Paired immunoglobulin-like receptors and their MHC class I recognition

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Summary
The immunoglobulin-like receptors provide positive and negative regulation of immune cells upon recognition of various ligands, thus enabling those cells to respond properly to extrinsic stimuli. Murine paired immunoglobulin-like receptor (PIR)-A and PIR-B, a typical receptor pair of the immunoglobulin-like receptor family, are expressed on a wide range of cells in the immune system, such as B cells, mast cells, macrophages and dendritic cells, mostly in a pair-wise fashion. The PIR-A requires the homodimeric Fc receptor common γ chain for its efficient cell-surface expression and for the delivery of an activation signal. In contrast, PIR-B inhibits receptor-mediated activation signals in vitro upon engagement with other activating-type receptors, such as the antigen receptor on B cells and the high-affinity Fc receptor for immunoglobulin E on mast cells. Recent identification of major histocompatibility complex (MHC) class I molecules as the physiological ligands for PIR has enabled us to attribute various immunological phenotypes observed in PIR-B-deficient mice to the consequences of the absence of a balanced interaction between PIR and MHC class I molecules expressed ubiquitously. Thus, PIR-A and PIR-B constitute a novel and physiologically important MHC class I recognition system.

Keywords: graft-versus-host disease; Th2 response; hypersensitivity; transplantation; autoimmunity; paired immunoglobulin-like receptors

Introduction

Paired immunoglobulin-like receptors (PIR) were first identified as those homologous to human Fc receptor (FcR) for immunoglobulin A (IgA), FcζRI. In 1997, Hayami et al. reported the isolation of several cDNA clones coding for a novel molecule from a B10.A mouse macrophage cDNA library during the course of experiments to obtain a hypothetical murine counterpart of FcζRI. The gene product was initially designated as p91 because of the calculated molecular weight of the mature polypeptide backbone. A similar approach by Kubagawa et al. yielded a novel gene family, the members of which constitute a set of PIR-A and PIR-B genes in the BALB/c splenic library. Because p91 and PIR-B were the most plausibly identical molecules based on their 98% sequence identity, the nomenclature has been standardized as PIR to avoid confusion (Fig. 1).

These initial studies could not detect PIR binding to IgA nor other immunoglobulins, suggesting that they are not receptors for immunoglobulins. Instead, PIR-A and PIR-B are now proposed as orthologues of human leucocyte immunoglobulin-like receptors (LIILR, also termed immunoglobulin-like transcripts/leucocyte immunoglobulin-like receptors/monocyte/macrophage immunoglobulin-related receptors (ILT/LIR/MIR)), based on their similarities in structure, expression profiles and genomic localization. The analogy of PIR-B to the inhibitory isoform of LIILR, namely LIIRB, and the findings that constitutive phosphorylation of PIR-B in splenocytes was reduced in β2-microglobulin (β2M)-deficient (B2m−/−) mice led to the notion that PIR-B may recognize classical or non-classical major histocompatibility complex (MHC) class I molecule(s). The observation that human leucocyte antigen (HLA)-G, a non-classical MHC class Ib on fetal trophoblast cells, binds PIR-B supports this notion. Recently, Nakamura et al. have shown that, in fact, the extracellular portion of PIR can bind to murine MHC class I molecules both in vitro and in vivo. In the present review, the physiological significance of PIR will be discussed in relation to the MHC class I recognition.
**PIR genes**

The PIR (CHIR) and LILR gene families are orthologs. At least six PIR-A genes (Pira) and a single PIR-B gene (Pirb) have been identified in the mouse chromosome. Consider gene evolution of PIR, it is interesting to see PIR from other species. The LILR are human orthologues of PIR. Structures of rat PIR-A and PIR-B homologues (Fig. 1) were quite similar to those of respective mouse PIR, indicating that the structural hallmarks of the PIR gene family are conserved in rats and mice. Similarly, genes for chicken PIR homologues (termed CHIR) have been mapped, supporting the notion that the PIR and LILR gene families are orthologs. It is also proposed that PIR-B could bind human HLA-B27.

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**Expression and protein structure of PIR**

PIR-A and PIR-B are expressed on various haematopoietic cell lineages, including B cells, mast cells, macrophages, granulocytes and dendritic cells (DC), mostly in a pairwise fashion, but are not expressed on T and natural killer (NK) cells (Fig. 1). Amino acid sequences of PIR-A and PIR-B ectodomains are highly homologous (over 92% identity). The deduced structure of PIR-B is a type I transmembrane glycoprotein with six extracellular immunoglobulin-like domains, a hydrophobic transmembrane segment and an intracellular polypeptide with four ITIM or ITIM-like sequences (consensus: (I/L/V/S)xYxx(L/V); Fig. 1). The PIR-B is highly homologous to...
several human and mouse immunoglobulin-like receptors, including murine gp49B1 (31% homology at the amino acid level), human KIR (34%), human FcγRI (29%), bovine FcγRII (32%), and less homologous to human and mouse FcγRIIB (17%).

Similarly, PIR-A molecules have six immunoglobulin-like extracellular domains, but in contrast to PIR-B, they contain unique pretransmembrane, transmembrane and short cytoplasmic sequences harbouring no ITIM-like motifs (Fig. 1). In addition, their transmembrane domains contain a positively charged residue, arginine, which presumably is crucial for the association of the FcR common γ subunit (Fcγγ), which itself is critical for expression on the cell surface and for delivery of the activation signal.

Although comparison of the available sequences of PIR extracellular portions from 129/Sv, B10.A and BALB/c mice indicated a fairly high sequence similarity, multiple substitutions of amino acid residues were observed, especially in the first four extracellular domains. The polymorphic nature of PIR has been one of the important characteristics to rationalize the notion that PIR can bind polymorphic class I molecules, similar to the situation for LILR and KIR, which have many polymorphic substitutions in their extracellular domains of some which recognize polymorphic MHC class I molecules.

Monoclonal and polyclonal antibodies to PIR identified cell-surface glycoproteins of approximately 85 and 120,000 MW on B cells, granulocytes and macrophages.

Using a fibroblast transfection experiment as well as FcRγ-deficient (Fcγγ) mice, it was shown that approximately 120,000 MW PIR-B is normally expressed on the cell surface without any other subunits, whereas approximately 85,000 MW PIR-A requires association with a homodimeric Fcγγ for its cell surface expression.

Interestingly, cell surface levels of PIR molecules on myeloid and B lineage cells reportedly increased with cellular differentiation and activation. Surface PIR levels are highest on marginal zone B cells and the B1 B cells express higher PIR levels than the B2 B cells.

### Ligands for PIR

PIR-B tyrosine phosphorylation status was examined in either B2m−/−, transporters associated with antigen processing (TAP)1-deficient or MHC class II-deficient mice, and the level of PIR-B tyrosine phosphorylation was reduced by approximately 50% in B2m−/− mice, but is not significantly altered in TAP1- or MHC class II-deficient mice. Non-classical MHC class I or class I-like molecules were speculated to be more likely candidates for native PIR ligands than classical MHC class I molecules.

To obtain direct evidence for the PIR binding to MHC class I molecules, Nakamura et al. took advantage of surface plasmon resonance analysis of the possible interaction between recombinant PIR-B ectodomain and various murine MHC class I (H-2) molecules. They found that recombinant PIR-B ectodomain bound to the monomeric H-2 molecules, H-2Ld, H-2Dd, H-2Kb, H-2Kk and H-2Kd, at affinities of $K_d = 1 \times 10^{-7}$ m, the values comparable, for example, to those between IgG and low-affinity FcγRs, FcγRIIB and FcγRIII. It is well known that the low-affinity FcγRs can bind multimeric IgG or IgG immune complexes very efficiently in vivo. In fact, the recombinant PIR-B ectodomain bound tetrameric H-2 at higher affinities of $K_d = 10^{-8}$–$10^{-9}$ m, indicating the physiological significance of the low but substantial affinities of PIR to monomeric MHC class I molecules.

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### Activation signalling via PIR-A

Engagement of transfected PIR-A expressed on the rat mast cell line RBL-2H3 with antibodies elicited a calcium mobilization and degranulation response, indicating that PIR-A plays a role in the activation of mast cells. The molecule associating with the PIR-A transmembrane region was Fcγγ, and the FcγRIβ chain in RBL-2H3, in which two positively and negatively charged residues of PIR-A, Arg626 and Glu643, respectively, were pivotal in the activating function of PIR-A and its association with the FcR subunits. Events in the downstream signal of Fcγγ have been studied extensively in relation to FcγRI on mast cells and activating type FcRs, FcγRI and FcγRIII, on macrophages. However, the exact physiological nature of PIR-A/Fcγγ signalling has not been studied until recently because of a lack of information regarding the ligand for PIR-A.

H-2 molecules are widely expressed on various murine haematopoietic and non-haematopoietic cells. Verification of the tetrameric H-2 binding to cellular PIR-A and PIR-B
suggests that H-2 molecules expressed on neighbouring cells (trans) or on the same cell (cis) may induce the constitutive phosphorylation of FcRγ in PIR-A receptor complex or ITIMs of PIR-B cytoplasmic domain (Fig. 2). It was examined whether the binding of tetrameric H-2 molecules to PIR-A on macrophages or PIR-B on B cells could induce up-regulated tyrosine phosphorylation of the receptor system. The phosphotyrosylation of FcRγ was substantially up-regulated in peritoneal macrophages from wild-type mice upon tetrameric H-2 stimulation. Note, constitutive phosphotyrosylation of FcRγ occurred in peritoneal macrophages without any specific stimuli, suggesting the constitutive phosphorylation of PIR-A by surrounding or neighbouring, native H-2 molecules. The phosphorylation was further increased in PIR-B deficiency. These observations indicate that PIR-A can deliver activation signal intracellularly via tyrosine phosphorylation of FcRγ upon interaction of PIR-A with H-2 molecules, and the signal is continuously inhibited by PIR-B coexpressed on the same cells (Fig. 2).

Inhibitory signalling via PIR-B

Taking advantage of B cells that express PIR-B but not PIR-A, the signalling nature via PIR-B upon interaction with H-2 has been investigated. Various H-2 tetramers were added to splenic B cells, and then phosphotyrosylation of PIR-B was monitored by immunoprecipitation and immunoblot analysis. The enhanced phosphotyrosylation of PIR-B after stimulation with every H-2 tetramer was observed, although the enhancement was rather small, about 1.8-fold higher than the non-stimulation control at most. Importantly, the constitutive PIR-B tyrosine phosphorylation in splenic B cells was observed without any specific stimuli. These notions are consistent with the hyper-activated state of B cells from PIR-B-deficient (Pirb−/−) mice in the absence of any specific stimulation of BCR in vitro and in vivo (Fig. 2).

Most of the inhibitory isoforms of immunoglobulin-like receptors exert their negative regulation of cells by recruiting SH2-containing tyrosine phosphatases SHP-1 and/or SHP-2 to their phosphorylated ITIM. Exceptionally, a unique inhibitory FcγR, namely FcγRIIB, recruits SH2-containing inositol 5-phosphatase (SHP-1) to the phosphorylated ITIM. PIR-B functions as the former type receptor. In vitro mutation analysis of cytoplasmic tyrosine residues in the ITIM of PIR-B indicated that the tyrosine in the third ITIM plays the most crucial role in mediating the inhibitory signal for B-cell receptor (BCR)-mediated cell activation as assessed by calcium mobilization and nuclear factor (NF)-AT activation. Also in vitro, synthetic phosphotyrosyl peptides corresponding to the third and fourth ITIMs of PIR-B can bind SHP-1, SHP-2 and SHIP-1 in cell extracts from macrophages, as well as in a B-cell line. However, the PIR-B-mediated inhibition was markedly reduced in SHP-1 and SHP-2 double-deficient DT40 chicken B cells, whereas this inhibition was unaffected in SHIP-deficient cells, suggesting that PIR-B can negatively regulate BCR activation by redundant functions of SHP-1 and SHP-2, but not by SHIP. In addition, in the mast cell line RBL-2H3, coaggregation of transfected PIR-B with FcεRI induced the PIR-B to recruit SHP-1 but not SHP-2 or SHIP. Even in the absence of the coaggregation, weak constitutive association of the PIR-B cytoplasmic domain with SHP-1 was observed. The third and fourth ITIM tyrosine residues were crucial for inhibition of RBL-2H3 cell degranulation and calcium mobilization. A similar approach with bone marrow-derived cultured mast cells reached to a conclusion that PIR-B expressed on bone marrow mast cells was constitutively phosphorylated and associated with SHP-1. Co-ligation of PIR-B and FcεRI inhibited IgE-mediated mast cell activation.

A reverse approach yielded an interesting observation about the preferential association of SHP-1 to PIR-B. The SHP-1 was shown to associate with a 130 000 MW tyrosyl-phosphorylated species, termed P130, in murine macrophages, suggesting that the P130 may be an SHP-1 regulator and/or substrate. Interestingly, P130 consisted of two transmembrane glycoproteins, namely PIR-B and the signal-regulatory protein (SIRP) family member BIT.
Furthermore, PIR-B was hyperphosphorylated in macrophages from SHP-1-inactive motheaten viable mice, whereas it was hypophosphorylated in SHP-1-deficient motheaten macrophages, suggesting a model in which SHP-1 dephosphorylates specific sites on PIR-B while protecting other sites from dephosphorylation via its SH2 domains. The PIR-B also associates with two tyrosyl phosphoproteins and a tyrosine kinase activity.\(^6\)

Irrespective of the cell activation status, PIR-B molecules in macrophages and B cells are constitutively phosphorylated possibly via continuous interaction with surrounding H-2 molecules, and PIR-B in splenocytes was constitutively associated with SHP-1 and Lyn.\(^7\) In Lyn-deficient mice, PIR-B tyrosine phosphorylation was greatly reduced.\(^7\) The cross-linking of PIR-B on chicken DT40 cells inhibits the BCR-induced tyrosine phosphorylation of immunoglobulin \(z\)/immunoglobulin \(\beta\), Syk, Btk and phospholipase \(C\gamma2\).\(^37\) The constitutive association of SHP-1 with PIR-B suggests that the inhibitory function of PIR-B may not necessarily require coligation of BCR and PIR-B by any unknown ligand shared by them. This speculation was shown to be the case in splenic B cells from Pirb\(^{−/−}\) mice, as described below.

**Dominant expression of PIR-B in vivo**

Because physiological ligand for PIR was identified as H-2 molecules, PIR may modulate inflammatory and immune responses by constitutive engagement with self H-2 molecules. Recently, Pirb\(^{−/−}\) mice have provided us with insight into the physiological significance of the H-2 recognition by PIR in the immune response, especially in antigen presentation, humoral immunity and transplantation as follows.\(^9,30\)

Because an available monoclonal antibody to PIR, 6C1, recognizes both PIR-A and PIR-B, it was not known which receptor is dominantly expressed on various cell surfaces. Given that the deletion of one receptor does not influence the expression of the other, PIR-B deficiency will tell us the surface expression of PIR-B by flow cytometry with 6C1, whereas cells devoid of FcR\(\gamma\) will enable us to estimate PIR-B expression. Comparison of flow cytometric data on cells from either Pirb\(^{−/−}\)–30 or Fcrg\(^{−/−}\) mice\(^38\) revealed the more dominant expression of PIR-B than PIR-A on splenic macrophages, splenic DC and bone marrow-derived cultured mast cells and exclusive expression of PIR-B on splenic B cells. The consistent observations from either PIR-B or FcR\(\gamma\) deletion suggested that the PIR-B deletion did not largely alter PIR-A expression in these cells, and vice versa. Thus, PIR-A and PIR-B surface expression is characteristic for each cell type, at least in its resting state, and the suppression of these cells by dominantly expressed PIR-B may have a physiological importance, such as in maintaining their resting state upon continuous interaction with H-2.

**Pirb\(^{−/−}\) B cells, neutrophils and macrophages are hyperresponsive**

The PIR-B molecules in macrophages and B cells are constitutively phosphorylated,\(^7\) and this is presumably induced by constitutive interaction with H-2 molecules. Pirb\(^{−/−}\) splenic B cells showed significantly enhanced proliferation upon anti-BCR F(\(ab\)')\(_2\) stimulation. When stimulated with anti-BCR whole IgG antibodies after blocking with anti-Fc\(\gamma\)RIIB monoclonal antibody, the enhanced Pirb\(^{−/−}\) B-cell proliferation was more pronounced due to masking of an inhibitory effect by Fc\(\gamma\)RIIB. This indicates that the inhibitory effects by PIR-B and Fc\(\gamma\)RIIB are additive, possibly because of the fact that PIR-B uses the SHP-1 cascade for inhibition whereas Fc\(\gamma\)RIIB uses SHIP.\(^30\) These results indicate that Pirb\(^{−/−}\) B cells are hypersensitive to stimulation via BCR ligation. Consistent with this observation, Pirb\(^{−/−}\) mice showed a higher IgM response against T-independent antigens trinitrophenol (TNP)-Ficoll and TNP-lipopolysaccharide (LPS). The enhanced response upon TNP-LPS challenge suggested that the Pirb-B inhibitory effect could also be exerted independently of BCR ligation. Enhanced tyrosine phosphorylation of cellular proteins in Pirb\(^{−/−}\) B cells even in the resting state indicated the constitutive activation of Pirb\(^{−/−}\) B cells.\(^30\) Thus, Pir-B may down-regulate BCR signalling by interacting with H-2 molecules (Fig. 2). A Pir-B deficiency may generally render B cells active and hyperresponsive to stimulation via BCR. However, serum IgM levels were not increased in naïve Pirb\(^{−/−}\) mice. In addition, anti-double-stranded DNA antibodies were not detected in adult Pirb\(^{−/−}\) mice of C57BL/6 background, in contrast with CD22-deficient mice, in which hyper-IgM and autoantibody production is evident.\(^39,40\)

It was pointed out that PIR-B is directly involved in integrin signalling in neutrophils and macrophages. Pereira \textit{et al.}\(^41\) observed that Lyn-deficient neutrophils showed enhanced respiratory burst, granule release, and a hyper-adhesive phenotype upon engagement of surface integrins. Lyn-deficient macrophages also showed a hyperadhesive phenotype. It was shown that the Lyn plays an essential role in the adhesion-dependent phosphorylation of the ITIM of the inhibitory receptors SIRP\(\alpha\) and PIR-B, which in turn recruit the phosphatase SHP-1. Reduced mobilization of SHP-1 to the membrane in Lyn-deficient neutrophils results in a hyperadhesive and hyperactive phenotype. Thus, it is apparent that Lyn kinase functions in these cases as a negative regulator in integrin signalling via PIR-B and SIRP\(\alpha\). In line with this observation, Pirb\(^{−/−}\) neutrophils displayed enhanced respiratory burst, secondary granule release, and a hyperadhesive phenotype upon integrin engagement.\(^42\) Bone marrow-derived macrophages from Pirb\(^{−/−}\) mice were also hyper-adhesive and spread more rapidly than wild-type cells upon cross-linking of the cellular \(\beta_2\) integrins. Phosphorylation and
activation of proteins involved in integrin signalling did occur in such Pirb−/− macrophages. Thus, PIR-B is critical in the regulation of neutrophil and macrophage integrin signalling, suggesting a significant role of PIR in inflammatory responses.

Altered humoral response in PIR-B deficiency

Th2-type humoral responses were augmented in Pirb−/− mice upon immunization with TNP-keyhole limpet haemocyanin or ovalbumin with alum adjuvant, in terms of both IL-4-rich and interferon-γ (IFN-γ)-poor cytokine profiles and enhanced IgG1 and IgE production. At least one of the mechanisms for the T helper 2 (Th2)-skewed responses in Pirb−/− mice was suggested to be the immature phenotype of DC. Flow cytometric analysis of surface markers, such as MHC class II, CD80 and CD86, on bone marrow-derived cultured DC (BMDC) before and after antigen loading revealed that the BMDC from Pirb−/− mice were immature. The production of IL-12, a Th1-polarizing cytokine, was diminished upon antigen loading of BMDC from Pirb−/− mice. Successful adoptive transfer of a Th2-prone response by Pirb−/− BMDC into wild-type mice strongly suggested that the impaired maturation of DC would be responsible for the skewing. To test the possibility that intracellular signalling may differ between Pirb−/− BMDC and wild-type cells, BMDC were stimulated with granulocyte–macrophage colony-stimulating factor (GM-CSF), an inducer cytokine for DC development, and were examined for their protein tyrosine phosphorylation profile. PIR-B was tyrosine phosphorylated in resting wild-type DC similar to that found in resting B cells. The tyrosine phosphorylation of PIR-B was augmented upon GM-CSF stimulation, indicating that PIR-B is involved in the cytokine signalling. Upon GM-CSF stimulation, altered phosphorylation profiles of total cellular phosphotyrosyl proteins as well as GM-CSF receptor common β chain were observed in Pirb−/− DC when compared to those of the wild-type cells. It is conceivable that PIR-B deficiency leads to altered phosphorylation profiles of GM-CSF signalling in the absence of inhibitory signal initiated upon H-2 binding of PIR-B, which yields immature DC. Thus, PIR-B and H-2 interaction is also critical for DC maturation and for regulating humoral responses.

Transplantation and PIR

Feto–maternal tolerance is important physiological system for maternal unresponsiveness to fetus, a kind of transplant for mothers. The non-classical MHC class I molecule HLA-G is selectively expressed on fetal trophoblast tissue at the feto–maternal interface in pregnancy. It has long been suggested that HLA-G may inhibit maternal NK cells through interaction with particular inhibitory KIRs. However, this feto–maternal tolerance may be substantially supported by inhibitory immunoglobulin-like receptors expressed on maternal DC and monocytes. On transfectants in fact, HLA-G tetramers can bind to inhibitory KIR LILRB1 (or ILT2) and LILRB2 (or ILT4). HLA-G tetramer binding to blood monocytes is largely caused by binding to LILRB2. Interestingly, the human HLA-G can also bind to murine PIR-B and modifies the function of murine DC. HLA-G tetrameric complexes inhibit maturation of murine BMDC in vitro, similar to what occurs in HLA-G-transgenic mice. PIR-B is highly phosphorylated on BMDC from HLA-G-transgenic mice. It should be important to dissect these observations in the murine system, in which mouse MHC class Ib molecule, such as Qa-1, may modulate PIR-B inhibitory function in murine DC.

Matching HLA haplotypes between donor and recipient is a critical determinant of the outcome of organ, tissue, and haematopoietic stem cell transplantation that is a crucial technology for therapy of malignant leukaemia. However, graft-versus-host disease (GVHD) caused by mismatched HLA often hampers successful transplantation. In GVHD, activation of donor alloreactive cytotoxic T cells requires prior activation of donor CD4+ T cells recognizing host allogeneic MHC class I molecules with peptides on the host antigen-presenting cells. To test whether the possible interaction between allogeneic MHC class I molecules and PIR in transplantation, Nakamura et al. employed a lethal GVHD model, in which sublethally irradiated, Pirb−/− or wild-type host mice received allogeneic splenocytes intravenously. Pirb−/− mice showed accelerated, lethal GVHD because of the augmented activation of the recipient’s DC with the concomitant up-regulation of PIR-A and enhanced production of IFN-γ from DC, CD4+ T cells and cytotoxic T cells.

In a rat model of heart transplantation, Liu et al. reported the importance of CD8+ T suppressor cells. CD8+ T suppressor cells from tolerant rats could transfer the tolerance to naive hosts and induced the up-regulation of rat PIR-B in allogeneic DC and heart endothelial cells. The heart allograft with PIR-B-positive endothelial cells were retransplanted to a secondary allogeneic recipient, they did not elicit rejection. In the human system, it was reported that human T suppressor cells rendered professional and non-professional antigen-presenting cells, such as DC and endothelial cells, tolerogenic inducing the down-regulation of costimulatory molecules and up-regulation of the inhibitory immunoglobulin-like receptors of human PIR-B orthologues, LILRB4 (or ILT3) and LILRB2 (or ILT4).

Conclusion

Various immune cells may have adopted recognition of ubiquitously expressed, MHC class I molecules as a
common strategy to inhibit cellular activation. Thus, PIR-A and PIR-B provide a novel system for MHC class I recognition with physiological and pathological significance, where PIR-A and PIR-B regulate the threshold for activation of cells, such as B cells, mast cells, neutrophils, macrophages and DC, and prevent undesired reaction to autologous tissues in a constitutive fashion. They become critical for successful tissue and haematopoietic stem cell transplantation. Further analysis of detailed structural features of interaction between PIR and MHC class I molecules as well as characteristics of Pirb<sup>−/−</sup> and Fcγ<sup>−/−</sup> mice will provide us with further information about the physiological role of PIR and pathophysiological aspects of its involvement in infection, hypersensitivity and autoimmune diseases.

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**References**

29 Shiroishi M, Tsuimoto K, Amano K et al. Human inhibitory receptors Ig-like transcript 2 (ILT2) and ILT4 compete with CD8 for MHC class I binding and bind preferentially to HLA-G. Proc Natl Acad Sci USA 2003; 100:8856–61.
43 Magram J, Connaughton SE, Warrier RR et al. IL-12-deficient mice are defective in IFNγ production and type 1 cytokine responses. Immunity 1996; 4:471–81.