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Promoting the Effector Function of Natural Killer (NK) Cells with a Focus on Bispecific Redirecting Antibodies

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Dear editor,

Natural killer (NK) cells are recognized as the main constituents of the immune system due to their intrinsic potentials in recognizing and eliminating tumor cells or targeted cells infected by viruses. Recently, the NK cells have been further considered since they can be used in monoclonal antibody (mAb)-based due to the expression of the Fc γ receptor (also known as CD16).

Different studies have reported that the therapeutic efficacy of different cancer therapies (e.g., radiotherapy, chemotherapy, and immunotherapy) is affected by various functionalities of the NK cells (1). Moreover, these therapies also have a significant impact on the functionary of the NK cells (1). In this regard, in the case of radiotherapy, the NK cells irradiated under low dose radiation exhibit enhanced cytotoxicity compared to non-irradiated NK cells (1). However, high dose radiation reduces the viability and antitumor activity of the NK cells (1). Moreover, radiotherapy have the following indirect effects on the NK cells: The modulation of activation and the inhibitory NK ligands on tumor cells, leading to a higher sensitivity of tumor cells to the NK cell-mediated cytotoxicity (by upregulating MICA/B and ULPB1-3 and downregulating the KIR2D ligands such as HLA-ABC and HLA-G), the production and secretion of damage-associated molecular patterns (DAMPs) by tumor cells, and the improvement of the NK cell tumor trafficking (e.g., by inducing tumor cells to increase the production of CXCL16 as a CXCR6 ligand) (1, 2).

Furthermore, several studies on chemotherapy have documented that chemotherapeutic agents directly affect the functionality of the NK cells by reducing their viability and activity and modulating the upregulation of the PD-1 expression on these cells (1). Moreover, these cytotoxic agents also tend to have indirect effects on the NK cells. In this regard, they induce tumor cells to upregulate the TRAIL ligands and PD-L1 and secrete DAMPs (1). These agents also mediate the modulation of the NKG2D, DNAM1, and NKp30 ligands by cancer cells known as NK cell-activating ligands (1).

Considering protein kinase inhibitors, the JAK inhibitors suppress the cytotoxic activity of the NK cells; however, Scr kinase inhibitors, BRAF inhibitors, GSK3β inhibitors, and TAM inhibitors are among protein kinase inhibitors promoting the cytotoxic activity of the NK cells (1).

Recently, T cell-redirecting bispecific antibodies (TRBAs) have been of higher interest due to the promising preclinical and clinical outcomes (3, 4). More specifically, a TRBA results from the fusion of two single-chain variable fragments (scFv) via a linker. Of note, an scFv is a fusion protein made of the light chain (VL) and the heavy chain (VH) variable regions of a full-length mAb (5). The linker peptide used in scFvs is usually a glycine-rich flexible peptide among 10 to 25 amino acids (5). The average molecular weight of an scFv is about 27 kDa. scFvs are popular alternatives to full-length mAbs since they, despite the presence of a linker, retain the specificity of the original antibody (5).

In the conventional TRBA, one of the scFvs is specific to a tumor marker of interest, and the other scFv recognizes and binds the CD3 receptor present on the T cells (3, 4). As the other bispecific mAbs, TRBAs establish a bridge between the T cells and the malignant cells, against which one of their scFv is redirected. Independent of major histocompatibility complex (MHC) molecules or the other relevant costimulatory molecules, TRBAs make the T cells enforce tumoricidal effects against the tumor cells of inter-

est (3, 4). To date, several TRBAs developed against different target antigens involved in a series of oncological indications have been under preclinical and clinical investigation. In 2014, the US Food and Drug Administration (FDA) granted Blinatumomab (under the trade name Blincyto) approval for the treatment of individuals with Philadelphia chromosome-negative relapsed or refractory acute lymphoblastic leukemia (ALL).

Furthermore, Solitomab, an EpCAM-specific TRBA, has also been under clinical investigation (NCT00635596) in Germany. In particular, this trial was to evaluate the safety index and the maximal tolerable dose of Solitomab in 65 patients with lung, prostate, colon, gastric, and ovarian cancers. In addition to the aforementioned TRBAs, some other candidate therapeutics have also been developed using this platform to target different antigens, indicating different malignancies. For example, TRBAs against CD66e, EphA2, HER2, EGFR, CSPG4, and CD33 have been developed or are currently under development, highlighting the potential of this approach to developing potential therapeutics (3, 4). Moreover, the other target antigens could also be considered to develop TRBAs against a wide range of solid and hematologic malignancies (all of which have been comprehensively discussed elsewhere) (6, 7).

In addition to the T cells, the NK cells might also be exploited for therapeutic purposes in a similar approach to TRBAs. To be more specific, the NK cell-redirecting bispecific antibodies (NKRBAs) can also be introduced simply by fusing two scFvs via a linker peptide of interest. One of the scFvs would target CD16 to engage the NK cells, and the other ones should be a tumor marker-specific scFv.

Moreover, single variable domain on a heavy chain (VHH) antibodies, also known as Nanobodies®, are made of the antigen-binding region of heavy-chain only antibodies (HcAb) (8). With a molecular weight of about 15kD, VHHs are tiny in size, facilitating their unique applications in various diagnostic and therapeutic fields (8). In this regard, our research team is currently developing a novel bispecific VHH based on the fusion of a CD16-specific VHH to an EGFR-specific VHH. This construct is hoped to induce strong NK cell effector responses against EGFR-expressing tumors in preclinical and clinical investigations.

In addition to developing anti-cancer mAb-based therapeutics, NKRBAs could also be developed to eliminate viruses. Regarding SARS-CoV-2, our research team is currently developing a novel therapeutic underpinned by the fusion of a CD16-specific VHH to the extracellular domain of ACE2, to which SARS-CoV-2 spike protein binds (9). This therapeutic might contribute to eliminating the SARS-CoV-2 viral particles after careful preclinical and clinical evaluations (9).

Footnotes

Authors' Contribution: PSKs structured, wrote, and revised the manuscript. AS planned, structured, and contributed to the writing and revision of the manuscript.

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