Antitumoral immune response: NKT cell-mediated immunotherapy

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Introduction

Cancer is a group of related diseases characterized by an uncontrolled growth of cells beyond their limits that can happen in any tissue of the body. Cancer awareness has been increasing because of its high incidence, morbidity and mortality, being a key subject in research for new, more effective therapies.

The immune system is able to fight against tumors with a process known as cancer immunoediting. In this aspect, natural killer T (NKT) cells have been proved useful to control tumor growth and are therefore being studied for potential new strategies. Two main subsets of NKT cells have been described, type I or iNKT cells and type II or vNKT cells, with opposing roles in mediating tumor immunity. The therapeutic studies focus on iNKT cell activity because they generally enhance tumor immunity, taking advantage of both direct and adjuvant iNKT cell activity. So far the results are preliminary but promising.

Aims

- Describe NKT cells and their general role in the immune system, characterizing the two main subtypes of NKT cells as well as the immunoregulatory axis in which they are involved.
- Explain the role of NKT cells in tumor immunity, focusing in the mechanisms by which iNKT cells seem to mediate cell response against tumors.
- Describe the current state of the iNKT cell-based immunotherapy and the main experimental procedures used in former studies.

Methodology

Search in Pubmed database and in official websites of the World Health Organization and Clinical Trials using the following keywords: NKT cells, iNKT cells, tumor, immunotherapy. Original and review papers were selected according to their date of publication, quality and specific topic.

RESULTS

Functions and characterization of NKT cells

NKT cells are an heterogeneous subset of lymphocytes that express receptors present both in NK cells (NK1.1) and T cells (αβ TCR and CD3). The TCR recognizes glycolipid antigens presented by **CD1d**, a class I-like MHC molecule.

Two subsets of NKT cells are defined according to heterogeneity of TCR rearrangements:

Type I NKT cells (iNKT)	Type II NKT cells (vNKT)
Invariant TCR	Diverse TCR
Reactive with α-GalCer	Reactive with sulfatides
Enhance tumor immunity	**Suppress** tumor immunity

Functions in general immunity

They play an important intermediary role between innate and adaptive immunity, being able to respond quickly to stimuli with abundant production of cytokines (mainly T_H1 and T_H2) that influence the polarization of the adaptive response.

Depending on the specific subset and nature of activation, they have immune enhancing or immunosuppressive roles. Type I and type II NKT cells have contrasting roles in most of the cases and cross-regulate each other.

Tolerance | Autoimmunity | Allergies | Infections | Tumor immunosurveillance

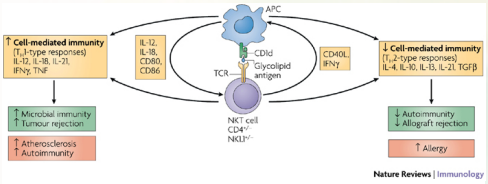


Fig. 1- NKT cell functions. Positive responses for the host are represented in green, while deleterious responses are represented in red (1)
CD40L, CD40 ligand; IFNγ, interferon-γ; IL, interleukin; TCR, T-cell receptor; TGFβ, transforming growth factor-β; TNF, tumour-necrosis factor

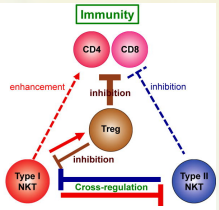
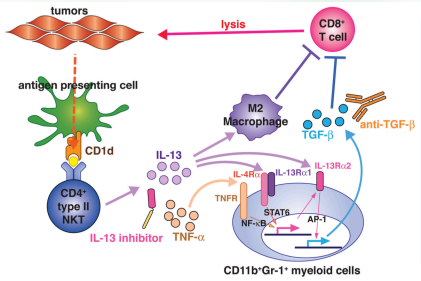


Fig. 2- Relationship among type I NKT cells, type II NKT cells and regulatory T cells (2)

vNKT cell regulatory pathway



CD4+ vNKT produce IL-13, which together with TNF-α present in the microenvironment induces CD11b+Gr-1+ myeloid cells to secrete TGF-β, that suppresses specific cytotoxic T cells (CD8+) activity against the tumor. In some tumors, IL-13 can suppress CD8+ T cells stimulating M2 macrophages.

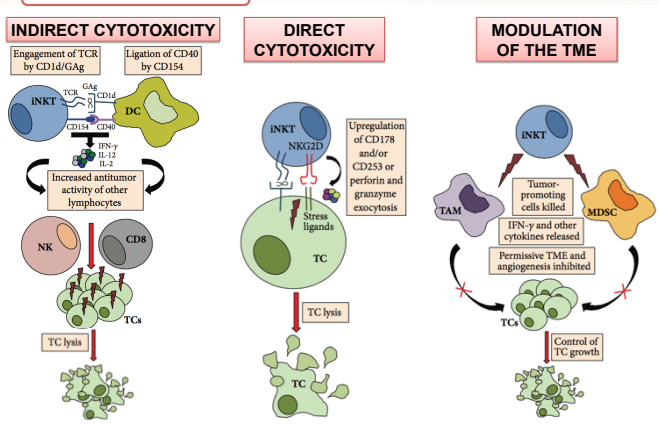
The pathway can be completely abrogated by:

- removing NKT or myeloid cells
- blocking IL-13 or TGF-β

Fig. 3- IL-13 produced by vNKT cells suppresses cytotoxic T cell tumor immune surveillance through the IL-4R-STAT6 pathway to induce TGF-β production by CD11b+Gr-1+ cells (3)

iNKT cells in tumor immunosurveillance

Mechanisms of action



iNKT cells produce T_H1 cytokines, mainly IFN-γ, that contribute to dendritic cell maturation and activate downstream effector cells with cytotoxic activity (NK cells and $CD8^+$ T cells)	iNKT cells produce perforin and granzyme and present upregulation of CD178 (or FasL) or CD253 (TRAIL), showing direct direct cytotoxicity against tumor cells expressing CD1d	iNKT cells target tumor-supporting cells, like TAMs and MDSCs, and limit angiogenesis, setting an immune-permissive environment that allows them to control tumor growth

Fig. 4- Possible mechanisms of iNKT cell-mediated antitumor responses (4)
Lightning bolt: exertion of direct cytotoxicity; DC: dendritic cell; GAg: glycolipid antigen; TC: tumor cell; TAM: tumor-associated macrophage; MDSC: myeloid-derived suppressor cell; TME: tumor microenvironment.

Clinical approaches

Use of iNKT cells in cancer immunotherapy

- **Stimulation of iNKT cells:**
  - **Direct administration of α-GalCer:** it is the strongest activator of iNKT cell activity, producing both direct and indirect anti-tumor activities. It inhibits tumor growth and increases survival in murine models. However, in clinical trials, no significant clinical responses were observed.
  - **Administration of APCs pulsed with α-GalCer:** it induces better anti-tumor responses from NKT cells than free α-GalCer administration while maintaining safety. Clinical trials reported partial responses.
- **Transfer of activated iNKT cells to the patient:**
  - ***Ex vivo* activation of autologous iNKT cells:** Isolated iNKT cells or PBMCs (which include iNKT cells, among other effector cells) from the patients were expanded and stimulated *in vitro* and infused back to patients. The best results were observed when iNKT and α-GalCer-pulsed APCs are infused together.
  - **Adoptive transfer therapy of iPS-derived iNKT cells:** Functional iNKT cells are generated from induced pluripotent stem cells and infused to patients. Better results are obtained if an activator of T_H1 polarization is also infused. This method is especially important in patients with low levels of NKT cells.

Conclusions and future prospects

- NKT cells are intermediates between innate and adaptive immunity, mediating immune regulatory functions.
- NKT cells are divided in iNKT cells and vNKT cells according their TCR rearrangements, and in consequence respond to distinct lipid antigens presented by CD1d.
- iNKT cells potentiate the antitumor response whereas vNKT cells suppress it.
- iNKT cells mediate antitumor response through 3 mechanisms: indirect and direct cytotoxicity and modulation of tumor microenvironment.
- iNKT cell-based cancer therapy has two main approaches: stimulation of the patient's iNKT cells through injection of exogenous ligands or infusion of iNKT cells activated *in vitro*.
- Differences of behavior of the different subsets, the simultaneous unspecific activation of several subfamilies of NKT cells, the influence of the microenvironment and the severity of cancer in the patients undergoing the studies could partially explain the lack of conclusive results.

Together these results indicate that iNKT cell-based cancer immunotherapy strategies are promising. However, more studies to understand the mechanisms of action and the interactions with other cell populations are required.

References

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