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Abstract

Natural Killer T (NKT) cells are a conserved subpopulation of lymphocytes that recognize glycolipid antigens in a CD1d context. Upon activation through their semi-invariant T cell receptor, these cells rapidly release large amounts of immunomodulating Th1 and Th2 cytokines. NKT cells have therefore been implicated in immune responses controlling various diseases including infection, cancer, transplantation and autoimmunity. Stimulation of the immunoregulatory capacity of NKT cells by the prototypical antigen α-galactosylceramide, results in amelioration of disease in several animal models. This review will focus on the current knowledge of human NKT cells and their role in autoimmune diseases. The features of these cells and their importance in regulation of autoimmunity suggest that NKT cell based therapies might be an interesting approach for the treatment of autoimmune diseases.

Introduction

The term “NK T cells” was first used in mice to define a subset of T cells that expressed the NK cell associated marker NK1.1 (CD161). It is now generally accepted that the term “NKT cells” refers to CD1d-restricted T cells, present in mice and humans, co-expressing a heavily biased, semi-invariant TCR and NK cell markers [1]. This TCR consists in humans of an invariant TCR Vα24Jα18 chain combined with a variable TCR Vβ11 chain (Vα14Jα281 and Vβ8, Vβ7 or Vβ2 chains on mouse NKT cells) [2,3,4,5,6]. These properties distinguish NKT cells from conventional $CD4^+$ and $CD8^+$ T cells, which use a diverse TCR to recognize peptide antigens bound by polymorphic MHC molecules.

In humans, only 0.2% of peripheral blood T cells are NKT cells. They are also found in the human liver but their numbers are lower than in liver of mice [7,8,9]. Interestingly, NKT cells express markers associated with recently activated or memory T cells, even in germ-free mice and in human cord blood [10,11]. This suggests that these cells might recognize an endogenous antigen present in the periphery [12].

Human and mouse NKT cells segregate into $CD4^+CD8^-$ and $CD4^-CD8^-$ (double negative, DN) cell subsets which differ in their functional properties. In humans, about 40 to 60% of invariant NKT cells are $CD4^+$, with high donor-to-donor variability, while the remaining cells lack CD4 expression. In humans, CD8α expression is common, but only very few $CD8\beta^+$ NKT cells exist (CD8 expression is even absent in mice) [13,14]. The $CD4^+$ subset potently produces both Th1 and Th2 cytokines whereas the DN population selectively produces Th1 cytokines IFN-γ and TNF-α and preferentially upregulates perforin in response to IL-2 or IL-12 [13,15]. Additionally, some chemokine receptors are differentially expressed on the subsets (CCR4 on $CD4^+$ NKT cells; CCR1, CCR6 and CXCR6 on DN NKT cells), suggesting different functions for these populations [16],[17].

Thymic development of NKT cells

Due to the unavailability of a specific marker for NKT cells, it was first thought that $NK1.1^+$ T cells (first considered as NKT cells) could develop in the liver, independent of the thymus [18]. Most recent experimental studies however, conclude that NKT cells are a thymus dependent population that has undergone TCR β selection [19]. The TCR α chain of NKT cells is invariant at the amino acid level, but can vary at the nucleotide

level, which suggests that the characteristic NKT TCR gene rearrangement is achieved randomly alongside TCR β and α chain rearrangement of mainstream precursors [19].

The divergence of double positive ($CD4^+CD8^+$) thymocytes to the NKT cell lineage is likely to be triggered by positive selection. Inhibition of CD1d trafficking through endosomes or inhibition of endosomal proteases all result in the failure of NKT cell positive selection [20,21], thereby demonstrating the importance of the selecting ligand. A significant discovery has been made by the identification of the lysosomal glycosphingolipid isoglobotrihexosylceramide (iGb3) as possible mediator of NKT cell development, since mice lacking β-hexosaminidase b, the enzyme needed for iGb3 production, show a severe reduction in NKT cells [22]. However, the presence of this glycolipid in the thymus of mice and humans remains to be formally demonstrated.

This molecule has to be presented in a CD1d context as confirmed by the NKT cell deficiency of $CD1d^{-/-}$ animals [23]. Interestingly, the CD1d/glycolipid complex must be presented to the developing NKT cell by hematopoietically derived $CD4^+CD8^+$ thymocytes, rather than thymic epithelial cells responsible for conventional positive selection [24]. Possibly, $CD4^+CD8^+$ cells provide essential signaling for entry to the NKT lineage which is not available on the epithelial cells. Homotypic SLAM family receptor interactions, together with SLAM-associated protein (SAP)-FynT association, appear to play a critical role in this signaling in both mice and humans [25,26]. Negative selection of NKT cells is mediated by dendritic cells, occurs at an earlier stage and the window of susceptibility is far narrower for NKT cells than for other T cells [27,28].

In mice, only 30 % of the NKT cells that are exported to the periphery express a more mature phenotype, which suggests that NKT cells undergo a late phase of peripheral development [29]. Human NKT cells may follow a similar path of thymic development,

as shown by the need for SAP in human NKT cell development. Additionally, it was reported that the number of human $CD4^+$ NKT cells in peripheral blood is mainly determined by thymic output, while $CD4^-$ NKT cells undergo extensive peripheral expansion [30]. However, both subsets further develop their functions in the periphery, suggesting a similar peripheral maturation as mouse NKT cells.

Glycolipid antigen presentation

NKT cells are activated by glycolipid antigens presented in the monomorphic MHC I-like molecule, CD1d [2]. The CD1d molecule is highly conserved among mammalian species [5,31]. It is primarily expressed on cells of hematopoietic origin, including thymocytes, B cells, macrophages and dendritic cells (DC) and can be induced in T cells upon activation [32].

CD1d trafficking has been studied in great detail in mice. After assembly with β2-microglobulin in the endoplasmatic reticulum, CD1d is rapidly transported from the Golgi apparatus to the plasma membrane along the secretory pathway [33]. Internalization from the plasma membrane is dependent on the CD1d cytoplasmatic tail, which directs trafficking through early and late endosomes to lysosomes [34]. Normal endosomal targeting of CD1d, presence of CD1d molecules in lysosomes and internalization of the glycolipid by the APC are essential for successful antigen presentation [35,36]. The binding cleft of the CD1d molecule is narrower but deeper than that found in conventional MHC class I and class II molecules and consists of two binding pockets. The non-polar lining of the antigen binding groove makes the molecule ideal for the presentation of hydrophobic antigens, such as glycolipids [37].

NKT cell research has significantly expanded with the discovery of α-galactosylceramide (α-GalCer), a glycolipid that binds to CD1d molecules and

selectively activates both mouse and human NKT cells [38]. α-GalCer was originally isolated from the sea sponge *Agelas mauritianus* during a screen for novel antitumor molecules [39]. As a result of this discovery α-GalCer loaded, genetically engineered, CD1d-tetramers have been used extensively to study NKT cell biology [8]. A truncated form of α-GalCer, OCH, could also activate NKT cells, but induced a higher production of Th2 cytokines in NKT cells than α-GalCer [40]. However, since α-glycosphingolipids are not present in mammalians, α-GalCer probably mimics self-antigens that are recognized by NKT cells. In search of the endogenous ligand of NKT cells, the first molecule identified to be associated with CD1d in major amounts *in vivo* was glycosylphosphatidylinositol (GPI), but it could not stimulate NKT cells [41]. It was therefore hypothesized that GPI could act as a chaperone, shielding the hydrophobic ligand-binding groove during CD1d trafficking until the physiological NKT cell ligand is encountered in an endosomal compartment [42]. The disialoganglioside GD3, commonly found on tumors of neuroectodermal origin, could activate a subset of α-GalCer reactive mouse NKT cells [43]. More importantly, it has now been reported that both human and mouse NKT cells are activated by the lysosomal glycosphingolipid iGb3, identifying it as a (possibly the) natural NKT cell antigen, involved in NKT cell development and NKT cell immune function [22].

NKT cell response to activation

The most striking property of NKT cells is their capacity to secrete large amounts of cytokines (IFN-γ, IL-4, IL-2, IL-5, IL-10, IL-13, GM-CSF and TNF-α) within minutes after TCR stimulation. This feature distinguishes them from naïve MHC class I and II restricted T cells that acquire their ability to secrete cytokines during proliferation after

primary stimulation. Unlike conventional T cells, mouse NKT cells activate IL-4 and IFN-γ transcription during thymic development and populate the periphery with both cytokine loci previously modified by histone acetylation [44]. Abundant mRNA transcripts for IL-4 and IFN-γ were detected in resting NKT cells which may allow the rapid production and secretion of these cytokines [44]. Activation of NKT cells also leads to upregulation of CD40L, resulting in IL-12 production by dendritic cells upon CD40 triggering [45]. Upon TCR engagement, NKT cells also have cytotoxic activities through the release of perforins and granzymes and by the expression of membrane bound members of the TNF family (such as FasL) [46]. Quickly upon activation, NKT cells become undetectable when assessed by flow cytometry with α-GalCer loaded CD1d-tetramer staining, which results from downregulation of the TCR [47,48]. Similarly, NK1.1 becomes downregulated in mice within 8 to 12 hours. The former is re-expressed within 24 hours, while the latter remains suppressed for up to 2 weeks. The cells dramatically expand even up to 10-fold their normal numbers within 2-3 days of stimulation before contracting to baseline levels on subsequent days [47,48]. Activation of NKT cells in vivo leads to subsequent activation of other cells, such as NK cells, B cells, DC, macrophages and conventional T cells in mice as well as humans [49,50]. Consequently, NKT cell activation results in a cascade of immune reactions, providing a possible explanation for their regulatory effects.

Effector and regulatory functions of NKT cells *in vivo*

Numerous studies have shown that NKT cells can influence the Th1/Th2 balance in immune responses against infectious agents, tumors, alloantigens and self antigens. NKT cells contribute to host responses against a variety of pathogens, including

bacteria, fungi, protozoa, helminthes and viruses (reviewed in [51]). In most cases, NKT cell protection against disease is dependent on the production of IFN-γ. They can also contribute to pathogen clearance by activation of macrophages, B cells and recruitment of neutrophils [52,53]. Additionally, in the response to helminth antigens, NKT cells were critically important for the generation of a Th2 response [54]. It has long been speculated that the NKT TCR may be directly involved in the recognition of pathogen derived antigens. Indeed, NKT cells can become activated upon infection with LPS containing bacteria, although this is dependent on DC activation by LPS and subsequent presentation of iGb3 and production of IL-12 [55]. In contrast, NKT cells might be stimulated by direct recognition of glycosylceramides derived from the cell wall of LPS negative bacteria, presented by DC [55,56]. This indicates that NKT cells might be important initiators of the immune response in some bacterial infections, whereas in others they may represent only one of several secondary responding cell types in others [57].

Another important role for NKT cells is found in anti-tumor immunity. Studies using chemical mutagenesis suggest that NKT cells contribute to natural tumor immunosurveillance [58]. Anti-tumor effects of α-GalCer have been observed against various tumors of different origins and their metastases [59,60]. The effect was dependent on NKT cell induced IL-12 production of DC that, together with NKT cell derived IFN-γ, secondarily stimulates NK and $CD8^+$ cells which function as direct anti-tumor effectors [59]. Improved efficiency of tumor rejection and prolonged IFN-γ responses were observed when α-GalCer pulsed DC were used instead of α-GalCer administration alone [61]. Paradoxically, NKT cells have been reported to also suppress tumor-specific $CD8^+$ cytotoxic T cell responses, resulting in tumor recurrence [62].

Thus, NKT cells can either promote or inhibit the development of protective anti-tumor responses.

The regulatory activity of NKT cells also seems to be implicated in the induction or maintenance of immune tolerance. The involvement of NKT cells was demonstrated in a model of immune privilege in the eye, known as anterior chamber-associated immune deviation [63]. Furthermore, NKT cells were required for the induction of allograft tolerance and survival in mouse models of transplant graft acceptance [64,65,66].

In addition, NKT cells help to maintain tolerance to self-antigens and can thereby prevent autoimmunity [67].

Relevance of NKT cells in autoimmune diseases

Many autoimmune diseases are characterized by Th1 polarized T cell responses and therefore, a role for NKT cells in the regulation of autoimmune diseases has been proposed. Additionally, reduced NKT cell numbers have been reported in autoimmune prone mice as well as in patients suffering from autoimmune diseases. Furthermore, estrogen promotes IFN-γ production by NKT cells, which suggests a possible contribution to the sexual dimorphism found in autoimmune diseases [68].

Type 1 Diabetes

Studies of type 1 diabetes have provided the most evidence that NKT cells are involved in autoimmune regulation. A defect in the number and function of NKT cells in NOD mice has been reported. This type of diabetes could be ameliorated by enrichment of NKT cell numbers through adoptive transfer or transgenic overexpression of the invariant TCR [69,70]. The genetic control of NKT cell numbers in NOD mice was

mapped to type 1 diabetes loci [71]. CD1d deficient mice developed diabetes earlier, had a greater disease penetrance and more severe disease [72,73]. In concordance, upregulation of CD1d expression restored the immunoregulatory function of NKT cells and prevented autoimmune diabetes [74]. Protection conferred by NKT cells was associated with a Th2 shift within the pancreatic islets, and IL-4 has been implicated as a key mediator of immunoregulation [69,75]. Stimulation of naïve T cells expressing a transgenic, diabetogenic TCR with their auto-antigen in the presence of NKT cells did not block the initial activation of the pathogenic T cells. However, both the production of IL-2 and IFN-γ and later proliferation were inhibited [76]. This suppression required cell contact and diabetes was still prevented in absence of cytokines involved in other functions of NKT cells [77]. These findings suggest that NKT cells may avert and ameliorate type I diabetes by preventing the differentiation of autoreactive T cells into effector cells.

In addition to a role in the natural course of type 1 diabetes, activation of NKT cells by administration of α-GalCer also prevented the onset and recurrence of diabetes in NOD mice [78,79,80]. Protection was associated with the induction and recruitment of tolerogenic DC [80]. Consequently, pathogenic T cells entering the pancreatic lymph nodes underwent great levels of apoptosis and the survivors became functionally anergic [81]. Protection also often coincided with suppression of pathogenic autoreactive T and B cells, and in the generation of tolerogenic islet autoantigen specific T cells with a protective cytokine production profile [78,79]. Furthermore, treatment of mice with OCH prevented the development of diabetes and insulitis in NOD mice more profoundly than α-GalCer [82].

Conflicting results have been found regarding the numerical reduction and dysfunction of NKT cells in human type 1 diabetes [83,84,85]. However, this might be attributed to different methods to detect NKT cells and to differences in the patient populations tested. In addition, it was reported that the NKT cell levels in blood of NOD mice are a poor representation of those in other organs where the deficiencies were observed, re-establishing the correlation between NKT cell deficiency and type 1 diabetes [86]. Furthermore, gene expression profiles of regulatory NKT cells from identical twins discordant for type I diabetes showed multiple differences [87].

Experimental autoimmune encephalomyelitis and multiple sclerosis

Murine NKT cells correlate with the pathogenesis of experimental autoimmune encephalomyelitis (EAE). SJL mice tend to develop chronic EAE and were shown to have NKT cells that are reduced in number and to have a defective IL-4 production [88]. Transgenic overexpression of the invariant TCR in NOD mice protected from EAE. This was associated with a striking inhibition of antigen-specific IFN-γ production, but was independent of IL-4 [89]. Additionally, studies using α-GalCer induced NKT cell stimulation confirmed the capacity of NKT cells to modulate the disease by inducing Th2 cytokine profiles [90,91,92]. However, others reported that the protective effect was associated with an enhanced IFN-γ production by liver-confined NKT cells [92]. Furthermore, the efficacy of α-GalCer treatment depended on the administration route, timing and dose [93,92]. In an effort to overcome these problems, combination therapy of α-GalCer and CD86 blocking antibodies or treatment with OCH was shown to selectively induce a Th2 response by the NKT cells and resulted in a reduced development of EAE [91,94,40].

Multiple sclerosis (MS) is a Th1 mediated autoimmune diseases which is directed to myelin antigens in the central nervous system [95]. A decrease in Vα24 mRNA was demonstrated in peripheral blood of MS patients [96] that seemed to coincide with the relapse state of the disease [97]. Additionally, the IL-4 secretion of DN NKT cells was reduced [98]. However, $CD4^+$ NKT cell lines obtained from patients in remission, showed a strong Th2 bias compared to patients in relapse [99]. These results support an immunoregulatory function of NKT cells in MS.

Rheumatoid arthritis and its animal models

NKT cells are now reported to be reduced in a whole range of diseases that are characterized by autoreactive tissue damage, including rheumatoid arthritis (RA) [100]. In addition, the invariant TCR transcripts were decreased in the rheumatoid synovium [101]. It was reported that NKT cells in RA patients showed a Th1 bias in blood and did not expand upon stimulation with α-GalCer in some patients [102,103]. Remarkably, NKT cells in synovial fluid of all patients responded to stimulation and displayed a Th0 cytokine profile [102]. Furthermore, the expression levels of soluble CD1d were lower in RA patients [104]. Recently, it was reported that collagen induced arthritis could be suppressed by activation of NKT cells with OCH, but not with α-GalCer [105]. Neutralization of IL-4 or IL-10 with monoclonal antibodies abolished disease protection by OCH, which indicates a critical role for these cytokines in disease protection. However, NKT cells might also be involved in the end-stage effector phase of arthritis, as these cells promoted antibody-induced joint inflammation by suppressing TGF-β [106].

Other autoimmune diseases

Lupus-prone mice as well as patients suffering from systemic lupus erythematosus (SLE) have reduced numbers of NKT cells, which suggests a protective effect of these cells on lupus development [107,100]. However, a possible pathogenic role of NKT cells has been supported by experiments showing that administration of α-GalCer exacerbated disease [108]. Transfer of activated NKT cells induced an autoimmune-like inflammation in young lupus-prone mice [109] and treatment with an anti-CD1d blocking antibody resulted in disease amelioration [108]. As for the EAE model, NKT cell activation with alpha-GalCer could suppress or promote pristane-induced lupus-like autoimmunity in mice, in a strain-dependent manner [110]. Finally, NKT cells have also been implicated in the regulation of a murine model of colitis [111] as well as in inflammatory bowel disease [100] and Wegener's granulomatosis [112].

Therapeutic application of α-GalCer based therapies for autoimmune diseases?

The striking conservation of the NKT/CD1d system among mammalian species offers a great opportunity for the use of α-GalCer as therapeutic agent in human autoimmune diseases [2]. However, clinical application of α-GalCer should be employed with caution since there are several concerns that may limit translation of the preclinical studies. It is still not clear how α-GalCer-mediated NKT cell stimulation can suppress Th1 mediated autoimmune diseases and also prevent tumor metastases and infections [113]. Moreover, in many of the studies of autoimmune diseases in mice, timing and dosage of α-GalCer had a significant impact on the disease outcome [93]. This illustrates the potential risk of augmenting unwanted Th1 responses that can worsen the

disease when administering α-GalCer. Additionally, although it is generally accepted that NKT cells might play a protective role in autoimmunity, some studies show that they may also contribute to autoimmune pathogenesis. In some animal models of autoimmune hepatitis, systemic lupus erythrematosis and autoimmune arthritis, it was shown that NKT cells may play an essential role in the pathogenesis of these diseases [106,114,108]. Stimulation of these pathogenic NKT cells would again lead to exacerbation of the disease. Furthermore, α-GalCer also exacerbates allergic airway inflammation in mice, which raises the possibility that long-term treatment may promote allergic reactions [115]. Finally, high dose administration of α-GalCer in mice has been reported to result in fatal liver damage [116]. However, it is likely that the adverse events in humans will be less pronounced since the number of NKT cells in humans is lower compared to mice. These findings clearly emphasize the need for detailed studies on the mechanisms involved in NKT cell mediated promotion of Th1 and Th2 responses and on the effect of α-GalCer administration in humans.

In spite these concerns, preliminary human trials with α-GalCer have already been performed. In a phase I clinical trial using α-GalCer as a treatment for human cancers, no adverse events were reported, even at high dosages [117]. Although these studies show promising results on the therapeutic use of α-GalCer in human cancer patients, more research will be necessary to fully comprehend the consequences of an α-GalCer mediated NKT cell activation in autoimmune diseases. More concerns on the effect of administration of α-GalCer have raised since the discovery that stimulation of NKT cells by α-GalCer causes the cells to become unresponsive, at least in mice [118]. Possibly, some of the potential problems associated with α-GalCer based therapy can be overcome by the use of analogues with superior activity in inducing Th2 responses

(OCH) or by combination therapies with cytokines (IL-7) or antibodies (anti-CD86 antibodies) that suppress Th1 and/or enhance Th2 cytokine production by NKT cells [105,91]. These approaches have already shown significant effect in animal models of autoimmune diseases. Furthermore, the identification of iGb3 might lead to a better understanding of the function of NKT cells in autoimmune diseases and as such give rise to the development of NKT cell based therapies for these diseases.

Conclusions

Much progress has recently been made in the understanding of the NKT cell biology. The importance of the immunoregulatory function of NKT cells was often demonstrated in animal models of several human diseases. Furthermore, the decreased frequency and the altered properties observed in patients suffering from autoimmune diseases implicate a (major) role of these cells in the regulation of autoimmune responses. Although the underlying mechanism of NKT cell dysfunction is not clear, it is likely that defects in their thymical development or in antigen synthesis may form the base of the NKT cell deficiency observed in many autoimmune diseases. Therapeutic approaches using α-GalCer or its analogues provide promising tools to treat autoimmune diseases. However, careful analysis of the *in vivo* consequences of α-GalCer administration or NKT cell activation should be performed to exclude unwanted Th1 responses that may worsen disease rather than suppress it. Hence, a more detailed insight in the molecular and cellular mechanisms involved in NKT cell function should help the development of new therapeutic strategies for autoimmune diseases.

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References

Reference List

1. Godfrey DI, MacDonald HR, Kronenberg M, Smyth MJ, Van Kaer L: NKT cells: what's in a name? Nat Rev Immunol 4:231, 2004.

2. Godfrey DI, Hammond KJ, Poulton LD, Smyth MJ, Baxter AG: NKT cells: facts, functions and fallacies. Immunol Today 21:573, 2000.

3. Joyce S: CD1d and natural T cells: how their properties jump-start the immune system. Cell Mol Life Sci 58:442, 2001.

4. Taniguchi M, Harada M, Kojo S, Nakayama T, Wakao H: The regulatory role of V(alpha) 14 NKT cells in innate and acquired immune response. Annu Rev Immunol 21:483, 2003.

5. Brigl M, Brenner MB: CD1: Antigen Presentation and T Cell Function. Annu Rev Immunol 22:817, 2004.

6. Seino K, Taniguchi M: Functional roles of NKT cell in the immune system. Front Biosci 9:2577, 2004.

7. Prussin C, Foster B: TCR V alpha 24 and V beta 11 coexpression defines a human NK1 T cell analog containing a unique Th0 subpopulation. J Immunol 159:5862, 1997.

8. Benlagha K, Weiss A, Beavis A, Teyton L, Bendelac A: In vivo identification of glycolipid antigen-specific T cells using fluorescent CD1d tetramers. J Exp Med 191:1895, 2000.

9. Norris S, Doherty DG, Collins C, McEntee G, Traynor O, Hegarty JE, O'Farrelly C: Natural T cells in the human liver: cytotoxic lymphocytes with

dual T cell and natural killer cell phenotype and function are phenotypically heterogenous and include Valpha24-JalphaQ and gammadelta T cell receptor bearing cells. Hum Immunol 60:20, 1999.

10. Der Vliet HJ, Nishi N, de Gruijl TD, von Blomberg BM, van den Eertwegh AJ, Pinedo HM, Giaccone G, Scheper RJ: Human natural killer T cells acquire a memory-activated phenotype before birth. Blood 95:2440, 2000.

11. Park SH, Benlagha K, Lee D, Balish E, Bendelac A: Unaltered phenotype, tissue distribution and function of Valpha14(+) NKT cells in germ-free mice. Eur J Immunol 30:620, 2000.

12. Godfrey DI, Pellicci DG, Smyth MJ: Immunology. The elusive NKT cell antigen--is the search over? Science 306:1687, 2004.

13. Gumperz JE, Miyake S, Yamamura T, Brenner MB: Functionally distinct subsets of CD1d-restricted natural killer T cells revealed by CD1d tetramer staining. J Exp Med 195:625, 2002.

14. Takahashi T, Chiba S, Nieda M, Azuma T, Ishihara S, Shibata Y, Juji T, Hirai H: Cutting edge: analysis of human V alpha 24+CD8+ NK T cells activated by alpha-galactosylceramide-pulsed monocyte-derived dendritic cells. J Immunol 168:3140, 2002.

15. Lee PT, Benlagha K, Teyton L, Bendelac A: Distinct functional lineages of human V(alpha)24 natural killer T cells. J Exp Med 195:637, 2002.

16. Kim CH, Johnston B, Butcher EC: Trafficking machinery of NKT cells: shared and differential chemokine receptor expression among Valpha24(+)Vbeta11(+) NKT cell subsets with distinct cytokine-producing capacity. Blood 100:11, 2002.

17. Bollyky PL, Wilson SB: CD1d-restricted T-cell subsets and dendritic cell function in autoimmunity. Immunol Cell Biol 82:307, 2004.

18. Kameyama H, Kawamura T, Naito T, Bannai M, Shimamura K, Hatakeyama K, Abo T: Size of the population of CD4+ natural killer T cells in the liver is maintained without supply by the thymus during adult life. Immunology 104:135, 2001.

19. MacDonald HR: Development and selection of NKT cells. Curr Opin Immunol 14:250, 2002.

20. Honey K, Benlagha K, Beers C, Forbush K, Teyton L, Kleijmeer MJ, Rudensky AY, Bendelac A: Thymocyte expression of cathepsin L is essential for NKT cell development. Nat Immunol 3:1069, 2002.

21. Elewaut D, Lawton AP, Nagarajan NA, Maverakis E, Khurana A, Honing S, Benedict CA, Sercarz E, Bakke O, Kronenberg M, Prigozy TI: The adaptor protein AP-3 is required for CD1d-mediated antigen presentation of glycosphingolipids and development of Valpha14i NKT cells. J Exp Med 198:1133, 2003.

22. Zhou D, Mattner J, Cantu C, III, Schrantz N, Yin N, Gao Y, Sagiv Y, Hudspeth K, Wu YP, Yamashita T, Teneberg S, Wang D, Proia RL, Levery SB, Savage PB, Teyton L, Bendelac A: Lysosomal glycosphingolipid recognition by NKT cells. Science 306:1786, 2004.

23. Bendelac A, Rivera MN, Park SH, Roark JH: Mouse CD1-specific NK1 T cells: development, specificity, and function. Annu Rev Immunol 15:535, 1997.

24. Gapin L, Matsuda JL, Surh CD, Kronenberg M: NKT cells derive from double-positive thymocytes that are positively selected by CD1d. Nat Immunol 2:971, 2001.

25. Pasquier B, Yin L, Fondaneche MC, Relouzat F, Bloch-Queyrat C, Lambert N, Fischer A, Saint-Basile G, Latour S: Defective NKT cell development in mice and humans lacking the adapter SAP, the X-linked lymphoproliferative syndrome gene product. J Exp Med 201:695, 2005.

26. Borowski C, Bendelac A: Signaling for NKT cell development: the SAP-FynT connection. J Exp Med 201:833, 2005.

27. Chun T, Page MJ, Gapin L, Matsuda JL, Xu H, Nguyen H, Kang HS, Stanic AK, Joyce S, Koltun WA, Chorney MJ, Kronenberg M, Wang CR: CD1d-expressing dendritic cells but not thymic epithelial cells can mediate negative selection of NKT cells. J Exp Med 197:907, 2003.

28. Pellicci DG, Uldrich AP, Kyparissoudis K, Crowe NY, Brooks AG, Hammond KJ, Sidobre S, Kronenberg M, Smyth MJ, Godfrey DI: Intrathymic NKT cell development is blocked by the presence of alpha-galactosylceramide. Eur J Immunol 33:1816, 2003.

29. Berzins SP, Uldrich AP, Pellicci DG, McNab F, Hayakawa Y, Smyth MJ, Godfrey DI: Parallels and distinctions between T and NKT cell development in the thymus. Immunol Cell Biol 82:269, 2004.

30. Baev DV, Peng XH, Song L, Barnhart JR, Crooks GM, Weinberg KI, Metelitsa LS: Distinct Homeostatic Requirements of CD4+ and CD4- Subsets of V{alpha}24-invariant Natural Killer T cells in Humans. Blood 2004.

31. Brossay L, Chioda M, Burdin N, Koezuka Y, Casorati G, Dellabona P, Kronenberg M: CD1d-mediated recognition of an alpha-galactosylceramide by natural killer T cells is highly conserved through mammalian evolution. J Exp Med 188:1521, 1998.

32. Exley M, Garcia J, Wilson SB, Spada F, Gerdes D, Tahir SM, Patton KT, Blumberg RS, Porcelli S, Chott A, Balk SP: CD1d structure and regulation on human thymocytes, peripheral blood T cells, B cells and monocytes. Immunology 100:37, 2000.

33. Kang SJ, Cresswell P: Calnexin, calreticulin, and ERp57 cooperate in disulfide bond formation in human CD1d heavy chain. J Biol Chem 277:44838, 2002.

34. Jayawardena-Wolf J, Benlagha K, Chiu YH, Mehr R, Bendelac A: CD1d endosomal trafficking is independently regulated by an intrinsic CD1d-encoded tyrosine motif and by the invariant chain. Immunity 15:897, 2001.

35. Spada FM, Koezuka Y, Porcelli SA: CD1d-restricted recognition of synthetic glycolipid antigens by human natural killer T cells. J Exp Med 188:1529, 1998.

36. Chiu YH, Park SH, Benlagha K, Forestier C, Jayawardena-Wolf J, Savage PB, Teyton L, Bendelac A: Multiple defects in antigen presentation and T cell development by mice expressing cytoplasmic tail-truncated CD1d. Nat Immunol 3:55, 2002.

37. Zeng Z, Castano AR, Segelke BW, Stura EA, Peterson PA, Wilson IA: Crystal structure of mouse CD1: An MHC-like fold with a large hydrophobic binding groove. Science 277:339, 1997.

38. Kawano T, Cui J, Koezuka Y, Toura I, Kaneko Y, Motoki K, Ueno H, Nakagawa R, Sato H, Kondo E, Koseki H, Taniguchi M: CD1d-restricted and TCR-mediated activation of valpha14 NKT cells by glycosylceramides. Science 278:1626, 1997.

39. Morita M, Motoki K, Akimoto K, Natori T, Sakai T, Sawa E, Yamaji K, Koezuka Y, Kobayashi E, Fukushima H: Structure-activity relationship of alpha-galactosylceramides against B16-bearing mice. J Med Chem 38:2176, 1995.

40. Miyamoto K, Miyake S, Yamamura T: A synthetic glycolipid prevents autoimmune encephalomyelitis by inducing TH2 bias of natural killer T cells. Nature 413:531, 2001.

41. Joyce S, Woods AS, Yewdell JW, Bennink JR, De Silva AD, Boesteanu A, Balk SP, Cotter RJ, Brutkiewicz RR: Natural ligand of mouse CD1d1: cellular glycosylphosphatidylinositol. Science 279:1541, 1998.

42. De Silva AD, Park JJ, Matsuki N, Stanic AK, Brutkiewicz RR, Medof ME, Joyce S: Lipid protein interactions: the assembly of CD1d1 with cellular phospholipids occurs in the endoplasmic reticulum. J Immunol 168:723, 2002.

43. Wu DY, Segal NH, Sidobre S, Kronenberg M, Chapman PB: Cross-presentation of disialoganglioside GD3 to natural killer T cells. J Exp Med 198:173, 2003.

44. Stetson DB, Mohrs M, Reinhardt RL, Baron JL, Wang ZE, Gapin L, Kronenberg M, Locksley RM: Constitutive cytokine mRNAs mark natural killer (NK) and NK T cells poised for rapid effector function. J Exp Med 198:1069, 2003.

45. Kitamura H, Iwakabe K, Yahata T, Nishimura S, Ohta A, Ohmi Y, Sato M, Takeda K, Okumura K, Van Kaer L, Kawano T, Taniguchi M, Nishimura T: The natural killer T (NKT) cell ligand alpha-galactosylceramide demonstrates its immunopotentiating effect by inducing interleukin (IL)-12 production by dendritic cells and IL-12 receptor expression on NKT cells. J Exp Med 189:1121, 1999.

46. Kawano T, Nakayama T, Kamada N, Kaneko Y, Harada M, Ogura N, Akutsu Y, Motohashi S, Iizasa T, Endo H, Fujisawa T, Shinkai H, Taniguchi M: Antitumor cytotoxicity mediated by ligand-activated human V alpha24 NKT cells. Cancer Res 59:5102, 1999.

47. Wilson MT, Johansson C, Olivares-Villagomez D, Singh AK, Stanic AK, Wang CR, Joyce S, Wick MJ, Van Kaer L: The response of natural killer T cells to glycolipid antigens is characterized by surface receptor down-modulation and expansion. Proc Natl Acad Sci U S A 100:10913, 2003.

48. Crowe NY, Uldrich AP, Kyparissoudis K, Hammond KJ, Hayakawa Y, Sidobre S, Keating R, Kronenberg M, Smyth MJ, Godfrey DI: Glycolipid antigen drives rapid expansion and sustained cytokine production by NK T cells. J Immunol 171:4020, 2003.

49. Nieda M, Okai M, Tazbirkova A, Lin H, Yamaura A, Ide K, Abraham R, Juji T, Macfarlane DJ, Nicol AJ: Therapeutic activation of Valpha24+Vbeta11+ NKT cells in human subjects results in highly coordinated secondary activation of acquired and innate immunity. Blood 103:383, 2004.

50. Kronenberg M: Toward an understanding of NKT cell biology: progress and paradoxes. Annu Rev Immunol 23:877, 2005.

51. Hansen DS, Schofield L: Regulation of immunity and pathogenesis in infectious diseases by CD1d-restricted NKT cells. Int J Parasitol 34:15, 2004.

52. Nieuwenhuis EE, Matsumoto T, Exley M, Schleipman RA, Glickman J, Bailey DT, Corazza N, Colgan SP, Onderdonk AB, Blumberg RS: CD1d-dependent macrophage-mediated clearance of Pseudomonas aeruginosa from lung. Nat Med 8:588, 2002.

53. Kumar H, Belperron A, Barthold SW, Bockenstedt LK: Cutting edge: CD1d deficiency impairs murine host defense against the spirochete, Borrelia burgdorferi. J Immunol 165:4797, 2000.

54. Faveeuw C, Angeli V, Fontaine J, Maliszewski C, Capron A, Van Kaer L, Moser M, Capron M, Trottein F: Antigen presentation by CD1d contributes to the amplification of Th2 responses to Schistosoma mansoni glycoconjugates in mice. J Immunol 169:906, 2002.

55. Mattner J, Debord KL, Ismail N, Goff RD, Cantu C, III, Zhou D, Saint-Mezard P, Wang V, Gao Y, Yin N, Hoebe K, Schneewind O, Walker D, Beutler B, Teyton L, Savage PB, Bendelac A: Exogenous and endogenous glycolipid antigens activate NKT cells during microbial infections. Nature 434:525, 2005.

56. Kinjo Y, Wu D, Kim G, Xing GW, Poles MA, Ho DD, Tsuji M, Kawahara K, Wong CH, Kronenberg M: Recognition of bacterial glycosphingolipids by natural killer T cells. Nature 434:520, 2005.

57. Van Kaer L, Joyce S: Innate immunity: NKT cells in the spotlight. Curr Biol 15:R429, 2005.

58. Smyth MJ, Thia KY, Street SE, Cretney E, Trapani JA, Taniguchi M, Kawano T, Pelikan SB, Crowe NY, Godfrey DI: Differential tumor surveillance by natural killer (NK) and NKT cells. J Exp Med 191:661, 2000.

59. Smyth MJ, Cretney E, Takeda K, Wiltrout RH, Sedger LM, Kayagaki N, Yagita H, Okumura K: Tumor necrosis factor-related apoptosis-inducing ligand (TRAIL) contributes to interferon gamma-dependent natural killer cell protection from tumor metastasis. J Exp Med 193:661, 2001.

60. Kobayashi E, Motoki K, Uchida T, Fukushima H, Koezuka Y: KRN7000, a novel immunomodulator, and its antitumor activities. Oncol Res 7:529, 1995.

61. Toura I, Kawano T, Akutsu Y, Nakayama T, Ochiai T, Taniguchi M: Cutting edge: inhibition of experimental tumor metastasis by dendritic cells pulsed with alpha-galactosylceramide. J Immunol 163:2387, 1999.

62. Terabe M, Matsui S, Noben-Trauth N, Chen H, Watson C, Donaldson DD, Carbone DP, Paul WE, Berzofsky JA: NKT cell-mediated repression of tumor immunosurveillance by IL-13 and the IL-4R-STAT6 pathway. Nat Immunol 1:515, 2000.

63. Hong S, Van Kaer L: Immune privilege: keeping an eye on natural killer T cells. J Exp Med 190:1197, 1999.

64. Seino KI, Fukao K, Muramoto K, Yanagisawa K, Takada Y, Kakuta S, Iwakura Y, Van Kaer L, Takeda K, Nakayama T, Taniguchi M, Bashuda H, Yagita H, Okumura K: Requirement for natural killer T (NKT) cells in the induction of allograft tolerance. Proc Natl Acad Sci U S A 98:2577, 2001.

65. Ikehara Y, Yasunami Y, Kodama S, Maki T, Nakano M, Nakayama T, Taniguchi M, Ikeda S: CD4(+) Valpha14 natural killer T cells are essential for acceptance of rat islet xenografts in mice. J Clin Invest 105:1761, 2000.

66. Sonoda KH, Taniguchi M, Stein-Streilein J: Long-term survival of corneal allografts is dependent on intact CD1d-reactive NKT cells. J Immunol 168:2028, 2002.

67. Godfrey DI, Kronenberg M: Going both ways: immune regulation via CD1d-dependent NKT cells. J Clin Invest 114:1379, 2004.

68. Gourdy P, Araujo LM, Zhu R, Garmy-Susini B, Diem S, Laurell H, Leite-De-Moraes M, Dy M, Arnal JF, Bayard F, Herbelin A: Relevance of sexual dimorphism to regulatory T cells: estradiol promotes IFN-{gamma} production by invariant natural killer T cells. Blood 2004.

69. Hammond KJ, Poulton LD, Palmisano LJ, Silveira PA, Godfrey DI, Baxter AG: alpha/beta-T cell receptor (TCR)+CD4-CD8- (NKT) thymocytes prevent insulin-dependent diabetes mellitus in nonobese diabetic (NOD)/Lt mice by the influence of interleukin (IL)-4 and/or IL-10. J Exp Med 187:1047, 1998.

70. Lehuen A, Lantz O, Beaudoin L, Laloux V, Carnaud C, Bendelac A, Bach JF, Monteiro RC: Overexpression of natural killer T cells protects Valpha14-Jalpha281 transgenic nonobese diabetic mice against diabetes. J Exp Med 188:1831, 1998.

71. Esteban LM, Tsoutsman T, Jordan MA, Roach D, Poulton LD, Brooks A, Naidenko OV, Sidobre S, Godfrey DI, Baxter AG: Genetic control of NKT cell numbers maps to major diabetes and lupus loci. J Immunol 171:2873, 2003.

72. Wang B, Geng YB, Wang CR: CD1-restricted NK T cells protect nonobese diabetic mice from developing diabetes. J Exp Med 194:313, 2001.

73. Shi FD, Flodstrom M, Balasa B, Kim SH, Van Gunst K, Strominger JL, Wilson SB, Sarvetnick N: Germ line deletion of the CD1 locus exacerbates diabetes in the NOD mouse. Proc Natl Acad Sci U S A 98:6777, 2001.

74. Falcone M, Facciotti F, Ghidoli N, Monti P, Olivieri S, Zaccagnino L, Bonifacio E, Casorati G, Sanvito F, Sarvetnick N: Up-regulation of CD1d expression restores the immunoregulatory function of NKT cells and prevents autoimmune diabetes in nonobese diabetic mice. J Immunol 172:5908, 2004.

75. Laloux V, Beaudoin L, Jeske D, Carnaud C, Lehuen A: NK T cell-induced protection against diabetes in V alpha 14-J alpha 281 transgenic nonobese diabetic mice is associated with a Th2 shift circumscribed regionally to the islets and functionally to islet autoantigen. J Immunol 166:3749, 2001.

76. Beaudoin L, Laloux V, Novak J, Lucas B, Lehuen A: NKT cells inhibit the onset of diabetes by impairing the development of pathogenic T cells specific for pancreatic beta cells. Immunity 17:725, 2002.

77. Novak J, Beaudoin L, Griseri T, Lehuen A: Inhibition of T cell differentiation into effectors by NKT cells requires cell contacts. J Immunol 174:1954, 2005.

78. Sharif S, Arreaza GA, Zucker P, Mi QS, Sondhi J, Naidenko OV, Kronenberg M, Koezuka Y, Delovitch TL, Gombert JM, Leite-De-Moraes M, Gouarin C, Zhu R, Hameg A, Nakayama T, Taniguchi M, Lepault F, Lehuen A, Bach JF, Herbelin A: Activation of natural killer T cells by alpha-galactosylceramide treatment prevents the onset and recurrence of autoimmune Type 1 diabetes. Nat Med 7:1057, 2001.

79. Hong S, Wilson MT, Serizawa I, Wu L, Singh N, Naidenko OV, Miura T, Haba T, Scherer DC, Wei J, Kronenberg M, Koezuka Y, Van Kaer L: The natural killer T-cell ligand alpha-galactosylceramide prevents autoimmune diabetes in non-obese diabetic mice. Nat Med 7:1052, 2001.

80. Naumov YN, Bahjat KS, Gausling R, Abraham R, Exley MA, Koezuka Y, Balk SB, Strominger JL, Clare-Salzer M, Wilson SB: Activation of CD1d-restricted T cells protects NOD mice from developing diabetes by regulating dendritic cell subsets. Proc Natl Acad Sci U S A 98:13838, 2001.

81. Chen YG, Choisy-Rossi CM, Holl TM, Chapman HD, Besra GS, Porcelli SA, Shaffer DJ, Roopenian D, Wilson SB, Serreze DV: Activated NKT cells inhibit autoimmune diabetes through tolerogenic recruitment of dendritic cells to pancreatic lymph nodes. J Immunol 174:1196, 2005.

82. Mizuno M, Masumura M, Tomi C, Chiba A, Oki S, Yamamura T, Miyake S: Synthetic glycolipid OCH prevents insulitis and diabetes in NOD mice. J Autoimmun 23:293, 2004.

83. Wilson SB, Kent SC, Patton KT, Orban T, Jackson RA, Exley M, Porcelli S, Schatz DA, Atkinson MA, Balk SP, Strominger JL, Hafler DA: Extreme Th1 bias of invariant Valpha24JalphaQ T cells in type 1 diabetes. Nature 391:177, 1998.

84. Lee PT, Putnam A, Benlagha K, Teyton L, Gottlieb PA, Bendelac A: Testing the NKT cell hypothesis of human IDDM pathogenesis. J Clin Invest 110:793, 2002.

85. Oikawa Y, Shimada A, Yamada S, Motohashi Y, Nakagawa Y, Irie J, Maruyama T, Saruta T: High frequency of valpha24(+) vbeta11(+) T-cells observed in type 1 diabetes. Diabetes Care 25:1818, 2002.

86. Berzins SP, Kyparissoudis K, Pellicci DG, Hammond KJ, Sidobre S, Baxter A, Smyth MJ, Kronenberg M, Godfrey DI: Systemic NKT cell deficiency in NOD

mice is not detected in peripheral blood: implications for human studies. Immunol Cell Biol 82:247, 2004.

87. Wilson SB, Kent SC, Horton HF, Hill AA, Bollyky PL, Hafler DA, Strominger JL, Byrne MC: Multiple differences in gene expression in regulatory Valpha 24Jalpha Q T cells from identical twins discordant for type I diabetes. Proc Natl Acad Sci U S A 97:7411, 2000.

88. Yoshimoto T, Bendelac A, Hu-Li J, Paul WE: Defective IgE production by SJL mice is linked to the absence of CD4+, NK1.1+ T cells that promptly produce interleukin 4. Proc Natl Acad Sci U S A 92:11931, 1995.

89. Mars LT, Laloux V, Goude K, Desbois S, Saoudi A, Van Kaer L, Lassmann H, Herbelin A, Lehuen A, Liblau RS: Cutting edge: V alpha 14-J alpha 281 NKT cells naturally regulate experimental autoimmune encephalomyelitis in nonobese diabetic mice. J Immunol 168:6007, 2002.

90. Singh AK, Wilson MT, Hong S, Olivares-Villagomez D, Du C, Stanic AK, Joyce S, Sriram S, Koezuka Y, Van Kaer L: Natural killer T cell activation protects mice against experimental autoimmune encephalomyelitis. J Exp Med 194:1801, 2001.

91. Pal E, Tabira T, Kawano T, Taniguchi M, Miyake S, Yamamura T: Costimulation-dependent modulation of experimental autoimmune encephalomyelitis by ligand stimulation of V alpha 14 NK T cells. J Immunol 166:662, 2001.

92. Furlan R, Bergami A, Cantarella D, Brambilla E, Taniguchi M, Dellabona P, Casorati G, Martino G: Activation of invariant NKT cells by alphaGalCer administration protects mice from MOG35-55-induced EAE: critical roles for administration route and IFN-gamma. Eur J Immunol 33:1830, 2003.

93. Jahng AW, Maricic I, Pedersen B, Burdin N, Naidenko O, Kronenberg M, Koezuka Y, Kumar V: Activation of natural killer T cells potentiates or prevents experimental autoimmune encephalomyelitis. J Exp Med 194:1789, 2001.

94. Hayakawa Y, Takeda K, Yagita H, Van Kaer L, Saiki I, Okumura K: Differential regulation of Th1 and Th2 functions of NKT cells by CD28 and CD40 costimulatory pathways. J Immunol 166:6012, 2001.

95. Hellings N, Raus J, Stinissen P: Insights into the immunopathogenesis of multiple sclerosis. Immunol Res 25:27, 2002.

96. Illes Z, Kondo T, Newcombe J, Oka N, Tabira T, Yamamura T: Differential expression of NK T cell V alpha 24J alpha Q invariant TCR chain in the lesions of multiple sclerosis and chronic inflammatory demyelinating polyneuropathy. J Immunol 164:4375, 2000.

97. Demoulins T, Gachelin G, Bequet D, Dormont D: A biased Valpha24+ T-cell repertoire leads to circulating NKT-cell defects in a multiple sclerosis patient at the onset of his disease. Immunol Lett 90:223, 2003.

98. Gausling R, Trollmo C, Hafler DA: Decreases in interleukin-4 secretion by invariant CD4(-)CD8(-)V alpha 24J alpha Q T cells in peripheral blood of patientswith relapsing- remitting multiple sclerosis. Clin Immunol 98:11, 2001.

99. Araki M, Kondo T, Gumperz JE, Brenner MB, Miyake S, Yamamura T: T(h)2 bias of CD4(+) NKT cells derived from multiple sclerosis in remission. Int Immunol 15:279, 2003.

100. van der Vliet HJ, von Blomberg BM, Nishi N, Reijm M, Voskuyl AE, van Bodegraven AA, Polman CH, Rustemeyer T, Lips P, van den Eertwegh AJ, Giaccone G, Scheper RJ, Pinedo HM: Circulating V(alpha24+) Vbeta11+ NKT cell numbers are decreased in a wide variety of diseases that are characterized by autoreactive tissue damage. Clin Immunol 100:144, 2001.

101. Maeda T, Keino H, Asahara H, Taniguchi M, Nishioka K, Sumida T: Decreased TCR AV24AJ18+ double-negative T cells in rheumatoid synovium. Rheumatology (Oxford) 38:186, 1999.

102. Linsen L, Thewissen M, Baeten K, Somers V, Geusens P, Raus J, Stinissen P: Peripheral blood, but not Synovial Fluid NKT cells are biased towards a Th1-like phenotype in Rheumatoid Arthritis. Arthritis Res Ther 7:R493, 2005.

103. Kojo S, Adachi Y, Keino H, Taniguchi M, Sumida T: Dysfunction of T cell receptor AV24AJ18+, BV11+ double-negative regulatory natural killer T cells in autoimmune diseases. Arthritis Rheum 44:1127, 2001.

104. Kojo S, Tsutsumi A, Goto D, Sumida T: Low expression levels of soluble CD1d gene in patients with rheumatoid arthritis. J Rheumatol 30:2524, 2003.

105. Chiba A, Oki S, Miyamoto K, Hashimoto H, Yamamura T, Miyake S: Suppression of collagen-induced arthritis by natural killer T cell activation with OCH, a sphingosine-truncated analog of alpha-galactosylceramide. Arthritis Rheum 50:305, 2004.

106. Kim HY, Kim HJ, Min HS, Kim S, Park WS, Park SH, Chung DH: NKT cells promote antibody-induced joint inflammation by suppressing transforming growth factor {beta}1 production. J Exp Med 201:41, 2005.

107. Mieza MA, Itoh T, Cui JQ, Makino Y, Kawano T, Tsuchida K, Koike T, Shirai T, Yagita H, Matsuzawa A, Koseki H, Taniguchi M: Selective reduction of V alpha 14+ NK T cells associated with disease development in autoimmune-prone mice. J Immunol 156:4035, 1996.

108. Zeng D, Liu Y, Sidobre S, Kronenberg M, Strober S: Activation of natural killer T cells in NZB/W mice induces Th1-type immune responses exacerbating lupus. J Clin Invest 112:1211, 2003.

109. Morshed SR, Mannoor K, Halder RC, Kawamura H, Bannai M, Sekikawa H, Watanabe H, Abo T: Tissue-specific expansion of NKT and CD5+B cells at the onset of autoimmune disease in (NZBxNZW)F1 mice. Eur J Immunol 32:2551, 2002.

110. Singh AK, Yang JQ, Parekh VV, Wei J, Wang CR, Joyce S, Singh RR, Van Kaer L: The natural killer T cell ligand alpha-galactosylceramide prevents or promotes pristane-induced lupus in mice. Eur J Immunol 35:1143, 2005.

111. Shibolet O, Kalish Y, Klein A, Alper R, Zolotarov L, Thalenfeld B, Engelhardt D, Rabbani E, Ilan Y: Adoptive transfer of ex vivo immune-programmed NKT lymphocytes alleviates immune-mediated colitis. J Leukoc Biol 75:76, 2004.

112. Takagi D, Iwabuchi K, Iwabuchi C, Nakamaru Y, Maguchi S, Ohwatari R, Furuta Y, Fukuda S, Joyce S, Onoe K: Immunoregulatory defects of V alpha 24V+ beta 11+ NKT cells in development of Wegener's granulomatosis and relapsing polychondritis. Clin Exp Immunol 136:591, 2004.

113. van der Vliet HJ, Molling JW, von Blomberg BM, Nishi N, Kolgen W, van den Eertwegh AJ, Pinedo HM, Giaccone G, Scheper RJ: The immunoregulatory role of CD1d-restricted natural killer T cells in disease. Clin Immunol 112:8, 2004.

114. Takeda K, Hayakawa Y, Van Kaer L, Matsuda H, Yagita H, Okumura K: Critical contribution of liver natural killer T cells to a murine model of hepatitis. Proc Natl Acad Sci U S A 97:5498, 2000.

115. Akbari O, Stock P, Meyer E, Kronenberg M, Sidobre S, Nakayama T, Taniguchi M, Grusby MJ, DeKruyff RH, Umetsu DT: Essential role of NKT cells

producing IL-4 and IL-13 in the development of allergen-induced airway hyperreactivity. Nat Med 9:582, 2003.

116. Osman Y, Kawamura T, Naito T, Takeda K, Van Kaer L, Okumura K, Abo T: Activation of hepatic NKT cells and subsequent liver injury following administration of alpha-galactosylceramide. Eur J Immunol 30:1919, 2000.

117. Giaccone G, Punt CJ, Ando Y, Ruijter R, Nishi N, Peters M, von Blomberg BM, Scheper RJ, van der Vliet HJ, van den Eertwegh AJ, Roelvink M, Beijnen J, Zwierzina H, Pinedo HM: A phase I study of the natural killer T-cell ligand alpha- galactosylceramide (KRN7000) in patients with solid tumors. Clin Cancer Res 8:3702, 2002.

118. Parekh VV, Wilson MT, Olivares-Villagomez D, Singh AK, Wu L, Wang CR, Joyce S, Van Kaer L: Glycolipid antigen induces long-term natural killer T cell anergy in mice. J Clin Invest 115:2572, 2005.