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# Interleukin-12 Suppresses Immunoglobulin E Production but Enhances Immunoglobulin G4 Production by Human Peripheral Blood Mononuclear Cells

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**The effect of interleukin-12 (IL-12) on human immunoglobulin G4 (IgG4) and IgE production was examined with cells derived from filarial patients and European controls. IL-12 inhibited IgE release but enhanced IgG4 production in cultures of peripheral blood mononuclear cells stimulated with anti-CD2 plus IL-2. When purified T- and B-cell cocultures were examined, IL-12 again markedly enhanced IgG4, whereas IgE production was no longer inhibited.**

Filarial infections in humans are characteristically associated with Th2 profiles (18). Active infection marked by microfilaremiae in peripheral blood drives strong immunoglobulin G4 (IgG4) responses that dominate the antibody repertoire mounted to filarial parasites (14, 19). Anti-filarial IgE, however, remains relatively low, indicating that active infection with filarial parasites results in the differential regulation of IgG4 and IgE in vivo. This is an interesting observation, since both IgG4 and IgE are thought to be regulated by interleukin-4 (IL-4) and IL-13 (1, 7, 20). Uncoupling of the IgG4 and IgE responses has been described for mice in studies in which IgG1 (murine equivalent of IgG4) is still produced in IL-4 knockout mice in the total absence of IgE (13). A number of studies of regulation of human IgG4 and IgE have reported that several compounds, such as disodium cromoglycate or nerve growth factor, can regulate these isotypes differentially (9, 10).

In search of a mechanism that could lead to the uncoupling of IgG4 and IgE in filarial infections, we have chosen to investigate the effect of IL-12 on these isotypes for the following reasons. In a mouse model of filariasis, it was found that the implanting of microfilariae (mf) resulted in a strong IgG1 production but no IgE. The mf elicited a marked gamma interferon (IFN-γ) response, which in turn might be due to the ability of mf to induce IL-12 (16). Indeed, endogenous IL-12 is produced upon stimulation of peripheral blood mononuclear cells (PBMCs) with filarial parasite extracts, and it has been shown that IL-12 decreases IgE production triggered by parasite antigens in vitro (11). Many studies have addressed the question of how IgE responses are regulated, with obvious implications for therapy of atopic diseases mediated by this isotype. Less attention has been given to IgG4, an isotype which is thought to act as a blocking antibody and which, because of its inability to fix complement, might curtail immunogenic inflammation (24, 25). Accordingly, in filariasis, high IgG4 responses, in the face of relatively low IgE, are associated with the asymptomatic form of the infection in microfilaremics. In the present study, we have determined the effect of IL-12 on IgE and IgG4 production by cells from filarial subjects and European controls. We found that IL-12 inhibits IgE but augments IgG4 production.

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Blood samples from six filarial subjects residing in an area endemic for Brugian filariasis in Central Sulawesi, Indonesia (three positive for circulating mf as detected by filtration of 1 ml of a blood sample taken at night and three amicrofilaremics but with elevated anti-filarial IgG4, an indicator of active infection [5, 14]), and seven European control donors were used in this study. PBMCs were isolated, cryopreserved in Djakarta, Indonesia, as described before (26), and transported to Leiden University, where the study took place. Cells from European control donors were also cryopreserved before use.

For in vitro antibody production, cryopreserved PBMCs were thawed and cultured at a concentration of $5 \times 10^4$ cells/ml (100 μl) in 96-well round-bottom plates (Nunc, Roskilde, Denmark) at a final volume of 0.2 ml of culture medium as described before (22). PBMCs were stimulated with monoclonal antibodies against CD2 (6G4 and B2H4) and recombinant human IL-2 (CLB, Amsterdam, The Netherlands) in the presence or absence of recombinant IL-12 (0.1 to 2.5 ng/ml). In some experiments, neutralizing antibodies to IFN-γ were included (21). All experiments were carried out with replicates of six wells. After 12 days of incubation, supernatants were collected and kept frozen at −20°C until assayed. Cells were counted with a cell counter (Analys Instruments).

Filaria-specific T-cell lines were generated to *Brugia malayi* adult worm extract (14) by incubation of $2 \times 10^6$ PBMCs in 2 ml of culture medium containing 10% human serum and 12.5 μg of *B. malayi* antigen per ml for 7 days in 24-well flat-bottom plates, after which medium was refreshed and IL-2 (50 U/ml) was added. Cultures were refreshed regularly for an additional 2 weeks before T-cell lines were used for experiments. Five T-cell lines designated E10, E56, E68, RN003, and TCL3 were used in this study. Four of these lines produced high levels of IL-4 (100 to 1,200 pg/ml) but no detectable IFN-γ when stimulated with anti-CD2 plus IL-2.

To measure in vitro antibody production by T- and B-cell cocultures, T cells (originating from T-cell lines) were incubated with $CD19^+$ cells (purity of >99%; sorted on a Becton Dickinson FACStar) obtained from a European control donor. These cultures containing 20,000 T cells and 300 B cells (ratios optimal for antibody production) were stimulated with anti-CD2 plus IL-2 (1:4,000, 50 U/ml) for 12 days before supernatants were harvested. T-cell–B-cell cocultures were carried out in replicates of six.

For the determination of Igs, rabbit anti-human IgE (Dako) or mouse anti-human IgG4 (HP6023; Sigma) was used at 1

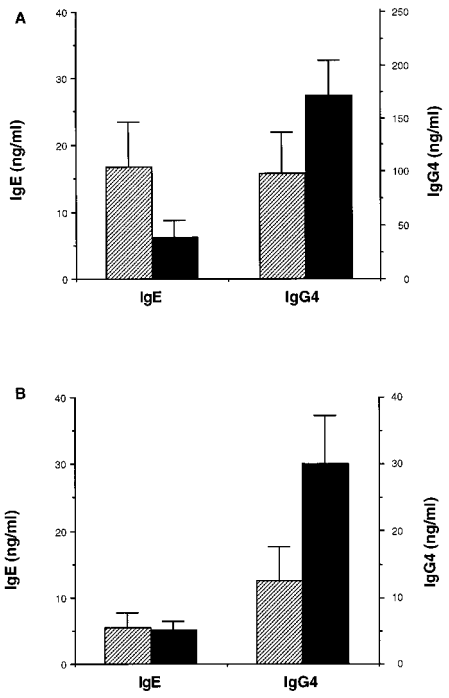


FIG. 1. Effect of IL-12 on IgE and IgG4 secretion by PBMCs derived from filarial patients (A [$n = 6$]) and European controls (B [$n = 7$]). PBMCs were stimulated with anti-CD2 plus IL-2 in the absence (hatched bars) or presence (solid bars) of 0.1 ng of IL-12 per ml. Mean values of IgG4 and IgE (nanograms per milliliter) in culture supernatants are depicted.

μg/ml to coat Maxisorp plates (Nunc) in 0.1 M carbonate buffer (pH 9.6). Rabbit anti-human IgG (Dako) was used at 1 μg/ml to coat Polysorp plates (Nunc). Culture supernatants were incubated overnight at 4°C. All antibody incubations were performed in 0.1 M Tris buffer (pH 7.4) containing 0.05% Tween 20 and 0.5% bovine serum albumin. For detection of IgE, biotinylated goat anti-human IgE (1:1,000; Vector) was added to the appropriate plates and incubated overnight. The plates were then washed and incubated for 4 h at room temperature with alkaline phosphatase-conjugated streptavidin (1:3,000; Boehringer Mannheim). For IgG and IgG4 determinations, alkaline phosphatase-conjugated goat anti-human IgG [$F(ab')_2$] (1:4,000; Jackson) was added, and the mixture was incubated for 4 h at room temperature. After washing, the plates were stained with *p*-nitrophenyl phosphate (Sigma). Reactions were stopped with NaOH (3 M), and $A_{405}$ was read. On each plate, a standard serum with known concentrations of IgE (Behringwerke AG, Marburg, Germany), IgG4, or IgG (CLB, Amsterdam, The Netherlands) was included to allow calculation of Ig concentrations in supernatants. The specificity of the IgG4 enzyme-linked immunosorbent assay (ELISA) was validated by assaying serum from a patient with a genetic deficiency in the gamma-4 chain. This serum contained normal IgG levels and was negative in the IgG4 ELISA.

Supernatants for cytokine measurement were taken on day 4 after stimulation. IFN-γ concentrations were quantified in supernatants by use of an ELISA with a lower detection limit of 3 U/ml (27). IL-4 and IL-13 concentrations were determined with a commercial kit (CLB) with a threshold of 3 pg/ml, according to the manufacturer's recommendations.

Differences in antibody isotype production and cellular responses were analyzed by Student's paired *t* test.

For measurable IgE production, it was necessary to culture PBMCs stimulated with anti-CD2 and IL-2 at low cell densities of 5,000 cells per well as reported before (23); under the same conditions, IgG4 could also be measured in the culture supernatants. Suboptimal concentrations of anti-CD2 plus IL-2 (1:8,000 antibody dilution and 12.5 U of IL-2 per ml) were used to study the modulatory role of IL-12 in IgG4 and IgE release. The effect of IL-12 was investigated with cells from six filarial subjects and seven European controls. As shown in Fig. 1, PBMCs from filarial subjects produced both IgG4 and IgE. In contrast, PBMCs from European donors produced low levels of IgG4 and virtually no IgE. It should be noted that levels of cell proliferation in response to anti-CD2 plus IL-2 were equivalent in both groups, and IL-12 enhanced proliferation in all subjects to the same extent (not shown). IgE production by filarial patients, however, was significantly inhibited by IL-12 ($P = 0.03$ [Fig. 1A]), confirming previous reports demonstrating that IgE release in PBMCs is suppressed by IL-12 (2, 11, 12). It was not possible to determine whether IL-12 had a similar effect on IgE production by European controls, because PBMCs from these donors produced very low levels of IgE (just above background) (Fig. 1B). To simplify culture conditions and to determine whether IL-12 exerts its effect by a direct interaction with T and B cells, we used Th2 cells (originating from cell lines generated to filarial worm extracts) and cultured them with $CD19^+$ B cells purified from a European control donor. Interestingly, IgE production, which was considerable in these cultures, could not be inhibited by IL-12 (Fig. 2). The latter confirms reports by King et al., who were unable to inhibit human IgE production by IL-12 in anti-CD40 plus IL-4-activated purified B cells, thus suggesting that the observed suppressive effect is indirect (11). IgG4 secretion was enhanced in both European controls and filarial subjects (Fig. 1) and in T-cell–B-cell cocultures (Fig. 2). This increase was statistically significant in T-cell–B-cell cocultures ($P < 0.05$) and for European controls ($P = 0.05$). In filarial subjects, the already high IgG4 production was further enhanced by IL-12, but this effect did not reach statistical significance. One other study has examined the effect of IL-12 on human IgG4 and IgE (2). In that study, IgE was suppressed by IL-12, and the effect of this cytokine on IgG4 was tested in three experiments, in two of which there was a decrease in IgG4 production and in one of which there was an increase in IgG4 production. The data were expressed as optical densities, making the interpretation of the magnitude of an effect difficult. In our study, total IgG was not significantly affected by IL-12 in PBMCs (decreases from 2,399 to 2,048 ng/ml in filarial subjects and from 3,714 to 2,678 ng/ml in European controls) or T-cell–B-cell cocul-

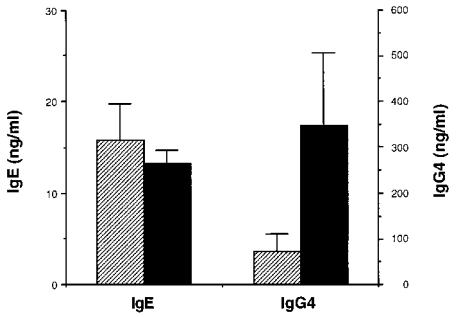


FIG. 2. Effect of IL-12 on IgG4 and IgE secretion by T- and B-cell cocultures. T cells were cocultured with B cells and stimulated with anti-CD2 plus IL-2 in the absence (hatched bars) or presence (solid bars) of 2.5 ng of IL-12 per ml. Mean values of IgG4 (measurable in all five culture supernatants) and IgE (measurable in three of the culture supernatants) are depicted.

TABLE 1. Effect of neutralizing antibodies to IFN-γ on IL-12-induced IgG4 enhancement[a]

| T-cell line | Effect on IgG4 enhancement (ng/ml)[b] | | | |
|---|---|---|---|---|
| | Control | IL-12 | MD-1[c] | IL-12 and MD-1 |
| TCL3 | 99.2 | 762.3[d] | 62.4 | 1,490.8[e] |
| RN003 | 205.9 | 706.8[d] | 75.0 | 482.8[e] |

[a] Cocultures of T and B cells were stimulated with anti-CD2 plus IL-2 (1:4,000, 50 U/ml) and tested for the effect of IL-12 (2.5 ng/ml).
[b] Data represent geometric means of IgG4.
[c] Neutralizing antibodies to IFN-γ (MD-1) were used at a concentration of 2.5 μg/ml.
[d] Value is significantly different from that of the control ($P < 0.05$).
[e] Value is significantly different from that of the control ($P < 0.05$) but not from that of IL-12.

tures (increase from 8,183 to 10,762 ng/ml), indicating that IgG4 is enhanced specifically. Because IgG4 constitutes a small fraction of the total IgG, its increase does not substantially change the total IgG levels, indicating that the IgG4 effect was specific. Indeed, isotypes of IgG1, IgG2, or IgG3 were not enhanced by IL-12 (unpublished data). Consistent with our findings, IL-12 has been reported not to affect IgG secretion by B cells in human PBMCs (12). However, IL-12 enhanced IgG production in one study which utilized purified B cells alone stimulated directly with IL-2 and *Staphylococcus aureus* Cowan strain I (8). This discrepancy might be attributed either to the nature of the stimulus or to the cell culture system used. Several studies have addressed the effect of IL-12 on antibody isotypes in the mouse. Unfortunately the data from the murine models are inconsistent: in vivo administration of IL-12 has been shown to (i) increase protein antigen-stimulated IgG responses (3), (ii) decrease IgG triggered by goat anti-mouse-delta (17), and (iii) increase IgG2b and IgG2c antibody responses to alloantigens and decrease the IgG1 antibody response (4). The ability of IL-12 to inhibit IgE production in PBMC cultures has been shown not to be mediated solely by IFN-γ (7, 12). In our hands, the effects of neutralizing antibodies to IFN-γ were inconsistent in the PBMC cultures. However, in T- and B-cell cocultures, it was clear that the enhancement of IgG4 by IL-12 was not mediated by IFN-γ, since no IFN-γ was detected in culture supernatants, and antibodies to IFN-γ that could neutralize up to 15 U of IFN-γ did not affect the IgG4 enhancement (Table 1).

The precise mechanism by which IL-12 augments IgG4 specifically remains unknown. A number of possibilities can be considered. (i) IL-12 expands the already switched B cells, in which case it has to be postulated that it expands $IgG4^+$ B cells only. (ii) IL-12 sensitizes B cells to switch to IgG4. (iii) IL-12 acts on T cells to upregulate soluble or surface-bound molecules, which in turn are involved in inducing a specific switch to IgG4. Preliminary data from T-cell–$IgD^-$ B-cell coculture experiments indicate that IL-12 does not enhance IgG4 production in such cultures, which contradicts point i. Furthermore, we found no evidence that IL-12 significantly changed IL-4 or IL-13 levels in the T-cell–B-cell cocultures in which IgG4 enhancement was observed (Table 2). In PBMC cultures stimulated with anti-CD2 and IL-2, there was no detectable IFN-γ or IL-4 (because of the low cell densities utilized). Upon addition of IL-12, which enhanced IgG4, it was possible to measure IFN-γ (geometric mean of 150 U/ml; $n = 4$), but there was still no IL-4. The possibility that other cytokines or surface molecules might play a role in enhancing a selective switch to IgG4 will be addressed in future studies.

TABLE 2. The effect of IL-12 on cytokine release

| T-cell line[a] | Cytokine release[b] | | | | | |
|---|---|---|---|---|---|---|
| | IL-4 (pg/ml) | | IL-13 (pg/ml) | | IFN-γ (U/ml) | |
| | −IL-12 | +IL-12 | −IL-12 | +IL-12 | −IL-12 | +IL-12 |
| E10 | 370 | 580 | 162 | 64 | <3 | <3 |
| TCL3 | 3,005 | 2,644 | 4,089 | 4,322 | <3 | <3 |
| RN003 | 1,088 | 2,038 | 1,023 | 1,005 | <3 | <3 |
| E68 | 111 | 101 | ND[c] | ND | <3 | 9.6 |

[a] T cells originated from the T-cell lines designated E10, TCL3, RN003, and E68 as indicated.
[b] IL-4, IL-13, and IFN-γ levels in supernatants were determined on day 4 after stimulation. All T- and B-cell cocultures were stimulated with anti-CD2 plus IL-2 (1:4,000, 50 U/ml) in the presence or absence of IL-12 (2.5 ng/ml).
[c] ND, not determined.

In summary, antibodies of the IgG4 subclass are incapable of fixing complement, are functionally monovalent, and have been shown to act as blocking antibodies to IgE (6, 24, 25). This isotype is highly elevated in infections with the nematode parasites *B. malayi* and *Wuchereria bancrofti*, particularly when circulating mf are detectable (14, 15). With respect to mechanisms that may lead to differential regulation of IgG4 and IgE in filariasis, it was noted that (i) parasite extracts when added to PBMCs were capable of inducing IL-12 release and that (ii) mice transplanted with microfilariae exhibited high levels of IFN-γ, raising the possibility that microfilariae induce IL-12 release, which in turn stimulates IFN-γ (11, 16). Following observations that IL-12 can inhibit IgE, it was reasoned that this cytokine might be involved in differential regulation of IgG4 and IgE in filariasis.

In the present study, we have shown that IL-12 can influence IgG4 and IgE differentially. Any IL-12 released upon interaction of mf with the immune system may play a significant role in the uncoupled expression of these two isotypes during active infection in lymphatic filariasis.

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## REFERENCES

1. **Aversa, G., J. Punnonen, B. G. Cocks, R. de Waal Malefyt, G. Vega, S. M. Zurawski, G. Zurawski, and J. E. de Vries.** 1993. An interleukin 4 (IL-4) mutant protein inhibits both IL-4 or IL-13-induced human immunoglobulin G4 (IgG4) and IgE synthesis and B cell proliferation: support for a common component shared by IL-4 and IL-13 receptors. J. Exp. Med. **178:**2213–2218.
2. **Garraud, O., C. Nkenfou, J. E. Bradley, F. B. Perler, and T. B. Nutman.** 1995. Identification of recombinant filarial proteins capable of inducing polyclonal and antigen-specific IgE and IgG4 antibodies. J. Immunol. **155:** 1316–1325.
3. **Germann, T., M. Bongartz, H. Dlugonska, H. Hess, E. Schmitt, L. Kolbe, E. Kolsch, F. J. Podlaski, M. K. Gately, and E. Rude.** 1995. Interleukin-12 profoundly up-regulates the synthesis of antigen-specific complement-fixing IgG2a, IgG2b and IgG3 antibody subclasses in vivo. Eur. J. Immunol. **25:** 823–829.
4. **Gracie, J. A., and J. A. Bradley.** 1996. Interleukin-12 induces interferon-γ-dependent switching of IgG alloantibody subclass. Eur. J. Immunol. **26:**1217–1221.
5. **Haarbrink, M., A. Terhell, K. Abadi, S. van Beers, M. Asri, and M. Yazdanbakhsh.** 1995. IgG4 antibody assay in the detection of filariasis. Lancet **346:**853–854.
6. **Hussain, R., R. W. Poindexter, and E. A. Ottesen.** 1992. Control of allergic reactivity in human filariasis. Predominant localization of blocking antibody to the IgG4 subclass. J. Immunol. **148:**2731–2737.
7. **Jabara, H. H., S. M. Fu, R. S. Geha, and D. Vercelli.** 1990. CD40 and IgE: synergism between anti-CD40 monoclonal antibody and interleukin-4 in the induction of IgE synthesis by highly purified human B cells. J. Exp. Med. **172:**1861.

8. **Jelinek, D. F., and J. K. Braaten.** 1995. Role of IL-12 in human B lymphocyte proliferation and differentiation. J. Immunol. **154:**1606–1613.
9. **Kimata, H., A. Yoshida, C. Ishioka, T. Kusunoki, S. Hosoi, and H. Mikawa.** 1991. Nerve growth factor specifically induces human IgG4 production. Eur. J. Immunol. **21:**137–141.
10. **Kimata, H., A. Yoshida, C. Ishioka, and H. Mikawa.** 1991. Disodium cromoglycate (DSCG) selectively inhibits IgE production and enhances IgG4 production by human B cells in vitro. Clin. Exp. Immunol. **84:**395–399.
11. **King, C. L., J. Hakimi, M. T. Shata, and A. Medhat.** 1995. IL-12 regulation of parasite antigen-driven IgE production in human helminth infections. J. Immunol. **155:**454–461.
12. **Kiniwa, M., M. Gately, U. Gubler, R. Chizzonite, C. Fargeas, and G. Delespesse.** 1992. Recombinant interleukin-12 suppresses the synthesis of immunoglobulin E by interleukin-4 stimulated human lymphocytes. J. Clin. Invest. **90:**262–266.
13. **Kuhn, R., K. Rajewsky, and W. Muller.** 1991. Generation and analysis of interleukin-4 deficient mice. Science **254:**707–710.
14. **Kurniawan, A., M. Yazdanbakhsh, R. van Ree, R. Aalberse, M. E. Selkirk, F. Partono, and R. M. Maizels.** 1993. Differential expression of IgE and IgG4 specific antibody responses in asymptomatic and chronic human filariasis. J. Immunol. **150:**3941–3950.
15. **Kwan-Lim, G.-E., K. P. Forsyth, and R. M. Maizels.** 1990. Filaria-specific IgG4 response correlates with active *Wuchereria bancrofti* infection. J. Immunol. **145:**4298–4305.
16. **Lawrence, R. A., J. E. Allen, J. Osborne, and R. M. Maizels.** 1994. Adult and microfilarial stages of the filarial parasite *Brugia malayi* stimulate contrasting cytokine and Ig isotype responses in BALB/c mice. J. Immunol. **153:**1216–1224.
17. **Morris, S. C., K. B. Madden, J. J. Adamovicz, W. C. Gause, B. R. Hubbard, M. K. Gately, and F. D. Finkelman.** 1994. Effects of IL-12 on in vivo cytokine gene expression and Ig isotype selection. J. Immunol. **152:**1047–1056.
18. **Ottesen, E. A.** 1992. Infection and disease in lymphatic filariasis: an immunological perspective. Parasitology **104:**S71–S79.
19. **Ottesen, E. A., F. Skvaril, S. P. Tripathy, R. W. Poindexter, and R. Hussain.** 1985. Prominence of IgG4 in the IgG antibody response to human filariasis. J. Immunol. **134:**2707–2712.
20. **Punnonen, J., G. Aversa, B. G. Cocks, A. N. J. Mckenzie, S. Menon, G. Zurawski, R. D. Malefyt, and J. E. de Vries.** 1993. Interleukin-13 induces interleukin-4-independent IgG4 and IgE synthesis and CD23 expression by human B-cells. Proc. Natl. Acad. Sci. USA **90:**3730–3734.
21. **van der Meide, P. H., M. Dubbeld, and H. Schellekens.** 1985. Monoclonal antibodies to human immune interferon and their use in a sensitive solid-phase ELISA. J. Immunol. Methods **79:**293–305.
22. **van der Pouw Kraan, C. T. M., H. J. A. M. Rensink, R. Rappuoli, and L. A. Aarden.** 1995. Costimulation of T cells via CD28 inhibits human IgE production: reversal by pertussis toxin. Clin. Exp. Immunol. **99:**473–478.
23. **van der Pouw Kraan, T., C. Van Kooten, R. van Oers, and L. A. Aarden.** 1991. Human transferrin allows efficient IgE production by anti-CD3-stimulated human lymphocytes at low cell densities. Eur. J. Immunol. **21:**385–390.
24. **van der Zee, J. S., P. van Swieten, and R. C. Aalberse.** 1986. Serologic aspects of IgG4 antibodies. II. IgG4 antibodies form small, nonprecipitating immune complexes due to functional monovalency. J. Immunol. **137:**3566.
25. **Xu, Y., R. Oomen, and M. H. Klein.** 1994. Residue at position-331 in the IgG1 and IgG4 CH2 domains contributes to their differential ability to bind and activate complement. J. Biol. Chem. **269:**3469–3474.
26. **Yazdanbakhsh, M., W. A. Paxton, Y. C. M. Kruize, E. Sartono, A. Kurniawan, A. V. Wout, M. E. Selkirk, F. Partono, and R. M. Maizels.** 1993. T cell responsiveness correlates differentially with antibody isotype levels in clinical and asymptomatic filariasis. J. Infect. Dis. **167:**925–931.
27. **Yazdanbakhsh, M., E. Sartono, Y. C. M. Kruize, A. Kurniawan, T. van der Pouw-Kraan, P. H. van der Meide, M. E. Selkirk, F. Partono, R. Q. Hintzen, R. A. W. van Lier, and R. M. Maizels.** 1993. Elevated levels of T cell activation antigen CD27 and increased interleukin-4 production in human lymphatic filariasis. Eur. J. Immunol. **23:**3312–3317.

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