

## Interleukin 18: Tipping the Balance Towards a T Helper Cell 1 Response

Susan L. Swain

*The Trudeau Institute, Saranac Lake, NY 12983*

IL-18 was identified as a factor promoting IFN- $\gamma$  production (1) and it was originally called IFN- $\gamma$ -inducing factor (IGIF). Further study indicated IL-18 had a structure related to IL-1 (2) and later it was found that the IL-18 receptor resembles that for IL-1 (3–4). IL-18, like IL-1 and agents interacting with Toll receptors, signals via MyD88 which activates TNF receptor-associated factor and ultimately nuclear factor  $\kappa$ B (5). Like IL-1, IL-18 is made as an inactive precursor that is cleaved by caspase-1 (interleukin-1-converting enzyme) to produce active cytokine (6). Many cell types have been reported to produce IL-18, including macrophages and dendritic cells (7); IL-18 mRNA or protein is also seen in Kupffer cells (8), astrocytes and microglia (9), intestinal and airway epithelial cells (10), and in keratinocytes (11) and osteoblasts (12). What induces IL-18 has not been extensively studied, but IL-18 is found after bacterial (13) and viral (14) infection and, by inference, in many other infectious diseases. IL-18 production from many cells is constitutive or prolonged after induction (15). An important, but not well-explored, role for IL-18 can also be inferred from the fact that poxviruses make a homologue of IL-18-binding protein, a natural suppressor of IL-18 (16) and also an inhibitor of interleukin-1-converting enzyme (17).

**Targets and Roles of IL-18.** Major targets of IL-18 include macrophages, NK cells (18), T cells (19), and perhaps B cells (20). A major effect of IL-18 is the induction of cytokine synthesis. IL-18 induces IFN- $\gamma$  production from T cells (1, 21), and IL-13 from NK cells and T cells (22), especially in concert with other signals (21).

Two papers in this issue (23, 24) provide compelling evidence that IL-18 plays a key role in protection against infectious disease and shed further light on the nature of that role as well as the mechanism by which it occurs. Papers from Neighbors et al. studying the role of IL-18 in protection against *Listeria monocytogenes* (*Listeria*), and from Helmby et al. in protection against the helminth, *Trichuris muris* (*Trichuris*), both indicate that IL-18 promotes Th1 polarization of the immune response even when IFN- $\gamma$  is not involved, suggesting a broader range of IL-18 targets and actions than was described previously. Together these

studies reinforce the concept of IL-18 as a highly pleiotropic, but decidedly pro-Th1 cytokine, that dramatically enhances both innate and acquired immune responses.

IL-18 induces macrophages to produce IFN- $\gamma$ , but the results of Neighbors and colleagues indicate that it also stimulates macrophages to produce of TNF- $\alpha$  and nitric oxide (NO), and that both of these are critical in IL-18's important role in protection against *Listeria* (23). This is likely to prove to be a key aspect of IL-18 action because such a mechanism could explain the procytotoxic activities of IL-18 (18, 19). Neighbors and colleagues have shown convincingly, using both cytokine knockout and blocking antibody studies, that the action of IL-18 is dominant over those of IL-12 and IFN- $\gamma$  in promoting resistance to *Listeria*, and that the effect is largely independent of those cytokines, but dependent on TNF- $\alpha$ . They also directly demonstrate the ability of IL-18 to induce macrophages to produce TNF- $\alpha$  and NO (23). These results begin to explain the observations in the literature that IL-18 seems to be required for protection against a broad range of pathogens including *Mycobacteria* (25), *Salmonella* (26), *Shigella* (26), *Leishmania* (27), *Staphylococci* (28), and *Cryptococci* (29).

**IL-18 and Cytokine Polarization.** Early studies of IL-18 stressed its IFN- $\gamma$ -inducing abilities and promoted its role as an inducer of Th1 responses (1, 19, 20), but more recently a number of studies and reviews have suggested IL-18 can also enhance production of Th2 cytokines and promote IgE synthesis (30, 31). The papers in this issue indicate that the major in vivo role is likely to be weighted towards IL-18 inducing a Th1 response. Not only did IL-18 mediate protection against *Listeria*, but in the *Trichuris* model, the absence of IL-18-converted B6 mice which were susceptible to low doses of the helminth, to a highly resistant state that is more profound than that seen in IL-12-deficient mice (24). In contrast Balb/c mice, which are normally resistant to *Trichuris* via a mechanism dependent on IL-13- and IL-4-mediated expulsion of the nematode, become susceptible after IL-18 treatment. In both cases susceptibility correlates with low IL-13 (not IL-4) levels. The authors conclude that IL-18 plays a key role in gastrointestinal nematode infections via downregulation of IL-13 (24). The authors also were able to visualize very early production of IL-18 after infection in the intestine by macrophages and dendritic cells, which correlates with the susceptible phenotype (24).

Address correspondence to Susan Swain, Trudeau Institute, Inc., 100 Algonquin Ave., P.O. Box 59, Saranac Lake, NY 12983. Phone: 518-891-3080; Fax: 518-891-5126; E-mail: sswain@northnet.org

## IL-18: Pleiotropic Cytokine for Protection against Intracellular parasites

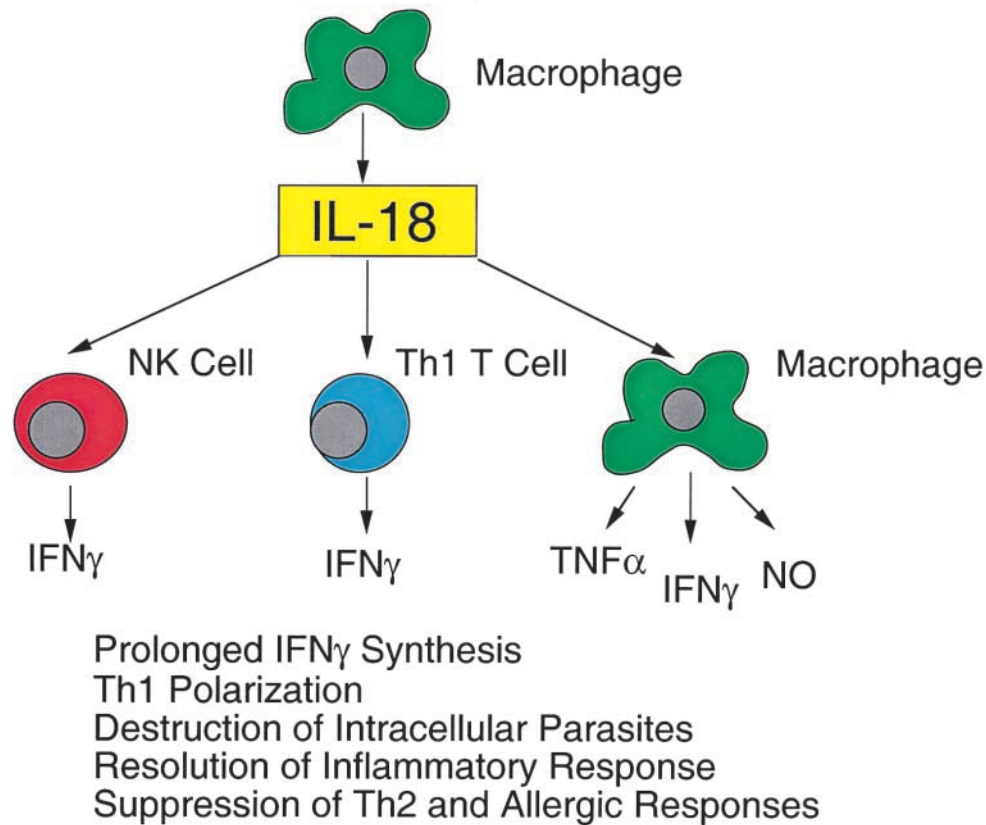


Figure 1.

The reason that IL-18 induces Th2 cytokines under some circumstances remains unexplained, but these new studies tip the balance in favor of a predominantly pro-Th1 action of IL-18. A cartoon summarizing the action of IL-18 in protection against infectious disease, derived from the recent and earlier studies, is in Fig. 1.

*Perspectives and Questions* One of the most novel activities of IL-18 is its ability to induce Th1 effectors to produce IFN- $\gamma$  in the absence of TCR signaling (21). IL-18 and IL-2 alone can induce prolonged IFN- $\gamma$  protein synthesis and, together with TCR triggering, there is a marked synergy resulting in high levels of IFN- $\gamma$  secreted for at least 5 d (21). This is in marked contrast to the effects of TCR triggering alone which results in only transient cytokine synthesis. The prolonged presence of IFN- $\gamma$  at sites of inflammation is liable to result in very dramatic biological effects both in the effector phase of the response but also in its subsequent downregulation (32, 33). Thus prolonged IFN- $\gamma$  production could provide a source of IFN- $\gamma$  that would be available late in the immune response to help downregulate excessive CD4 T cell expansion.

Finally, as IL-18 shares a common signaling pathway with IL-1 $\beta$  and other Toll receptor interacting components, IL-1 $\beta$  and agents signaling via toll receptors might be expected to induce prolonged rather than transient IFN- $\gamma$  production. It would also be of interest to deter-

mine if the other cytokines produced in response to IL-18 also show prolonged induction.

The regulation of IL-18 production also deserves further exploration. Some cells have been reported to make IL-18 constitutively (15), but certain infections apparently lead to upregulation of production. The consensus seems to be that macrophages and related cells are the major producers, but what cells make IL-18 in different circumstances and what conditions favor IL-18 production, processing, and subsequent blocking by IL-18-binding protein deserve further study.

*Conclusions.* IL-18 is emerging as a powerful, pleiotropic cytokine involved in determining the polarization of T cell responses and whether the responses to infectious organisms are protective or not. IL-18 is made by macrophages, dendritic cells, perhaps lymphocytes, and by nonimmune cells; and like IL-1, its actions are regulated by the requirement for proteinase cleavage and by blocking proteins, as well as by the expression of its receptor by the variety of potential targets. It has potent actions on macrophages, inducing TNF production and its consequences as well as NO production, on T cells and B cells inducing IFN- $\gamma$  especially in synergy with other cytokine inducers including IL-12 and Ag/APC. We are sure to hear much more about IL-18 as a critical multipotent inducer of innate and acquired immune responses.

## References

1. Okamura, H., H. Tsutsui, T. Komatsu, M. Yutsudo, A. Hakura, T. Tanimoto, K. Torigoe, T. Okura, Y. Nukada, K. Hattori, et al. 1995. Cloning of a new cytokine that induces IFN- $\gamma$  production by T cells. *Nature*. 378:88–91.
2. Bazan, J.F., J.C. Timans, and R.A. Kastelein. 1996. A newly defined interleukin-1? *Nature*. 379:591.
3. Hoshino, K., H. Tsutsui, T. Kawai, K. Takeda, K. Nakanishi, Y. Takeda, and S. Akira. 1999. Generation of IL-18 receptor-deficient mice: evidence for IL-1 receptor-related protein as an essential IL-18 binding receptor. *J. Immunol.* 162:5041–5044.
4. Born, T.L., E. Thomassen, T.A. Bird, and J.E. Sims. 1998. Cloning of a novel receptor subunit, ACPL, required for interleukin-18 signaling. *J. Biol. Chem.* 273:29445–29450.
5. Adachi, O., T. Kawai, K. Takeda, M. Matsumoto, H. Tsutsui, M. Sakagami, K. Nakanishi, and S. Akira. 1998. Targeted disruption of the MyD88 gene results in loss of IL-1- and IL-18-mediated function. *Immunity*. 9:143–150.
6. Gu, Y., K. Kuida, H. Tsutsui, G. Ku, K. Hsiao, M.A. Fieffing, N. Hayashi, K. Higashino, H. Okainura, K. Nakanishi, et al. 1997. Activation of interferon- $\gamma$  inducing factor mediated by interleukin-1 converting enzyme. *Science*. 275:206–209.
7. Stoll, S., H. Jonuleit, E. Schmitt, G. Muller, H. Yarnauchi, M. Kurimoto, J. Knop, and A.H. Enk. 1998. Production of functional IL-18 by different subtypes of murine and human dendritic cells (DC): DC-derived IL-18 enhances IL-independent Th1 development. *Eur. J. Immunol.* 28:3231–3239.
8. Matsui, K., T. Yoshimoto, H. Tsutsui, Y. Hyodo, N. Hayashi, K. Hiroishi, N. Kawada, H. Okamura, K. Nakanishi, and K. Higashino. 1997. *Propionibacterium acnes* treatment diminishes CD4<sup>+</sup> NK1.1<sup>+</sup> T cells but induces type I T cells in the liver by induction of IL-12 and IL-18 production from Kupffer cells. *J. Immunol.* 159:97–106.
9. Prinz, M., and U.K. Hanisch. 1999. Murine microglial produce and respond to interleukin-18. *J. Neurochem.* 72:2215–2218.
10. Takeuchi, M., Y. Nishizaki, O. Sano, T. Ohta, M. Ikeda, and M. Kurimoto. 1997. Immunohistochemical and immunoelectron microscopic detection of interferon- $\gamma$ -inducing factor (interleukin-18) in mouse intestinal epithelial cells. *Cell Tissue Res.* 289:499–507.
11. Stoll, S., G. Muller, M. Kurimoto, J. Saloga, T. Tanimoto, H. Yamauchi, H. Okamura, J. Knop, and A.H. Enk. 1997. Production of IL-18 (IFN- $\gamma$ -inducing factor) messenger RNA and functional protein by murine keratinocytes. *J. Immunol.* 159:298–302.
12. Udagawa, N., N.J. Horwood, J. Elliott, A. Mackay, J. Owens, H. Okamura, M. Kurimoto, T.J. Chambers, T.J. Martin, and M.T. Gillespie. 1997. Interleukin-18 (interferon- $\gamma$ -inducing factor) is produced by osteoblasts and acts via granulocyte/macrophage colony-stimulating factor and not via interferon- $\gamma$  to inhibit osteoclast formation. *J. Exp. Med.* 185:1005–1012.
13. Yankayalapati, R., B. Wize, S.E. Weis, B. Samten, W.M. Girard, and P.F. Bames. 2000. Production of interleukin-18 in human tuberculosis. *J. Infect. Dis.* 182:234–239.
14. Pirhonen, I., T. Sareneve, M. Kurimoto, I. Julkunen, and S. Matikainen. 1999. Virus infection activates IL-1 $\beta$  and IL-18 production in human macrophages by a caspase-1-dependent pathway. *J. Immunol.* 162:7322–7329.
15. Okamura, H., H. Tsutsui, S. Kashiwamura, T. Yoshimoto, and K. Nakanishi. 1998. Interleukin-18: a novel cytokine that augments both innate and acquired immunity. *Adv. Immunol.* 70:281–312.
16. Yang, Y., and B. Moss. 1999. IL-18 binding and inhibition of interferon  $\gamma$  induction by human poxvirus-encoded proteins. *Proc. Natl. Acad. Sci. USA.* 96:11537–11542.
17. Smith, V.P., N.A. Bryant, and A. Alami. 2000. Ectromelia, vaccinia and cowpox viruses encode secreted interleukin-18-binding proteins. *J. Gen. Virol.* 5:1223–1230.
18. Dao, T., W.Z. Mehal, and I.N. Crispe. 1998. IL-18 augments perforin-dependent cytotoxicity of liver NK-T cells. *J. Immunol.* 161:2217–2222.
19. Kanakaraj, P., K. Ngo, Y. Wu, A. Angulo, P. Ghazal, C.A. Harris, J.J. Siekierka, P.A. Peterson, and L.W. Fung. 1999. Defective interleukin (IL)-18-mediated natural killer and T helper cell type I responses in IL-1 receptor-associated kinase (IRAK)-deficient mice. *J. Exp. Med.* 189:1129–1138.
20. Yoshimoto, T., H. Okamura, Y. Tagawa, Y. Iwakura, and K. Nakanishi. 1997. Interleukin 18 together with interleukin 12 inhibits IgE production by induction of interferon- $\gamma$  production from activated B cells. *Proc. Natl. Acad. Sci. USA.* 94:3948–3953.
21. Yang, J., H. Zhu, T.L. Murphy, W. Ouyang, and K.M. Murphy. 2001. IL-18-stimulated GADD45b required in cytokine-induced, but not TcR-induced IFN- $\gamma$  production. *Nat. Immunol.* 2:157–164.
22. Hoshino T., R.H. Wiltrot, and H.A. Young 1999. IL-18 is a potent co-inducer of IL-13 in NK and T cells: a new potential role for IL-18 in modulating the immune response. *J. Immunol.* 162:5070–5077.
23. Neighbors, M., X. Xu, F.J. Barrat, S.R. Ruuls, T. Churakova, R. Debets, J.F. Bazan, R.A. Kastelein, J.S. Abrams, and A. O'Garra. 2001. A critical role for IL-18 in primary and memory responses to *Listeria monocytogenes* that extends beyond its effects on interferon  $\gamma$  production. *J. Exp. Med.* 194:343–354.
24. Helmbly, H., K. Takeda, S. Akira, and R.K. Grencis. 2001. Interleukin-18 promotes the development of chronic gastrointestinal helminth infection by downregulating IL-13. *J. Exp. Med.* 194:355–364.
25. Kobayashi, K., M. Kai, M. Gidoh, N. Nakata, M. Endoh, R.P. Singh, T. Kasaina, and H. Saito. 1998. The possible role of interleukin (IL)-12 and interferon- $\gamma$ -inducing factor/IL-18 in protection against experimental *Mycobacterium leprae* infection in mice. *Clin. Immunol. Immunopathol.* 88:223–231.
26. Garcia, V.E., K. Uyemura, P.A. Sieling, M.T. Ochoa, C.T. Morita, H. Okamura, M. Kurimoto, T.H. Rea, and R.L. Modlin. 1999. IL-18 promotes type I cytokine production from NK cells and T cells in human intracellular infection. *J. Immunol.* 162:6114–6121.
27. Sansonetti, P.J., A. Phalipon, J. Arondel, K. Thirumalai, S. Banerjee, S. Akira, K. Takeda, and A. Zychlinsky. 2000. Caspase-1 activation of IL-1 $\beta$  and IL-18 are essential for *Shigella flexneri*-induced inflammation. *Immunity*. 12:581–590.
28. Wei, X.Q., B.P. Leung, W. Niedbala, D. Piedrafita, G.J. Feng, M. Sweet, L. Dobbie, A.J. Smith, and F.Y. Liew. 1999. Altered immune responses and susceptibility to *Leishmania major* and *Staphylococcus aureus* infection in IL-18-deficient mice. *J. Immunol.* 163:2821–2828.

29. Kawakami, K., Y. Koguchi, M.H. Qureshi, A. Miyazato, S. Yara, Y. Kinjo, Y. Iwakura, K. Takeda, S. Akira, M. Kurimoto, and A. Saito. 2000. IL-18 contributes to host resistance against infection with *Cryptococcus neoformans* in mice with defective IL-12 synthesis through induction of IFN- $\gamma$  production by NK cells. *J. Immunol.* 165:941–947.
30. Yoshimoto, T., H. Mizutani, H. Tsutsui, N. Noben-Trauth, K. Yamanaka, M. Tanaka, S. Izumi, H. Okainura, W.E. Paul, and K. Nakanishi. 2000. IL-18 induction of IgE: dependence on CD4<sup>+</sup> T cells, IL-4 and STAT6. *Nat. Immunol.* 1:132–137.
31. Nakanishi, K., T. Yoshimoto, H. Tsutsui, and H. Okamura. 2001. Interleukin-18 regulates both Th1 and Th2 responses. *Annu. Rev. Immunol.* 19:423–474.
32. Cauley, L.S., E. Miller, M. Yen, and S.L. Swain. 2000. Superantigen-induced CD4 T cell tolerance mediated by myeloid cells and IFN- $\gamma$ . *J. Immunol.* 165:6056–6066.
33. Dalton, D.K., L. Haynes, C. Chu, S.L. Swain, and S. Wittmer. 2000. Interferon  $\gamma$  eliminates responding CD4 T cells during *Mycobacterial* infection by inducing apoptosis of activated CD4 T cells. *J. Exp. Med.* 192:117–212.