Fc γ receptors and the complement system in T cell activation

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Chapter 4

Murine Fc Receptors for IgG are redundant in facilitating presentation of immune complex derived antigen to $CD8^+$ T cells *in vivo*.

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Abstract

Antigen(Ag)–immunoglobulin (Ig)G complexes (IC) are more efficiently processed and presented than soluble Ag. IC can bind to various cell types via different types of Fc-Receptors or, upon binding to complement factors, by complement receptors. Murine professional antigen-presenting cells (APC) express four types of FcγReceptors (FcγR) via which they are able to capture IC; three activating receptors (FcγRI, III and IV) and one inhibitory receptor (FcγRII). It has been demonstrated that FcγR play a pivotal role in facilitating the presentation of Ag derived from IC. Nonetheless, relative little information is available on the relative contribution of the activating or inhibitory FcγR or complement to the presentation of immune-complexed Ag to CD8+ T cells.

To study the contribution of the different FcγR and complement receptors in IC-facilitated Ag-presentation, we analyzed the ovalbumin(OVA)-specific CD8+ T cell proliferation in FcγR- and complement component 3 (C3)-deficient mice after subcutaneous injection of OVA-IC. Here we show that the efficient Ag-presentation was FcγR-, but not C3-mediated, as it was inhibited in FcγRI/II/III-deficient mice but unaffected in the C3-depleted mice. Moreover, FcγRIV does not play a role under these conditions. However, no difference was found between wild-type and FcγRI/III-deficient or wild-type and FcγRII-deficient mice. These results indicate that Ag-presentation via the activating FcγR is not enhanced in the absence of FcγRII, and point to redundancy of the FcγR, including FcγRII, in the uptake and presentation of s.c. injected soluble IC to CD8+ T cells.

Keywords: FcγReceptors; Immune complexes; Antigen presentation; CD8+ T cells

1. Introduction

MHC class I molecules are generally complexed with peptides derived from newly synthesized cytosolic proteins. However, professional APC are also able to process and present exogenous antigens leading to the activation of naïve CD8+ T cells. Ag derived from apoptotic cells, antigen-heat shock protein-complexes and Ag-immunoglobulin (Ig)G complexes (IC) are shown to be presented by DC to T cells in a process called cross-presentation (Heath and Carbone, 2001). For the uptake of exogenous antigen, DC display several receptors like mannose-receptors, CD91, CD36, complement receptors and FcReceptors (FcR).

FcR are receptors for the Fc part of the constant region of Ig, providing a link between the cellular and the humoral part of the immune response. FcR are membrane glycoproteins, which belong to the Ig supergene family. There are different FcR for each Ig class: FcαR (IgA), FcεR (IgE), FcγR (IgG) and Fcα/μR (IgA and IgM).

In mice, four types of FcγR are described, i.e. three activating receptors, FcγRI (CD64), FcγRIII (CD 16), and the recently

Abbreviations: Ag, antigen; APC, antigen presenting cells; BSA, bovine serum albumin; C3, complement component 3; CFSE, 5(6)-carboxyfluorescein diacetate succinimidyl ester; CVF, cobra venom factor; FcγR, receptor for the Fc part of the IgG molecule; FCS, fetal calf serum; IC, immunecomplexes; Ig, immunoglobulin; ITAM, immunoreceptor tyrosine-based activation motif; ITIM, immunoreceptor tyrosine-based inhibitory motif; OVA, ovalbumin; PBS, phosphate-buffered saline

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identified FcγRIV (Mechetina et al., 2002; Nimmerjahn et al., 2005), and one inhibitory receptor, FcγRII (CD32) (Ravetch and Kinet, 1991; Ravetch and Bolland, 2001). The activating FcγR are associated with the signal transducing γ-chain (FcRγ) homodimer, which contains an immunoreceptor tyrosine-based activation motif (ITAM) in its cytoplasmic tail. In contrast, the inhibitory FcγR is a single-chain receptor containing an immunoreceptor tyrosine-based inhibitory motif (ITIM) in its cytoplasmic domain.

Activating FcγR are expressed by most effector cells of the immune system, particularly monocytes, macrophages, NK cells, mast cells, eosinophils, and neutrophils, while absent from lymphoid cells. The inhibitory receptor FcγRII is expressed on all hematopoietic cells except T and NK cells. In general, activating and inhibitory FcγR are co-expressed on the same cell. The IgG ligand will then be able to cross-link both receptor types, which is important for setting a threshold for activating stimuli. The relative expression of these two opposing signaling systems is likely to determine the cellular response (Ravetch and Clynes, 1998). B cells only express FcγRII on their surface. Here, FcγRII inhibits Ab responses to IgG-complexed Ag when it is co-ligated to the BCR (Ravetch and Bolland, 2001). Although FcγR are important to facilitate presentation of Ag complexed in IC, the relative contribution of the different FcγR or the possible negative regulation mediated by FcγRII on this process in vivo has not been studied in detail.

Mice-deficient for the different FcγR provide excellent tools to analyze the specific contribution of the individual FcγR to IgG-dependent immune functions (Ioan-Facsinay et al., 2002; Hazenbos et al., 1996). To study the relative contribution of the four different FcγR in facilitating presentation of IC-derived Ag, we analyzed the activation of OVA-specific $CD8^+$ T cells (OT-I) upon subcutaneous injection of OVA-IC in FcγR knock out and wild-type mice. Previous studies have already shown that Ag internalized through FcγR are more efficiently presented to T cells (Lanzavecchia, 1996; Regnault et al., 1999; Hamano et al., 2000). Our results confirm that IgG-complexed OVA is at least 10 times more efficient than soluble OVA in activating Ag-specific $CD8^+$ T cells in vivo. We now also show that this effect is absent in mice lacking FcγRI, II and III, but not in C3-deficient mice, indicating that the enhanced efficiency was complement independent and primarily FcγR mediated but independent of FcγRIV. However, no difference was found between wild-type and FcγRI/III-deficient mice, between wild-type and γ-chain-deficient mice or between wild-type and FcγRII-deficient mice.

These results indicate that the activating receptors FcγRI and FcγRIII as well as the inhibiting FcγRII can mediate the uptake and presentation of IC to T cells and illustrate that the activation of CTL via IC uptake by the activating FcγRI and FcγRIII is not enhanced in the absence of the inhibitory FcγRII.

2. Materials and methods

2.1. Mice

C57BL/6K^h (B6; H-2^b) mice and OT-I/Ly5.1 transgenic mice, which have a transgenic Vα2Vβ5 TCR specific for the $OVA_{257–264}$ epitope in the context of H2-K^b, were bred in our specific pathogen-free animal facility. FcγRI/II/III-deficient mice, FcγRI/III-deficient mice, FcγRII-deficient mice and their wild-type counterparts were generated in our own laboratory. C3-deficient mice and FcRγ-chain-deficient mice were kindly provided respectively by Dr. Carroll and Dr. Saito.

2.2. Depletion with cobra venom factor

To deplete complement factors in vivo, B6 mice were injected twice i.p. with 500 Units Cobra Venom Factor (CVF; Naja naja Kaoutchia; MP Biomedicals, Aurora, OH). The second injection took place 6 h after the first. To determine whether complement factors were depleted, serum of these mice was analyzed next day by ELISA for C3.

2.3. CFSE labeling

To follow T cell proliferation in vivo, OT-I/Ly5.1$^+$ cells were labeled with the intracellular fluorescent dye CFSE (Molecular Probes, Leiden, the Netherlands). Spleen and lymph node cells from OT-I/Ly5.1$^+$ mice were depleted for dendritic cells using CD11c-specific microbeads and the MACS system (LS$^+$ columns) (Milteny, CLB, Amsterdam, The Netherlands) according to manufacturer's instructions. Cells were washed with PBS containing 0.1% bovine serum albumin (BSA; Sigma–Aldrich, Zwijndrecht, the Netherlands) and resuspended to 10×10^6 cells/ml in PBS/0.1% BSA with 5 μM CFSE for 10 min at 37 °C. Ten percent heat-inactivated fetal calf serum (FCS; Bodinco, Alkmaar, the Netherlands) was added and cells were washed twice with IMDM (BioWhittaker, Europe), supplemented with 8% FCS, 100 IU/ml penicillin/streptavidin (BioWhittaker), 2 mM L-glutamine (Invitrogen, Breda, the Netherlands) and 20 μM 2-mercaptoethanol (Merck, Hohenbrunn, Germany), and resuspended in PBS/1% BSA. 3×10^6 $CD8^+V\alpha2^+V\beta5^+$ OT-I cells in 200 μl PBS/0, 5%BSA were injected i.v. into the mice.

2.4. Generation of OVA immune complexes

Immune complexes were generated by incubating soluble OVA (Sigma–Aldrich) with polyclonal OVA-specific rabbit IgG (rIgGαOVA) (MP Biomedicals), at a ratio of 1 μg OVA to 25 μg rIgGαOVA in PBS, for 30 min at 37 °C. IC were then injected s.c. into the right flank of the mice.

2.5. Antibodies

To analyze proliferation of the OT-I cells in vivo, draining and non-draining lymph nodes were collected from the mice. Lymph node cells were stained with APC-coupled anti-CD8α Ab (Ly-2) and PE-coupled anti-CD45.1 Ab (Ly5.1) (both from PharMingen, San Diego, CA). Stainings were performed for 20 min at 4 °C. $CFSE^+CD8^+Ly5.1^+$ cells were analyzed using a FACSCalibur® flow cytometer equipped with CellQuest software (Becton Dickinson, San Jose, CA).

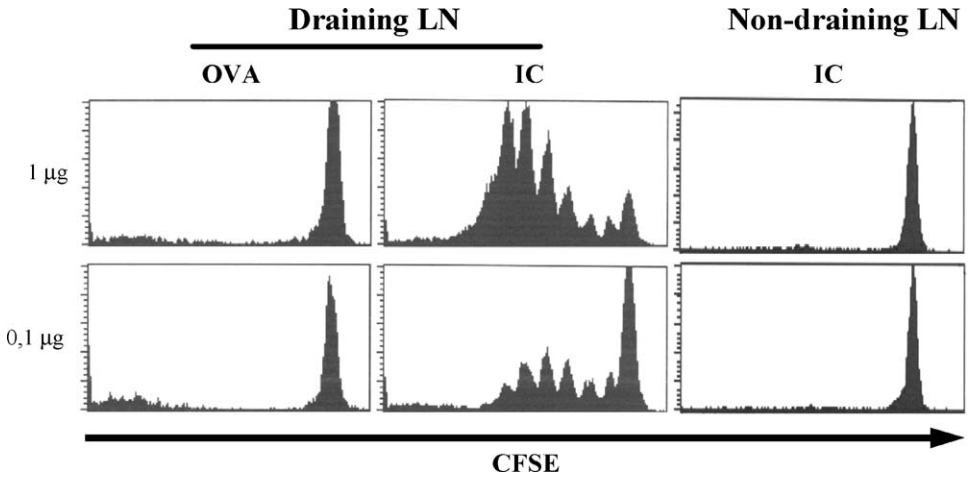

Fig. 1. IC are at least 10 times more efficient then soluble OVA in inducing T cell proliferation. CFSE labeled OT-I/Ly5.1$^+$ cells were transferred into wild-type mice. These mice were subsequently injected s.c. with different concentrations of soluble OVA or OVA complexed to anti-OVA IgG ($n = 2$). Three days after injection, proliferation of the CD8$^+$/Ly5.1$^+$ T cells was analyzed in the draining and non-draining lymph nodes. One representative experiment out of three performed is presented.

3. Results

3.1. Efficient IC-mediated CD8$^+$ T cell proliferation in vivo

Professional APC are able to capture, process and present exogenous Ag leading to activation of T cells. Several in vitro studies showed that Ag complexed with IgG are much more efficiently presented than soluble Ag in a FcγR-mediated fashion (Amigorena and Bonnerot, 1999; Manca et al., 1991). To confirm and extend these data, wild-type mice were injected i.v. with CFSE labeled OVA-specific T cells derived from OT-I/Ly5.1$^+$ mice and treated s.c. with different concentrations of soluble OVA or OVA bound to IgGαOVA. Three days after injection of the antigen, T cell proliferation in the draining lymph and non-draining nodes was analyzed.

In some, but not all mice, soluble OVA induced T cell proliferation in the draining lymph nodes when a dose of 10 μg/mouse (data not shown) was injected. No proliferation was detected when a dose of 1 μg/mouse or lower soluble OVA was administrated (Fig. 1). In contrast, injection of OVA incubated with anti-OVA IgG still resulted in significant proliferation of OT-I cells in the draining lymph nodes even when injected at a dose of 0.1 μg OVA/mouse. These results indicate that subcutaneously given IC are at least 10 times more efficient in inducing CD8$^+$ T cell proliferation than soluble OVA in vivo.

3.2. FcγR are involved in IC-mediated Ag-presentation, C3 is not

Uptake of IC can be mediated via FcγR, from which there are four types known in mice: FcγRI, II, III and IV. Alternatively, complement factors can form complexes with IC that subsequently can be taken up via complement receptors. To analyze which receptors are most important in IC uptake and presentation in vivo, Ag-presentation after injection of IC was analyzed in different FcγR-deficient and C3-deficient mice.

As shown in Fig. 2A and B, CD8$^+$ T cell proliferation was strongly reduced in the FcyRI/II/III$^{-/-}$ mice compared to the T cell proliferation in wild-type mice after injection of 0.1 μg IC. No difference in T cell proliferation was observed between wild-type mice and mice treated with cobra venom factor, which depletes active C3 and C5 and thereby inhibits the effector function of the complement system (Fig. 2C). Similar results were obtained in the C3$^{-/-}$ mice (data not shown). Together, these results indicate that, after s.c. injection of IC, FcγR play the most prominent role in uptake and presentation of Ag to CD8$^+$ T cells, while activation of C3 does not seem to be involved in this process.

3.3. No difference between WT and FcγRI/III-deficient mice

To investigate the contribution of the three activating FcγR, FcγRI, III and IV, to IC-facilitated Ag-presentation in vivo, T cell proliferation was compared between wild-type and γ-chain$^{-/-}$ mice, which lack FcγRI, III and IV, upon immunization with IC. As shown in Fig. 3A, no difference in CD8$^+$ T cell proliferation was found between wild-type en γ-chain$^{-/-}$ mice when immunized with IC s.c. However, recent publications suggest that the γ-chain$^{-/-}$ mice still express low amounts of FcγRI (Barnes et al., 2002). Therefore, similar experiments were also performed in the FcyRI/III$^{-/-}$ mice. Fig. 3B shows that also in these mice, proliferation of the CD8$^+$ T cells was equally efficient in wild-type and FcyRI/III$^{-/-}$ mice. Thus, FcγRI, III and IV are not crucial for IC-mediated activation of OVA-specific CTL.

3.4. FcγR seem to be redundant

As there is a reduced T cell proliferation in the FcyRI/II/III$^{-/-}$ mice upon s.c. injection of IC, which is not observed in γ-chain$^{-/-}$ and FcyRI/III$^{-/-}$ mice, we also wished to analyze the contribution of FcγRII to this process. Therefore, CD8$^+$ T cell

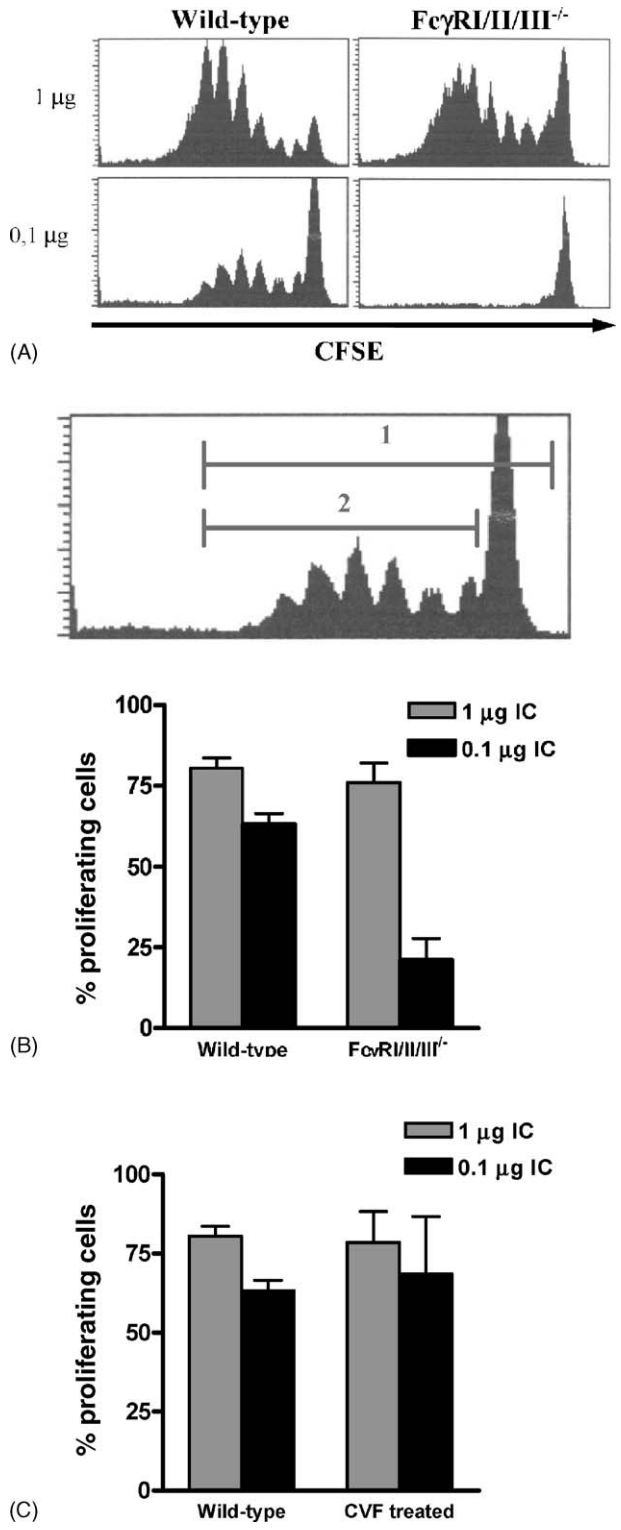

Fig. 2. Reduced T cell proliferation in FcγRI/II/III-deficient mice. CFSE labeled OT-I/Ly5.1$^+$ cells were transferred into wild-type mice, FcγRI/II/III$^{-/-}$ mice (A and B) or Cobra Venom Factor treated mice (C). These mice were injected s.c. with different concentrations of IC ($n = 2$). Three days after injection, proliferation of the CD8+/Ly5.1$^+$ T cells was analyzed in the draining lymph nodes. One experiment out of four (A and B) or two (C) performed is presented. B: In next figures the results will be shown in column graph as follow: the percentage of cell that are proliferating (2) from the total amount of CD8+/Ly5.1$^+$ cells (1). (A) would than look as shown here. One bar of the diagram represents the average S.D. of two animals per group. One experiment out of four performed is presented.

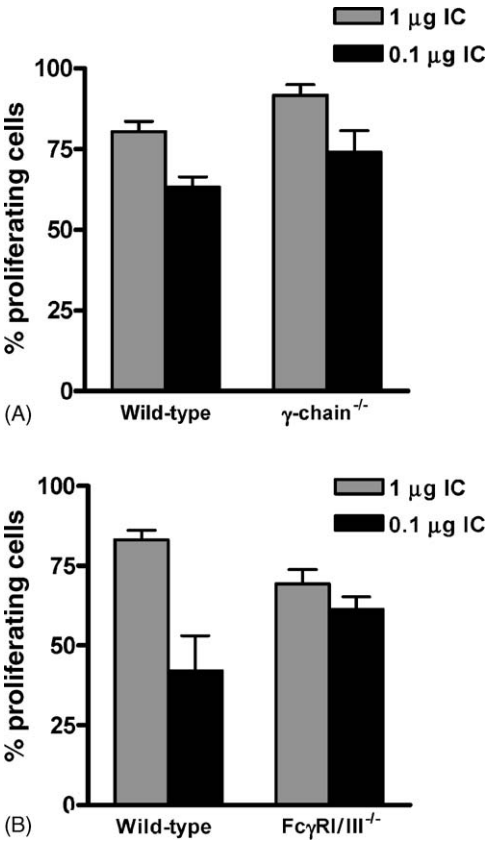

Fig. 3. No change in T cell proliferation in γ-chain-deficient mice and in FcγRI/III-deficient mice. CFSE labeled OT-I/Ly5.1$^+$ cells were transferred into wild-type mice, γ-chain$^{-/-}$ mice (A), or FcyRI/III$^{-/-}$ mice (B). These mice were injected s.c. with different concentrations of IC ($n = 3$ for γ-chain$^{-/-}$, $n = 4$ for FcyRI/III$^{-/-}$). Three days after injection, proliferation of the CD8+/Ly5.1$^+$ T cells was analyzed in the draining lymph nodes. One experiment out of three performed is presented.

proliferation in FcyRII$^{-/-}$ mice was analyzed after s.c. injection of IC. As shown in Fig. 4, there was also no change in T cell proliferation in mice lacking FcγRII, compared to wild-type mice. Because the absence of FcγRII does not lead to enhanced

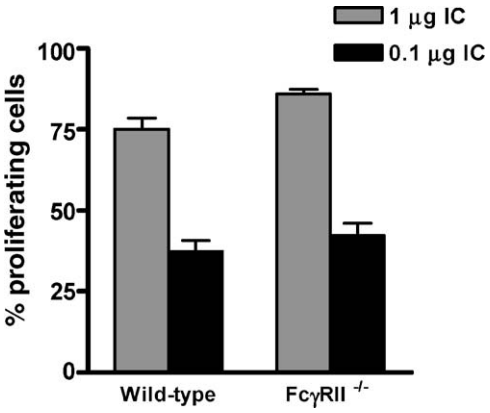

Fig. 4. No change in T cell proliferation in FcγRII-deficient mice. CFSE labeled OT-I/Ly5.1$^+$ cells were transferred into wild-type or FcyRII$^{-/-}$ mice. These mice were injected s.c. with different concentrations of IC ($n = 4$). Three days after injection, proliferation of the CD8+/Ly5.1$^+$ T cells was analyzed in the draining lymph nodes. One experiment out of three performed is presented.

activation of $CD8^+$ T cells following s.c. injection of IC, these findings indicate that FcγRII does not inhibit IC-facilitated $CD8^+$ T cell activation. Moreover, these observations indicate redundancy of the FcγR in the uptake and presentation of IC to $CD8^+$ T cells, as the lack of three FcγR (FcγRI, II and III) did lead to a reduced $CD8^+$ T cell proliferation, whereas the lack of one or two of these receptors did not.

4. Discussion

In this study, we investigated the contribution of the different murine FcγR and complement receptors to the activation OVA-specific $CD8^+$ T cells in vivo, following s.c. vaccination with OVA-IC. Previously, it has been shown that IC are much more efficiently presented than soluble OVA in a FcγR-mediated fashion (Amigorena and Bonnerot, 1999; Manca et al., 1991). In vitro data support that the enhancement of MHC class I-restricted Ag-presentation is mediated by the activating FcγR (FcγRI and FcγRIII), as γ-chain$^{-/-}$ DC are not activated by IC, and do not present IC-derived MHC class I-binding peptides more efficiently than peptides derived from soluble Ag alone (Regnault et al., 1999). Our results show that, also in vivo, FcγR, but not C3, are involved in IC-mediated Ag-presentation. However, in contrast to the studies described above, FcγR (including the inhibitory FcγRII) seem to be redundant, as deficiency in only one or two of the FcγR does not alter the IC-induced T cell proliferation. These contrasting findings could be related to the fact that we analyzed the contribution of the different FcγR in vivo, allowing participation of all APC- and DC-populations. Whereas the studies performed in vitro/ex vivo analyzed the contribution of the FcγR on selected populations of DC that might not represent all DC-subsets present in vivo.

Recently, a novel murine FcγR is described: FcγRIV (Mechetina et al., 2002; Nimmerjahn et al., 2005). Our observations exclude a role for FcγRIV in IC-mediated Ag-presentation under the conditions used. $CD8^+$ T cell proliferation was strongly reduced in the FcγRIV expressing FcγRI/II/III$^{-/-}$ mice and T cell proliferation was unaltered in the γ-chain deficient mice which do not express FcγRIV (and FcγRI and FcγRIII).

The FcγR-dependent cross-presentation is thought to be a unique property of DC as these cells, but not macrophages and B cells, possess a specialized cross-presentation transport system for MHC class I Ag-presentation (den Haan et al., 2000; Jung et al., 2002; Regnault et al., 1999; Rodriguez et al., 1999; Schuurhuis et al., 2002). Previous publications have shown that the enhanced cross-presentation of IC in vivo is depended on the gamma-chain-containing activating FcγR only in a subset of DC (the $CD8^-$) (den Haan and Bevan, 2002). The $CD8^+$ DC are shown to present IC also in the absence of the three activating receptors (FcRγ$^{-/-}$). This is in line with our results, as probably also FcγRII is involved in uptake and presentation of IC-derived antigens. FcγRII is thought to inhibit cellular responses to IC when it is cross-linked to the activating FcγR (Ravetch and Clynes, 1998). However, to date, there are no reports demonstrating that FcγRII negatively regulates Ag-presentation via activating FcγR. Recent studies argue against an inhibitory effect of FcγRII on FcγR-mediated Ag-presentation as it is shown that DTH reactions take place normally in FcγRII$^{-/-}$ animals (Getahun et al., 2004) and Ag-targeting to FcγRII even accelerates Ag-presentation (Yada et al., 2003). In contrast, an abundance of in vitro data demonstrate that FcγRII does indeed negatively regulate B cell activation (Ashman et al., 1996; Bijsterbosch and Klaus, 1985; Choquet et al., 1993; Minskoff et al., 1998; Muta et al., 1994; Nakamura and Cambier, 1998; Wagle et al., 1999). However, other in vitro data show that the B cell isoform of FcγRII is able to mediate capture and presentation of some Ag–Ab complexes and might play a role in BCR-independent Ag-presentation in vivo (Antoniou and Watts, 2002). These latter results are in line with our observation that in FcγRI/III-deficient mice, proliferation of OVA-specific $CD8^+$ T cells was not altered compared to wild-type mice, indicating that also FcγRII can mediate the uptake and presentation of IC.

It has been shown that resting DC induce peripheral tolerance of $CD8^+$ T cells, whereas activated DC induce priming (Probst et al., 2003; Probst et al., 2005). As we analyzed $CD8^+$ T cell proliferation only 3 days after IC injection, our results do not conclude that Ag-presentation mediated by FcγRII induces an effector T cell response. Earlier publications showed that targeting of antigen to the DC receptor DEC-205 in the steady state induced solid peripheral $CD8^+$ T cell tolerance (Bonifaz et al., 2002). Here, targeting of αDEC-205: Ag to DC in the steady state initially induced four to seven cycles of T cell proliferation, but the T cells were then deleted and the mice became specifically unresponsive to rechallenge with Ag. Other studies showed that FcγRII blockade leads to maturation of human monocyte-derived DC and to increased Ag-specific T cell immunity, suggesting that the balance between activating and inhibitory FcγR is important in modifying the immunity to IC (Dhodapkar et al., 2005). Therefore, it might be possible that cross-linking of the FcγRII receptor does not properly activate the DC, which finally results in a tolerogenic immune response. In contrast, cross-linking only the activating FcγR might results in a robust effector $CD8^+$ T cell response. Our results provide a rationale to further study this possibility as they indicate that FcγRII is able to mediate Ag-presentation after injection of IC, and thus could have the ability to silence CTL-reactions through the direct capture and presentation of immune-complexed Ag.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.molimm.2006.01.002.

References

Amigorena, S., Bonnerot, C., 1999. Fc receptor signaling and trafficking: a connection for antigen processing. Immunol. Rev. 172, 279–284.

Antoniou, A.N., Watts, C., 2002. Antibody modulation of antigen presentation: positive and negative effects on presentation of the tetanus toxin antigen via the murine B cell isoform of FcgammaRII. Eur. J. Immunol. 32, 530–540.

Ashman, R.F., Peckham, D., Stunz, L.L., 1996. Fc receptor off-signal in the B cell involves apoptosis. J. Immunol. 157, 5–11.

Barnes, N., Gavin, A.L., Tan, P.S., Mottram, P., Koentgen, F., Hogarth, P.M., 2002. FcgammaRI-deficient mice show multiple alterations to inflammatory and immune responses. Immunity 16, 379–389.

Bijsterbosch, M.K., Klaus, G.G., 1985. Crosslinking of surface immunoglobulin and Fc receptors on B lymphocytes inhibits stimulation of inositol phospholipid breakdown via the antigen receptors. J. Exp. Med. 162, 1825–1836.

Bonifaz, L., Bonnyay, D., Mahnke, K., Rivera, M., Nussenzweig, M.C., Steinman, R.M., 2002. Efficient targeting of protein antigen to the dendritic cell receptor DEC-205 in the steady state leads to antigen presentation on major histocompatibility complex class I products and peripheral CD8(+) T cell tolerance. J. Exp. Med. 196, 1627–1638.

Choquet, D., Partiseti, M., Amigorena, S., Bonnerot, C., Fridman, W.H., Korn, H., 1993. Cross-linking of IgG receptors inhibits membrane immunoglobulin-stimulated calcium influx in B lymphocytes. J. Cell Biol. 121, 355–363.

den Haan, J.M., Bevan, M.J., 2002. Constitutive versus activation-dependent cross- presentation of immune complexes by CD8(+) and CD8(−) dendritic cells in vivo. J. Exp. Med. 196, 817–827.

den Haan, J.M., Lehar, S.M., Bevan, M.J., 2000. CD8(+) but not CD8(−) dendritic cells cross-prime cytotoxic T cells in vivo. J. Exp. Med. 192, 1685–1696.

Dhodapkar, K.M., Kaufman, J.L., Ehlers, M., Banerjee, D.K., Bonvini, E., Koenig, S., Steinman, R.M., Ravetch, J.V., Dhodapkar, M.V., 2005. Selective blockade of inhibitory Fcgamma receptor enables human dendritic cell maturation with IL-12p70 production and immunity to antibody-coated tumor cells. Proc. Natl. Acad. Sci. U.S.A. 102, 2910–2915.

Getahun, A., Dahlstrom, J., Wernersson, S., Heyman, B., 2004. IgG2a-mediated enhancement of antibody and T cell responses and its relation to inhibitory and activating Fc gamma receptors. J. Immunol. 172, 5269–5276.

Hamano, Y., Arase, H., Saisho, H., Saito, T., 2000. Immune complex and Fc receptor-mediated augmentation of antigen presentation for in vivo Th cell responses. J. Immunol. 164, 6113–6119.

Hazenbos, W.L., Gessner, J.E., Hofhuis, F.M., Kuipers, H., Meyer, D., Heijnen, I.A., Schmidt, R.E., Sandor, M., Capel, P.J., Daeron, M., van de Winkel, J.G., Verbeek, J.S., 1996. Impaired IgG-dependent anaphylaxis and Arthus reaction in Fc gamma RIII (CD16) deficient mice. Immunity 5, 181–188.

Heath, W.R., Carbone, F.R., 2001. Cross-presentation in viral immunity and self-tolerance. Nat. Rev. Immunol. 1, 126–134.

Ioan-Facsinay, A., de Kimpe, S.J., Hellwig, S.M., van Lent, P.L., Hofhuis, F.M., van Ojik, H.H., Sedlik, C., da Silveira, S.A., Gerber, J., de Jong, Y.F., Roozendaal, R., Aarden, L.A., van den Berg, W.B., Saito, T., Mosser, D., Amigorena, S., Izui, S., van Ommen, G.J., van Vugt, M., van de Winkel, J.G., Verbeek, J.S., 2002. FcgammaRI (CD64) contributes substantially to severity of arthritis, hypersensitivity responses, and protection from bacterial infection. Immunity 16, 391–402.

Jung, S., Unutmaz, D., Wong, P., Sano, G., De los, S.K., Sparwasser, T., Wu, S., Vuthoori, S., Ko, K., Zavala, F., Pamer, E.G., Littman, D.R., Lang, R.A., 2002. in vivo depletion of CD11c(+) dendritic cells abrogates priming of CD8(+) T cells by exogenous cell-associated antigens. Immunity 17, 211–220.

Lanzavecchia, A., 1996. Mechanisms of antigen uptake for presentation. Curr. Opin. Immunol. 8, 348–354.

Manca, F., Fenoglio, D., Li, P.G., Kunkl, A., Celada, F., 1991. Effect of antigen/antibody ratio on macrophage uptake, processing, and presentation to T cells of antigen complexed with polyclonal antibodies. J. Exp. Med. 173, 37–48.

Mechetina, L.V., Najakshin, A.M., Alabyev, B.Y., Chikaev, N.A., Taranin, A.V., 2002. Identification of CD 16-2, a novel mouse receptor homologous to CD16/Fc gamma RIII. Immunogenetics 54, 463–468.

Minskoff, S.A., Matter, K., Mellman, I., 1998. Fc gamma RII-B1 regulates the presentation of B cell receptor-bound antigens. J. Immunol. 161, 2079–2083.

Muta, T., Kurosaki, T., Misulovin, Z., Sanchez, M., Nussenzweig, M.C., Ravetch, J.V., 1994. A 13-amino-acid motif in the cytoplasmic domain of Fc gamma RIIB modulates B-cell receptor signalling. Nature 369, 340.

Nakamura, K., Cambier, J.C., 1998. B cell antigen receptor (BCR)-mediated formation of a SHP-2-pp120 complex and its inhibition by Fc gamma RIIB1-BCR coligation. J. Immunol. 161, 684–691.

Nimmerjahn, F., Bruhns, P., Horiuchi, K., Ravetch, J.V., 2005. FcgammaRIV: a novel FcR with distinct IgG subclass specificity. Immunity 23, 41–51.

Probst, H.C., Lagnel, J., Kollias, G., van den, B.M., 2003. Inducible transgenic mice reveal resting dendritic cells as potent inducers of CD8+ T cell tolerance. Immunity 18, 713–720.

Probst, H.C., McCoy, K., Okazaki, T., Honjo, T., van den, B.M., 2005. Resting dendritic cells induce peripheral CD8+ T cell tolerance through PD-1 and CTLA-4. Nat. Immunol. 6, 280–286.

Ravetch, J.V., Bolland, S., 2001. IgG Fc receptors. Annu. Rev. Immunol. 19, 275–290.

Ravetch, J.V., Clynes, R.A., 1998. Divergent roles for Fc receptors and complement in vivo. Annu. Rev. Immunol. 16, 421–432.

Ravetch, J.V., Kinet, J.P., 1991. Fc receptors. Annu. Rev. Immunol. 9, 457–492.

Regnault, A., Lankar, D., Lacabanne, V., Rodriguez, A., Thery, C., Rescigno, M., Saito, T., Verbeek, S., Bonnerot, C., Ricciardi-Castagnoli, P., Amigorena, S., 1999. Fcgamma receptor-mediated induction of dendritic cell maturation and major histocompatibility complex class I-restricted antigen presentation after immune complex internalization. J. Exp. Med. 189, 371–380.

Rodriguez, A., Regnault, A., Kleijmeer, M., Ricciardi-Castagnoli, P., Amigorena, S., 1999. Selective transport of internalized antigens to the cytosol for MHC class I presentation in dendritic cells. Nat. Cell. Biol. 1, 362–368.

Schuurhuis, D.H., Ioan-Facsinay, A., Nagelkerken, B., van Schip, J.J., Sedlik, C., Melief, C.J., Verbeek, J.S., Ossendorp, F., 2002. Antigen–antibody immune complexes empower dendritic cells to efficiently prime specific CD8+ CTL responses in vivo. J. Immunol. 168, 2240–2246.

Wagle, N.M., Faassen, A.E., Kim, J.H., Pierce, S.K., 1999. Regulation of B cell receptor-mediated MHC class II antigen processing by FcgammaRIIB1. J. Immunol. 162, 2732–2740.

Yada, A., Ebihara, S., Matsumura, K., Endo, S., Maeda, T., Nakamura, A., Akiyama, K., Aiba, S., Takai, T., 2003. Accelerated antigen presentation and elicitation of humoral response in vivo by Fcgamma. Cell. Immunol. 225, 21–32.