cells are innate cytotoxic lymphoid cells that actively prevent neoplastic development, growth, and metastatic dissemination in a process called cancer immunosurveillance. An equilibrium between immune control and tumor growth is maintained as long as cancer cells evade immunosurveillance. Therapies designed to kill cancer cells and to simultaneously sustain host antitumor immunity are an appealing strategy to control tumor growth. Several chemotherapeutic agents, depending on which drugs and doses are used, give rise to DNA damage and cancer cell death by means of apoptosis, immunogenic cell death, or other forms of non-apoptotic death (i.e., mitotic catastrophe, senescence, and autophagy). However, it is becoming increasingly clear that they can trigger additional stress responses. Indeed, relevant immunostimulating effects of different therapeutic programs include also the activation of pathways able to promote their recognition by immune effector cells. Among stress-inducible immunostimulating proteins, changes in the expression levels of NK cell-activating and inhibitory ligands, as well as of death receptors on tumor cells, play a critical role in their detection and elimination by innate immune effectors, including NK cells. Here, we will review recent advances in chemotherapy-mediated cellular stress pathways able to stimulate NK cell effector functions. In particular, we will address how these cytotoxic lymphocytes sense and respond to different types of drug-induced stresses contributing to anticancer activity.

**Keywords:** natural killer cells, immunochemotherapy, cancer, stress, natural killer cell activating ligands, damage-associated molecular patterns, death receptors, PDL-1

## INTRODUCTION

Natural killer (NK) cells represent a crucial component of antitumor innate immune response displaying cytotoxic functions and secreting several cytokines/chemokines (1, 2).

Natural killer cell cytotoxic activity regulation depends on an integrated interplay between inhibitory receptors and numerous activating receptors acting in concert to efficiently eliminate tumor cells.

Relevant activating receptors for tumor cell recognition are NKG2D that recognizes MICA/B and ULBPs proteins, orthologs of the mouse RAE1 molecules, DNAM-1 that binds two ligands named poliovirus receptor (PVR/CD155) and Nectin-2 (CD112), and the receptors NKp30, NKp44, and

NKp46 belonging to the natural cytotoxicity receptors and shown to interact with a broad spectrum of ligands (3).

Natural killer cells also express inhibitory receptors for molecules of the major histocompatibility complex (MHC) class I, which are Ly49 receptors in mice, killer cell immunoglobulin-like receptors (KIRs) that bind to HLA-A, -B, and -C molecules in humans, and the CD94-NKG2A heterodimer in both species (4). In addition, NK cells express two inhibitory receptors for PVR, called TACTILE (CD96) and TIGIT, that counterbalance the DNAM-1-mediated activation of NK cells (5).

The activation of NK cells leads to the release of cytotoxic granules containing perforin and various granzymes and to cytokine production, most prominently interferon- $\gamma$  (IFN- $\gamma$ ) (6–8). In addition, the expression at the cell surface of death-inducing ligands belonging to the tumor necrosis factor (TNF) family, such as Fas ligand (FasL) and TNF-related apoptosis-inducing ligand (TRAIL), also drives the activation of the caspase enzymatic cascade through the binding to the death receptors (DRs), namely, Fas, DR4 (TRAIL-RI), and DR5 (TRAIL-RII), on target cells (9, 10).

More recently, immunological checkpoint molecules commonly associated with T cells, such as CTLA-4 and PD-1, have been described on NK cells as negative regulators of their immune function (11–13).

Conventional chemotherapies were initially designed to produce antiproliferative or cytotoxic effects on dividing tumor cells. However, as result of numerous demonstrations indicating that an endogenous antitumor immunity is essential for complete remission during tumor therapy (14–16) several antineoplastic drugs, even at low doses, have been reconsidered also as potential immunomodulatory agents (17).

In this context, it has becoming always more evident that dying or stressed cells release or expose stress molecules, called damage-associated molecular patterns (DAMPs) that can alert the immune system (18). Moreover, many chemotherapy-mediated stress pathways modulate the expression of NK cell activating and inhibitory ligands, rendering tumor cells more immunogenic.

In this review, we will summarize the effects of different chemotherapeutic agents on the activity of NK cells, emphasizing the immunomodulatory effects of both conventional and low concentrations of drugs at the interface between stressed or dying cancer cells and the immune system, in the attempt of exploiting them for therapeutic purposes.

## REGULATION OF NK CELL-ACTIVATING AND -INHIBITORY LIGAND EXPRESSION BY CHEMOTHERAPEUTIC DRUGS

A number of evidence indicate that chemotherapy-induced sensitization of tumor cells to immune effectors plays an important role in anticancer therapy. Indeed, different types of drug-induced stresses can modulate the expression of NK cell-activating/or -inhibitory ligands on cancer cells thus affecting their recognition and elimination by NK cells (Table 1). Besides genotoxic drugs or radiotherapy, many other pharmacological compounds already approved for the treatment of different malignancies or entered in clinical trials have been described to increase NK cell-activating

ligand expression (19–27). Moreover, most of these drugs are also able to downregulate NK cell-inhibitory ligand expression, so that different and multiple mechanisms concur to make tumor cells more susceptible to NK cell-mediated lysis (28–32).

In the case of genotoxic drugs or DNA replication inhibitors, the mechanisms regulating the NKp30 ligand B7-H6 expression on human cancer cells remain largely unknown (23), while much evidence indicate a major role for the DNA damage response (DDR) pathway in the upregulation of the stimulatory ligands for the NKG2D and DNAM-1 immunoreceptors. In addition, ionizing radiations represent classical stimuli to induce NKG2D ligand upregulation, through the induction of the DDR (33). The activation of the kinases ATM/ATR and the production of reactive oxygen species converge on the E2F1 factor able to activate MICA, MICB, and PVR transcription on multiple myeloma (MM) cells by doxorubicin and melphalan (34). On the other hand, a different pathway governing NKG2DLs expression by chemicals known to induce genotoxic stress has been characterized in murine lymphoma cells: DDR drives to the presence of cytosolic DNA and to STING/TBK1-dependent activation of the transcription factor IRF3, responsible for the upregulation of RAE1 expression (35). Interestingly, in murine leukemia cells, concomitantly to NKG2D ligand upregulation, DDR-activating therapeutic agents cause a loss of the inhibitory NK cell ligand Clr-b, thus enhancing the cytotoxicity mediated by NKRP1B<sup>+</sup> NK cells (36).

Non-lethal heat shock mimicking hyperthermia therapy can promote NKG2DL expression both in human and murine cancer cells but with different mechanisms. MICA and MICB upregulation occurs at the transcriptional level *via* HSF1 activation (37) and, with a similar mechanism, MICA and MICB expression on MM cells is enhanced by HSP90 chaperone inhibitors that activate this transcription factor (21). In a different way, increased surface expression of the mouse NKG2D ligand Mult1 depends on the inhibition of protein ubiquitination and lysosomal degradation (38).

Treatment of different tumor cell types with epigenetic drugs, like histone deacetylase inhibitors (HDACi) and DNA-methyltransferase inhibitors (DNMTi) (25–27, 39–43), leads to the upregulation of NKG2DLs and PVR surface levels, although it downregulates B7-H6 expression (44). For DNMTi the molecular mechanisms underlying NKG2DLs upregulation are still unclear, while different pathways cooperate in the regulation of these molecules in response to HDACi, and this might depend on the type of tumor and the dose of the drug used. In particular, valproic acid (VPA) has been reported to upregulate MICA/B with a mechanism dependent on PI3K/Akt pathway in pancreatic cancer cells (40), while the involvement of ERK in MICA/B and ULBP2 upregulation in response to VPA has been shown in MM cells (45). Moreover, Yang and colleagues proposed that the capability of the HDACi suberoylanilide-hydroxamic acid (SAHA) to increase MICA expression in hepatoma cancer cells is dependent on miR-17-92 cluster (46).

In MM cells, the bromodomain and extra terminal domain inhibitors (BETi) and immunomodulatory drugs (IMiDs) can block the repressive activity of the transcription factors IRF4 and IKZF1/3 on MICA and PVR promoters (19, 47). In addition,

**TABLE 1** | Chemotherapy-induced pathways and molecular targets able to modulate natural killer (NK) cell activating ligands and PDL-1 on cancer cells.

Class of chemotherapeutic agent	Pathway/molecular target	Ligand	NK cell cytotoxicity	Cancer cell type	Reference
<b>PROTEASOME INHIBITOR</b>					
Bortezomib	DNA damage response (DDR)	MICA	nd	Multiple myeloma (MM)	(24)
Low doses: 0.75–10 nM	nd	MICA/B, PVR, Nec-2	+	MM	(52)
	nd	MICA/B ULBP1–3, PVR, Nec-2	nd	MM	(22)
	nd	MICA/B	+	Hepatocellular carcinoma	(114)
<b>HISTONE DEACETYLASE INHIBITORS</b>					
Low dose: valproic acid (1 mM)	nd	MICA/B	+	Hepatocellular carcinoma	(41)
	ERK	MICA/B, ULBP2	+	MM	(45)
	PI3K/Akt	MICA/B	+	Pancreatic cancer	(40)
Trichostatin A	HDAC1/MICA promoter	MICA/B	+	Leukemia	(42)
Suberoylanilide-hydroxamic acid	miR-17-92	MICA	+	Hepatocellular carcinoma	(46)
<b>GENOTOXIC AGENTS</b>					
Low doses: doxorubicin (0.05–3.5 $\mu$ M); melphalan (1.5–22 $\mu$ M)	Reactive oxygen species-dependent DDR	MICA/B, ULBP1–3, PVR, Nec-2	+	MM	(22, 34)
Cisplatin	nd	B7-H6	+	Tumor cell lines	(23)
Ara-C, aphidicolin	STING/TBK/IRF3	RAE1	nd	B cell lymphoma	(35)
<b>GSK INHIBITORS</b>					
Low doses: LiCl (10 mM), BIO (1.5 $\mu$ M), SB21 (5 $\mu$ M)	STAT3 inhibition	MICA	+	MM	(20)
<b>BET INHIBITORS</b>					
Low dose: JQ1 (0.5 $\mu$ M)	IRF4	MICA	+	MM	(19)
	BRD4	PDL-1	nd	Lymphoma	(28)
<b>HSP90 inhibitors</b>					
Low doses: radicicol (2 $\mu$ M), 17-AAG (1 $\mu$ M)	HSR	MICA/B	+	MM	(21)
<b>MICROTUBULE ASSEMBLY INHIBITORS</b>					
Low dose: vincristine (0.05 $\mu$ g/ml)	p38 MAPK	PVR, MICA, ULBP1	+	MM	(50)
Cytochalasin D	DDR	MICA, ULBP1–3, PVR, Nec-2, B7-H6	+	Tumor cell lines	(51)
Nocodazole	Endoplasmic reticulum stress response				
Docetaxel					
<b>IMMUNOMODULATORY DRUGS</b>					
Low dose: lenalidomide (10 $\mu$ M)	IKZF1/3, IRF4	MICA, PVR	+	MM	(47)

Effects on an increased NK cell recognition and killing of drug-treated tumor cells are also reported (+). Low doses of drugs that do not affect cell vitality are indicated. nd, not done.

both these therapeutic agents can downregulate the expression of PD-L1 on cancer cells (28, 29, 31, 32). Indeed, BETi interrupt the activity of the epigenetic reader protein BRD4 on PD-L1 promoter region, by significantly reducing both the constitutive and IFN- $\gamma$  inducible expression of this ligand. In this regard, the downstream mediators of IFN- $\gamma$  signaling, JAK kinases, can be pharmacologically blocked to negatively regulate PD-L1 expression in cancer cells (48). Furthermore, drugs disrupting RAF/MEK/ERK signaling pathway, such as Sorafenib and the TLR3 agonists poly-IC, can synergistically reduce the percentage of tumor cells expressing PD-L1 and enhance NK and T cell activation in a mouse model of hepatocarcinoma (49).

Regarding drugs that disrupt the microtubule assembly, sub-lethal doses of Vincristine can activate p38 MAPK and regulate NKG2DL expression both at transcriptional and post-transcriptional level in MM cells (50). Moreover, Cytochalasin D, nocodazole, and docetaxel can enhance NKG2D, DNAM-1, and NKp30 ligands on tumor cell surface, with MICA upregulation being dependent on both DNA damage and endoplasmic reticulum (ER) stress response (51).

Different studies have been done by using proteasome inhibitors in MM cells. In this regard, low doses of bortezomib can induce the upregulation of both NKG2D and DNAM-1 ligands (22, 52, 53), and in accordance with these data, Jinushi

and colleagues reported a DDR-ATM-dependent upregulation of MICA surface levels (24). On the other hand, no significant change in NKG2DL expression was observed upon bortezomib treatment by Shi and colleagues (30). Interestingly, the latter study described the capability of bortezomib to downregulate HLA class I surface expression by sensitizing MM cells to NK cell-mediated lysis (30).

Chemotherapeutic agents can also contribute to the post-translational regulation of NK activating ligand expression by promoting the release of soluble NKG2DLs through the modulation of the expression and activity of metalloproteinases (MMP) and ADAM enzymes on cancer cells (54). Although an increased stimulation of the shedding process in response to genotoxic agents has been reported (55), some studies using different drugs describe an inhibitory effect. Indeed, gemcitabine treatment impaired ULBP2 shedding through downregulation of ADAM10 in pancreatic cancer (56). Likewise, the hypomethylating agents, azacitidine and decitabine, reduced MICA, MICB, and ULBP2 release in AML by increasing TIMP3 expression, a potent inhibitor of MMP family (57).

Thus, antitumor therapeutics can work also as activators of different “stress pathways” that enhance tumor sensitivity to NK cell cytolysis by modulating the expression of the activating and inhibitory ligands on tumor cells.

## MODULATION OF DRs BY CANCER THERAPEUTIC AGENTS

Many cancer therapeutic drugs can induce DR expression and redistribution (58) (Table 2). Several studies described a role for

different types of HDACi in the upregulation of TRAIL receptors on various malignant tumor cells (59–63). In this context, SAHA and trichostatin A (TSA) were shown to increase cell-surface expression of DR4 and DR5 in human MM cell lines (64). A study from Insinga et al. showed that different DR and their ligands (i.e., TRAIL, DR5, FasL, and Fas) are upregulated by HDACi on leukemic cells, but not in the normal counterpart of hematopoietic progenitors, promoting tumor apoptosis through the activation of the DR pathway (65).

A number of studies showed that bortezomib upregulated surface expression of TRAIL receptors on a variety of human tumor cell lines, enhancing their susceptibility to NK cell lysis with a mechanism mainly dependent on TRAIL (66). In another model, a bortezomib-treated murine renal carcinoma cell line is more susceptible to both NK-cell perforin/granzyme and recombinant TRAIL-mediated apoptosis, resulting in enhanced caspase-8 activity (67). Indeed, in human non-small cell lung cancer cells this drug has been shown to trigger TRAIL-induced apoptosis via DR5 upregulation (68). Several pieces of evidence reported that another proteasome inhibitor, namely, MG132, increases DR5 expression cooperating in establishing apoptosis in several cancer cells (69–71).

DR4 and DR5 were demonstrated to be DNA damaging-inducible and p53-regulated genes (72–76). Accordingly, many DNA damaging chemotherapeutic agents can regulate DR expression, rendering cancer cells more sensitive to DR-elicited apoptosis (74, 75, 77–81).

Altogether, these results suggest that the extrinsic apoptotic pathway has an important role in chemotherapy-induced apoptosis through the promotion of DRs-mediated recognition by cytotoxic lymphocytes. In addition, chemotherapies can promote

**TABLE 2** | Chemotherapy-induced pathways and molecular targets able to modulate death receptors (DRs) on cancer cells.

Class of chemotherapeutic agent	Pathway/molecular target	DR	Cancer cell type	Reference
<b>PROTEASOME INHIBITORS</b>				
Low doses: bortezomib (5–20 nM)	DNA damage response	DR5	Tumor cell lines, renal carcinoma	(66, 67)
MG132	CHOP	DR5	Prostate cancer	(71)
<b>HISTONE DEACETYLASE INHIBITORS</b>				
Sodium butyrate	Sp1	DR5 (caspase-3 activation)	Colorectal carcinoma	(59)
Trichostatin A (TSA), suberoylanilide-hydroxamic acid (SAHA)	p53-independent mechanism	DR5 (caspase member activation)	Tumor cell lines	(60)
Sodium butyrate				
Low doses: SAHA (500 nM), TSA (50 nM)	p21, p27, E2F	DR4, DR5 (increase of proapoptotic Bcl-2 family members)	Multiple myeloma	(64)
VPA	nd	DR5, FAS	Leukemia	(65)
<b>GENOTOXIC AGENTS</b>				
Cisplatin, mitomycin, doxorubicin, methotrexate, etoposide	p53-dependent mechanism	FAS, DR5, DR4	Tumor cell lines	(72–74, 77)
Etoposide	NF- $\kappa$ B	DR5	Tumor cell lines	(76)
Doxorubicin, Ara-C, etoposide	p53-independent mechanism	DR5	Leukemia cell lines	(81)

Low doses of drugs that do not affect cell vitality are indicated. nd, not done.



the cell death by regulating the balance between pro- and anti-apoptotic proteins toward apoptosis. Many evidence show that drugs may control the cell intrinsic apoptosis by altering Bax and Bcl-2 expression in different tumor cells (82–86).

## CHEMOTHERAPY-INDUCED DAMPS ALERTING NK CELLS

Many anticancer chemotherapies increase the immunogenic potential of cancer cells mainly through the establishment of immunogenic cell death, or other forms of non-apoptotic death, including autophagy, and the release of the so-called DAMPs, such as high-mobility group box 1 proteins (HMGB1), ATP, heat shock proteins (HSPs), and the ER chaperone calreticulin (87).

Damage-associated molecular patterns are intracellularly sequestered in normal physiological conditions, but they can be actively secreted or aberrantly exposed on the cell surface under conditions of cellular stress.

Engagement of various target receptors present on immune cells by DAMPs leads to the elicitation of a potent antitumor immunity. Mostly, DAMPs have been proposed to activate local APCs, thus promoting the adaptive immune system. For example, both HSP70 and HMGB1 boost dendritic cell (DC) maturation through toll-like receptor 4, favoring the induction of antigen-specific T cell-mediated antitumor immune responses (88, 89). Less is known about DAMP contribution to NK cell stimulation; thus, we will focus the attention on HMGB1 and HSPs, due to their ability to exert different effects on NK cell-mediated functions.

High-mobility group box 1 protein is an endogenous nuclear factor released both by activated immune cells or injured non-immune cells, and in the extracellular milieu acts as a DAMP alerting the immune system to danger and triggering immune response activation through the interaction either with multiple TLRs and the receptor for advanced glycation end products (RAGE), expressed on a variety of cells (90). In this regard, the chemotherapeutic agent cyclophosphamide has been recently shown to facilitate NK cell activation through a process involving HMGB1 release in a glioma mouse model (91). Accordingly, it was demonstrated that in HMGB1-deficient tumors, different innate immune cells, including NK cells, have impaired ability to reach the tumor tissue in response to DNA alkylating agent treatments (92). In addition, HMGB1 can be released by NK cells and can stimulate NK cell chemotaxis through RAGE, thus further amplifying their response to tumors (93) and can also play an important role in the cross-talk between NK and DC, by promoting DC maturation (94, 95). Interestingly, HGMB1 can induce autophagy (96), which may control the regulation of the innate and adaptive immune responses contributing to enhance antigen processing and presentation (97).

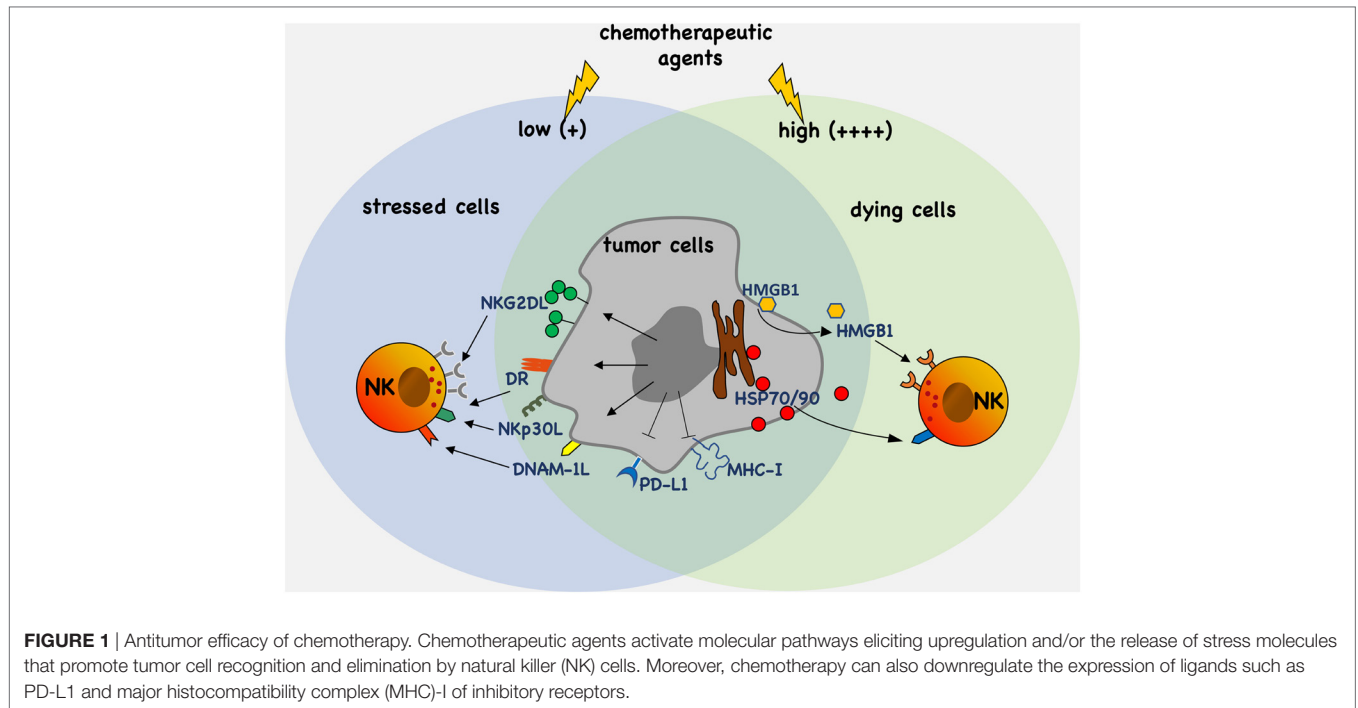
Heat shock proteins are localized in most intracellular compartments where they act as molecular chaperone by supporting protein folding and transport across membranes. Several studies demonstrated an unusual HSP70 cell membrane localization on transformed tumor cells (98–100). As already mentioned, stressful conditions can cause HSPs mobilization to the plasma membrane, or their release from cells, thus acting as potent danger signals. In this respect, therapeutic treatments including

radio and chemotherapy have been shown to produce an augmentation of HSP70 cell-surface expression on tumor cells (101, 102). Several studies have shown that membrane-bound HSP70 directly promotes NK cell mediated cytotoxicity *in vitro* (103, 104) and *in vivo* (105) thus, there is an increasing interest in the therapeutic potential of targeting HSP70. Interestingly, Elsner and colleagues have shown a synergistic potentiating effect of two stress-inducible immunological danger signals HSP70 and NKG2D ligands on cytotoxicity of human (106) and mouse NK cells (107), suggesting that the drug-mediated upregulation of activating ligands and HSP70 on the cancer cell surface might be an encouraging strategy aimed at promoting the antitumor NK cell responses. Moreover, several pieces of evidence demonstrate that extracellular-located HSPs can be associated to extracellular vesicles (108–112), and a number of chemotherapeutic agents, including etoposide (109), melphalan (110), cisplatin, and 5-fluorouracil (112), have been shown to stimulate an enhanced secretion of exosomes from different types of cancer cells. Notably, colon carcinoma-derived HSP70 associated to exosomes can stimulate NK cell migration and cytotoxic activity (108). In addition, we have recently demonstrated that HSP70 on the surface of MM-derived exosomes triggers NK cell-mediated IFN- $\gamma$  production through a mechanism dependent on TLR2 (110).

## DIRECT EFFECTS OF CHEMOTHERAPY ON NK CELL-MEDIATED FUNCTIONS

Alterations of NK cell activities upon administration of chemotherapeutic drugs can be different in terms of cytotoxicity and immunoregulatory activity; indeed, standard chemotherapeutic protocols used in the treatment of cancer patients mainly suppress NK cell-mediated killing against cancer cells and their cytokine production. However, several studies aimed at analyzing the NK cell behavior in patients undergoing cytotoxic chemotherapy have demonstrated different and variable effects depending on both the type and the dose of the drug used.

In this regard, by producing IFN- $\gamma$ , NK cells induce CD8<sup>+</sup> T cells to become CTLs, and also help to differentiate CD4<sup>+</sup> T cells toward a Th1 response. Moreover, NK cell-derived cytokines might also regulate antitumor antibody production by B cells. Thus, therapeutic strategies able to preserve NK functions in cancer patients are of pivotal importance, particularly those eligible for monoclonal antibody-based treatments. In this context, metronomic low cyclophosphamide (CTX) regimen was shown to potentially stimulate NK functions in terms of cytokine production and antitumor immunity (18). A number of drugs, including bortezomib, genotoxic agents, and epigenetic drugs, exert immunosuppressive effects at high concentrations, whereas at sub-lethal doses, they can render tumor cells more immunogenic without affecting the immune cell activity (113). As an example, low doses of bortezomib capable of stimulating NK cell activating ligand expression on MM (22, 52), do not alter NK cell degranulation against sensitive targets (52). In another study, low concentrations of bortezomib reduced IFN- $\gamma$  production without affecting NK cell cytotoxicity (114). Moreover, a combination of bortezomib with exogenous cytokine treatment enhanced the cytotoxic effects of NK cells against cancer cells in



two different models (115, 116). The treatment of NK cells with sub-lethal doses of doxorubicin, able to upregulate NKG2D and DNAM-1 ligands on MM cells, does not change the capacity of NK cell to degranulate in response to target cells, as well as the ability to produce IFN- $\gamma$  (34). Although the wide range of HDACi, structurally different from each other, can have both stimulatory and inhibitory effects on immune cell function, the most of them (i.e., romidepsin, vorinostat, TSA, and VPA) have been shown to suppress NK cell activity at therapeutically relevant concentrations (117–119). However, some reports describe a beneficial effect on NK cells as for the narrow-spectrum HDACi entinostat that can increase NKG2D expression on NK cells without affecting their cytotoxic activity (120). Furthermore, a recent study demonstrates that the HDACi panobinostat has the capability to potentiate the antitumor effects of trastuzumab by stimulating the antibody-dependent cell-mediated cytotoxicity (ADCC) mediated by NK cells (121). Regarding the DNTMi decitabine and 5-azacytidine, treatment of NK cells leads to increased reactivity toward different tumor cells (122, 123), while another study describes that 5-azacytidine exposure compromises their activity in AML and MDS patients (124).

Immunomodulatory drugs (lenalidomide, pomalidomide, and thalidomide) exert strong immunomodulatory effects involving both innate and adaptive immunity. In particular, these compounds activate both NK and T cells by inducing their proliferation, cytokine production, and cytotoxic activity (125) and promising clinical trials have been reported their use for the treatment of hematological malignancies, such as myeloma, lymphoma, and leukemia, as well as of solid tumors (126–128). Interestingly, Lagrue and colleagues demonstrated that lenalidomide enhances NK cell response (IFN- $\gamma$  production and cytotoxicity) by augmenting actin remodeling, thus rendering them

able to respond to lower densities of activating ligands on tumor cells (126). Furthermore, lenalidomide has synergistic effects on NK cell functions when used in combination with monoclonal antibodies able to promote ADCC that are already approved in therapeutic protocols, such as rituximab or elotuzumab (129, 130); indeed, novel strategies in the treatment of MM combines the use of lenalidomide and the anti-inhibitory KIR antibody (IPH2101) (131, 132).

## CONCLUSION

The modulation of the expression and/or the release of stress molecules has emerged as a new paradigm of the therapeutic possibilities associated with the use of chemotherapy (Figure 1). In this context, the characterization of novel drugs and regulatory pathways activated by cellular stress modifiers able to affect tumor growth and, at the same time, to improve the activities mediated by cytotoxic lymphocytes such as NK cells, will importantly contribute to the developing field of chemo-immunotherapy.

## AUTHOR CONTRIBUTIONS

AZ, CF, CB, MC, ASantoni, and ASoriani contributed equally to writing and critically revised the paper.

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**Conflict of Interest Statement:** 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.

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