Among vaccine-preventable diseases, measles is the preeminent killer of children worldwide. Infection with measles virus (MV) is associated with prolonged suppression of cell-mediated immune responses, a phenomenon that is thought to underlie the susceptibility to secondary infections that accounts for most measles-related mortality. Interleukin (IL)–12 is critical for the orchestration of cellular immunity. MV specifically ablates IL-12 production by monocyte/macrophages in vitro through binding to CD46, a complement regulatory protein that is an MV receptor. To address the effect of MV on IL-12 responses in vivo, cytokine production was examined in Gambian patients with measles. IL-12 production by peripheral blood monocytes from such patients is markedly suppressed, which provides a unifying mechanism for many of the immunologic abnormalities associated with measles. This suppression is prolonged, with significant, stimulus-specific inhibition of IL-12 production demonstrable months after recovery from acute infection. However, despite this suppression, IL-12 responsiveness remains intact.

Measles remains a devastating disease of children worldwide. This is especially true in sub-Saharan Africa, where a significant proportion of all deaths in children <5 years old is related to measles [1]. Indeed, the annual mortality due to the human immunodeficiency virus (HIV) pandemic has only recently surpassed the estimated 1 million yearly deaths due to measles [1, 2]. Like HIV, measles virus (MV) is profoundly immunosuppressive, with secondary infections accounting for most associated morbidity and mortality [3]. Abnormalities are found in all compartments of the immune system. However, as with HIV, alterations in cell-mediated immunity (CMI) are thought to be of greatest clinical import [4]. Dysregulation of CMI is prolonged: delayed-type hypersensitivity responses are inhibited for weeks to months after resolution of acute MV infection [5]. Despite this, most patients with measles develop successful MV-specific antibody and cytolytic T lymphocyte (CTL) responses [6].

The search for the mechanism of immunosuppression in measles has revealed many possibilities. Phenomena described in vitro and/or ex vivo include suppressed lymphoproliferative responses [7–10], abnormal immune cell apoptosis [11–14], and decreased NK cell and neutrophil activity [15, 16]. Thymic stromal cell infection, with associated thymic cell and/or thymocyte apoptosis, also has been demonstrated in vitro [17] and in murine (SCID/hu-thymus) models [11]. Furthermore, there is in vitro and in vivo evidence of a type 2 polarization in cytokine responses after measles [18].

Interleukin (IL)–12 provides a key link between innate and acquired immunity [19]. IL-12 is critical to the development of CMI, since it is a potent inducer of interferon (IFN)–γ from T and NK cells, is required for the development of Th1 responses and is an enhancer of NK cell cytotoxicity [19]. Monocyte/macrophages and dendritic cells, antigen-presenting cells that are thought to be the principal IL-12–producing cells in vivo, are also prime targets of MV in natural infection [20, 21]; thus, we previously examined the effects of MV on IL-12 production by such antigen-presenting cells in vitro. Of note, the interaction of MV with primary human monocyte/macrophages ablates IL-12 production [22]. In addition, engagement of CD46, a complement regulatory protein that is a cell surface receptor for both wild-type and highly passaged MV [23–25], with antibody or with its natural complement ligand leads to similar inhibition of monocytic production of IL-12 [22]. These findings have found generalization along 2 quite different paths. First, complement receptors other than CD46 regulate IL-12 production: ligands for complement receptor type 3 (CR3) and the C5a receptor inhibit IL-12 production by murine and human monocyte/macrophages [26–29]. Second, along with other important functional
abnormalities [12, 14, 30–32], dendritic cell production of IL-12 also is suppressed by MV [12, 33].

Suppression of IL-12 production by MV provides a plausible underlying mechanism for many of the abnormalities in CMI that are observed in measles. However, all reported data on IL-12 and MV derive from in vitro models of infection. To see whether IL-12 responses are similarly altered in vivo during natural infection, we examined monokine production in Gambian patients with measles.

Subjects and Methods

Patients and control subjects. For monokine studies, 25 patients with acute measles were recruited through the outpatient department of the Medical Research Council (MRC) Laboratories in The Gambia during a yearly measles epidemic (May–July 1999). Patients were examined and enrolled within 7 days of the onset of a generalized maculopapular rash. A clinical diagnosis of measles was made according to World Health Organization criteria [34]. Serologic confirmation by hemagglutination-inhibition assay subsequently was done on plasma, as described elsewhere [35]. All patients either had a titer ≥250 IU/mL or a ≥4-fold increase in titer at follow-up; 1 patient died from respiratory complications before follow-up. Six of the measles patients (24%) had written evidence of previous measles vaccination at age ≥9 months. Sixteen patients (64%) had a history of no vaccination. For 3 patients (12%), vaccination status was unknown. Twenty-seven healthy control subjects also were recruited from a group of infants and children enrolled in ongoing surveillance studies and from a group of adult employees of the MRC Laboratories.

Demographic characteristics of patients and control subjects are shown in table 1. There were no significant differences in age or sex between patients and control subjects. Sixteen of the enrolled measles patients were reexamined 32–54 days (mean, 6.6 weeks) after acute presentation. Nine of these patients were further examined 203–250 days (mean, 32.4 weeks) after initial presentation. Despite the decreasing numbers of patients examined during these follow-up visits, there remained no significant demographic differences between control subjects and the patients studied at these later times. Furthermore, among patients who were characterized, no significant differences were found in nutritional status, as assessed by weight-for-age z scores (calculated using Epi Info 6 software; CDC), between patients (n = 20) and control subjects (n = 15).

Heparinized blood (1–5 mL, depending on age and size) was obtained from each subject. Peripheral blood mononuclear cells (PBMC) were isolated by centrifugation over ficoll-hypaque (Nycomed). A complete blood cell count with differential was determined (Cellanalyzer CA530; Bromma) for most control subjects and for patients with acute disease.

For studies of IL-12 responsiveness, 9 patients (4 boys and 5 girls) with acute measles were similarly enrolled during a measles outbreak in 1997. The patients had a mean age of 5.6 years (range, 0.3–16.0 years).

Flow cytometric analysis of cytokine production. PBMC were cultured at 1.5 × 10^6 cells/mL (1400 μLwell) in RPMI containing 5% human serum, gentamicin (both from Sigma), and l-glutamine (Gibco BRL) in 24-cell plates (Ultra-Low Cluster plates; Costar). After being primed for 2 h with 300 IU/mL of IFN-γ (Pharmingen), PBMC were stimulated with 1 μg/mL of Salmonella typhimurium lipopolysaccharide (LPS; Sigma). Monensin (1 μM, Sigma) was added at the time of LPS stimulation. Twenty-two hours later, the cells were harvested, were washed in staining buffer (PBS with 1% fetal calf serum [Gibco] and 0.05% sodium azide [Malineckrodt]), and were blocked for 30 min with 20 μg/mL of murine IgG1 (Pharmingen) plus 34 μg/mL of human IgG (Dako). After a further wash, cyochrome-conjugated antibodies to CD2, CD3, and CD19 (Pharmingen) were added to PBMC for 30 min. Cells then were washed and fixed with 4% paraformaldehyde in PBS for 10–15 min. After further washing, the cells were stored overnight at 4°C in staining buffer. PBMC then were permeabilized with 0.1% saponin (Sigma) in staining buffer (permeabilization buffer), were washed twice, and were incubated for 30 min with phycoerythrin (PE)–conjugated antibodies to IL-12 p40 (clone 11.5; 1.25 μg/mL) or IL-12 p70 (clone 20C2, 0.7 μg/mL; Pharmingen) in the presence of saponin. Cells then were washed once in permeabilization buffer and were resuspended in staining buffer. All incubations were done on ice. Flow cytometric analysis was done using a FACS Calibur along with CellQuest Software (Becton Dickinson), and 15,000 events were acquired for each condition.

Although CD14 is a useful cell surface marker of monocyctic cells in freshly isolated PBMC, the stimulation conditions used in this study (IFN-γ followed by LPS) lead to marked down-regulation of CD14 expression [36, 37] (C.L.K., unpublished data), which obviates its usefulness as a marker of monocyctic cells under such conditions. To identify the monocyte/macrophage population by flow cytometry after such stimulation, we sequentially gated on this population on the basis of forward- and side-scatter properties (according to extensive experience with stimulated elutriated human monocytes) and excluded nonmonocytic cells within this gate by excluding cells that were positive for the cell surface markers CD3, CD2, and/or CD19 (figure 1A and 1B). Cyochrome- and PE-conjugated irrelevant murine IgG1 antibodies were used to set signal/noise parameters in each patient’s sample. Signal gates were adjusted further for specificity through analysis of the ability of

<table>
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<tr>
<th>Characteristic</th>
<th>Healthy control subjects (n = 27)</th>
<th>Measles patients</th>
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<tr>
<td></td>
<td>Acute (n = 25)</td>
<td>6-week follow-up (n = 16)</td>
</tr>
<tr>
<td>Age, mean years (range)</td>
<td>5.3 (0.3–31.0)</td>
<td>5.4 (0.75–23.0)</td>
</tr>
<tr>
<td>Boys, no. (%)</td>
<td>14 (51.8)</td>
<td>13 (52)</td>
</tr>
<tr>
<td>Girls, no. (%)</td>
<td>13 (48.2)</td>
<td>12 (48)</td>
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excess unconjugated IL-12 antibodies (50 μg/mL), to ablate the IL-12 signal in each patient’s sample (figure 1C and 1D).

Analysis of cytokine secretion. PBMC were cultured at 1.5 × 10^6 cells/mL (700 μL/well) in the above-described media in 48-well plates (Costar). Stimulation was with 0.01% Staphylococcus aureus Cowan strain I (SAC; Calbiochem) or an equivalent volume of medium. After 24 h, cell-free supernatants were harvested and stored at −20°C until analysis. IL-12 p40, IL-10, IL-6, and tumor necrosis factor (TNF-α) were measured by ELISA, using reagents from Phar- ming (sensitivities were 16, 20, 20, and 40 pg/mL, respectively). IL-12 p70 production was measured using the Quantikine HS kit (R&D Systems), with sensitivities of 0.16–1.6 pg/mL in the various assays. IFN-γ production was measured by ELISA, using reagents (Biosource Europe) with a sensitivity of 8 pg/mL. For statistical purposes, all values under the threshold of sensitivity of the ELISAs were assigned a value of 50% of the limit of detection.

Cytotoxic T cell assay and IFN-γ production. CTL activity was determined by a direct assay, as described elsewhere [38]. In brief, PBMC isolated from patients with acute measles were cultured for 3 days at 2 × 10^6 cells/mL (1 mL/well; 24-well plates) in RPMI-1640 medium (Sigma) supplemented with 10% fetal calf serum, L-glutamine, penicillin, streptomycin, and 10% IL-2 (lymphocult-T; Biotest). PBMC were cultured in the presence or absence of 10 ng/mL of recombinant IL-12 p70 (Genetics Institute). For target cells, HLA-matched allogeneic Epstein-Barr virus–transformed B cells were infected for 2 h with vaccinia virus recombinants that expressed the MV fusion protein (or β-galactosidase as a control) at an MOI of 5. After being washed, target cells were cultured for 16 h before being labeled with ^51 chromium (Amersham). A standard 4-h chromium-release assay was subsequently done using an effector:target ratio of 20:1 [38]. The percentage lysis in β-galactosidase–expressing targets was subtracted from that in MV fusion protein-expressing targets to give a value for MV fusion-specific lysis [38]. Just before doing the chromium release assay, we harvested cell-free supernatants of PBMC cultures for analysis of IFN-γ secretion.

Statistical analysis. Comparisons between measles patients and control subjects were analyzed using the Mann-Whitney U test (cytokine production, cell counts, and age) or the χ^2 test (sex). Wilcoxon’s rank sum test was used to compare cytokine results obtained from the same patient at different times (acute measles phase vs. recovery phase) or under different culture conditions (exogenous IL-12 vs. medium control). Spearman’s rank correlation test was used to compare IL-12 production with age.

Results

Prolonged suppression of monocytic production of IL-12 in the wake of measles: flow cytometric analysis. We characterized

Figure 1. Flow cytometric analysis of the stimulated production of interleukin (IL–12) by monocytes present within peripheral blood mononuclear cell populations. A, Monocyte/macrophage gating by forward- and side-scatter properties, after stimulation with interferon (IFN–γ) plus lipopolysaccharide. B, Negative control, using conjugated murine IgG1 antibodies. C, Specificity control, blocking intracellular IL-12 staining with excess unconjugated anti-IL-12 antibodies before the addition of conjugated anti–IL-12 antibodies. D, Specific, intracellular IL-12 staining in the peripheral blood mononuclear cells of a patient with acute measles, showing the percentage of monocytic cells expressing IL-12 p40 after stimulation with IFN–γ plus lipopolysaccharide. CyC, cyochrome; FSC-H, forward scatter; mIgG1, murine IgG1; PE, phycoerythrin; SSC-H, side scatter.
the stimulated production of IL-12 by peripheral blood monocytes in patients with acute measles and in healthy control subjects, using intracytoplasmic staining and flow cytometric techniques. In the absence of stimulation, IL-12 production is essentially undetectable by flow cytometric analysis (data not shown) [39]. Although much of the literature on human IL-12 regulation during infections has focused on SAC-driven IL-12 production (e.g., [40, 41]), the high capacity of SAC to bind immunoglobulins via surface-expressed protein A renders it unsuitable as a stimulus in flow cytometry assays. We thus used IFN-γ and LPS as stimuli in these studies. Of note, the percentage of monocytes that produced IL-12 in response to such stimulation was significantly suppressed in patients with acute measles. The median proportion of monocytes expressing the highly regulated heavy chain of IL-12 (IL-12 p40) after stimulation was 9.8% (range, 1.0%–36.9%) in patients, compared with 20.9% (range, 8.2%–38.9%) in healthy control subjects (P = .0002); for the functional IL-12 heterodimer (IL-12 p70), the relevant proportions were 0.7% (range, 0%–6.8%) and 7.4% (range, 2.6%–9.3%), respectively (P < .0001; figure 2).

There was no significant correlation between IL-12 production and patient age at the time of presentation with acute disease. In addition, there were no significant differences in IL-12 production between males and females among acute patients or control subjects, although female patients tended to have a higher proportion of monocytes expressing IL-12. Last, there were no significant differences in IL-12 production between patients with a documented history of previous measles vaccination and those with a documented history of no such vaccination (data not shown).

This suppression in the IL-12 productive capacity of patients’ monocytes ameliorated with time. Follow-up samples were obtained from 16 of the original 26 patients with measles 6 weeks after presentation with acute measles. Although there was a significant increase in the percentage of monocytes producing IL-12 p40 (P = .01) and IL-12 p70 (P = .0004) at this time, production of both remained significantly suppressed, compared with that in healthy control subjects. The median proportion of monocytes expressing IL-12 p40 was 17.1% (range, 3.4%–30%) in patients, compared with 20.9% (range, 8.2%–38.9%) in healthy control subjects (P = .04); for IL-12 p70 the relevant proportions were 3.0% (range, 0.8%–10.7%) and 7.4% (range, 2.6%–19.3%), respectively (P = .0004; figure 2).

Further follow-up samples were obtainable from 9 patients from the original measles cohort 32 weeks after acute presentation. Although the percentage of IL-12 p40-producing monocytes in patients was comparable to that in healthy control subjects at this time (P = .97), IL-12 p70 remained suppressed in patients, compared with control subjects (median proportions of 4.3% [range, 1.4%–7.1%] and 7.4% [range, 2.6%–19.3%], respectively; P = .008; figure 2). IL-12 productive capacity clearly increases with age in early childhood [42, 43]. Because the patients available for follow-up at 32 weeks tended to be younger than our cohort of control subjects, albeit not significantly, a posthoc comparison of IL-12 production was done for patients and control subjects, using only control subjects within the upper age range of these patients. IL-12 p70 production remained significantly suppressed at reanalysis (P = .0076).

**Figure 2.** Production of interleukin (IL)–12 by monocytes from Gambian patients with measles after stimulation with interferon (IFN)–γ plus lipopolysaccharide. IL-12 production was assessed by flow cytometry in peripheral blood mononuclear cells stimulated for 24 h with IFN-γ plus lipopolysaccharide during acute measles (n = 25), 6 weeks after acute presentation (n = 16), 32 weeks after acute presentation (n = 9), and in healthy age- and sex-matched control subjects (n = 27). Data are shown as box plots representing medians; 10th, 25th, 75th, and 90th percentiles; and outlying values. A. IL-12 p40 production. B. IL-12 p70 production. *P = .0002; †P = .04; ‡P < .0001; §P = .0004; and ¶P = .008, compared with normal control subjects.
secretion of IL-12 was significantly suppressed in PBMC from patients with acute measles (P = .03), whereas IL-6 and TNF-α remained unchanged (figure 4). As with IL-12 secretion, IL-10 secretion returned to levels comparable to those of healthy control subjects by 6 weeks after acute presentation. Production of all 3 cytokines (IL-10, TNF-α, and IL-6) was significantly higher in patients at 6 weeks than during acute disease (P = .03, P = .02, and P = .01, respectively). However, unlike secretion of IL-10, secretion of TNF-α and IL-6 also was significantly increased at 6 weeks, compared with that in healthy control subjects (P = .005 and P = .02, respectively), although production of both of these latter cytokines normalized by week 32 of follow-up (figure 4).

Maintenance of IL-12 responsiveness in measles: CTL responses and IFN-γ production. To examine IL-12 responsiveness during acute measles, PBMC from 9 patients with acute measles were cultured in the presence or absence of recombinant IL-12 p70 before the measurement of CTL activity specific for the MV fusion protein and of ex vivo IFN-γ secretion by such cells. Significant increases in both MV-specific lytic activity (P = .0076) and IFN-γ secretion (P = .0077) were seen after the addition of IL-12 p70 (figure 5).

Discussion

Despite the availability of a live attenuated vaccine, measles, among all vaccine-preventable diseases, remains the preeminent killer of children worldwide. The immunosuppressive nature of measles or, more historically, the idea that measles leaves a propensity for the development of lethal infections in its wake, has been appreciated for at least 150 years [44]. Considerable experimental effort has been expended in trying to understand the alterations in CMI induced by measles, beginning with the demonstration early in the twentieth century that delayed-type hypersensitivity responses to tuberculin are ablated during and after measles [45]. Mid-century, before the development of pharmacologic steroids, the immunosuppressive properties of measles infection were even harnessed for therapeutic purposes [46]. A satisfactory accounting of mechanism(s) has been elusive. It has been shown elsewhere that the interaction of MV with primary human monocytes and dendritic cells leads to profound inhibition of the production of IL-12 [12, 22, 33].

Table 2. Hematologic parameters in Gambian patients with measles and in healthy endemic control subjects.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Measles patients, acute (n = 21)</th>
<th>Healthy control subjects (n = 17)</th>
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<tbody>
<tr>
<td>Monocytes</td>
<td>5.6 × 10^9 (1.2–22.0)a</td>
<td>1.9 × 10^9 (0–4.1)</td>
</tr>
<tr>
<td>Lymphocytes</td>
<td>2.7 × 10^9 (0.9–11.9)b</td>
<td>4.1 × 10^9 (1.4–9.4)</td>
</tr>
<tr>
<td>Neutrophils</td>
<td>3.4 × 10^9 (1.0–11.5)c</td>
<td>1.8 × 10^9 (0.9–9.4)</td>
</tr>
<tr>
<td>Total leukocytes</td>
<td>6.5 × 10^9 (3.9–24.2)</td>
<td>6.9 × 10^9 (2.6–19.6)</td>
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NOTE. Data are median no. of leukocytes per liter (range). Granulocytes other than neutrophils were not assessed.

a P < .0001, compared with healthy control subjects.
b P < .03, compared with healthy control subjects.
c P = .05, compared with healthy control subjects.
Figure 4. Monokine secretion by peripheral blood mononuclear cells (PBMC) in Gambian patients with measles after stimulation with *Staphylococcus aureus* Cowan strain 1 (SAC). Monokine production was determined by ELISA in cell-free supernatants of SAC-stimulated PBMC during acute measles (n = 18–22), 6 weeks after acute presentation (n = 12–13), 32 weeks after acute presentation (n = 7–8), and in healthy age- and sex-matched control subjects (n = 18–21). A, Interleukin (IL)–10 secretion. B, Tumor necrosis factor (TNF)–α secretion. C, IL-6 secretion. *P = .03; †P = .07; ‡P = .02; ¶P = .005; and §P = .01.

Data presented here provide the first evidence that IL-12 production is similarly impaired in patients with naturally acquired measles infection, providing a potentially unifying mechanism for the prolonged abnormalities in cellular immune responses observed during and after measles.

Flow cytometric analysis revealed marked suppression of monocyte production of IL-12 in response to stimulation with IFN-γ and LPS during acute measles. Although production of the IL-12 p40 heavy chain normalized over time, production of the functional IL-12 p70 heterodimer remained suppressed relative to that in control subjects for months after acute presentation. Analysis of monokine production in PBMC supernatants in response to stimulation with SAC also demonstrated marked inhibition of IL-12 production during measles. Occurring in the face of increased monocyte numbers, this suppression of IL-12 secretion showed considerable specificity as TNF-α and IL-6 secretion remained unaffected. In a further mirroring of the cytokine profiles associated with the in vitro infection of monocytes with MV [22], IL-10 secretion also was found to be suppressed acutely. Unlike IFN-γ–plus LPS-driven IL-12 expression, however, SAC-driven IL-12 secretion had normalized by 6 weeks after acute presentation.

What explains the markedly different kinetics of IL-12 suppression revealed by these 2 assays? One possibility is that the percentage of monocytes able to produce IL-12 p70 remains suppressed for months but that a compensatory, long-term increase in monocytic cell numbers normalizes overall IL-12 productive capacity. In fact, the concordant up-regulation of all

Figure 5. Interleukin (IL)–12 responsiveness in Gambian patients with acute measles. The effect of the addition of recombinant IL-12 (10 ng/mL) on measles virus–specific cytotoxic T lymphocyte (CTL) and interferon (IFN)–γ production was assessed in measles patients at the time of acute presentation. Data shown represent individual patients (n = 9). A, Measles virus fusion protein–specific CTL activity (effector :target ratio, 20:1; P = .0076). B, IFN–γ secretion (P = .0077).
measured secreted monokines 6 weeks after acute presentation with measles (normalization of suppressed IL-10 and IL-12, augmentation of initially normal IL-6 and TNF-α) suggests the possibility of a compensatory increase in monocyte numbers at this time. These changes in IL-6 and TNF-α secretion were not sustained, however, which makes this an unlikely explanation for the differing kinetics of IL-12 suppression reported here. A far more likely explanation stems from the fact that the 2 assays differ not just in measurement techniques but in stimulation conditions: SAC stimulation for secretion and IFN-γ plus LPS stimulation for intracellular staining.

Of note, a similar, albeit reversed, disparity between SAC- and LPS-driven IL-12 production has been described elsewhere in HIV-infected subjects. In the case of HIV infection, IL-12 production in response to LPS (with or without IFN-γ) is maintained in the face of suppressed SAC-driven IL-12 production [47]. Although the critical signaling pathways remain poorly understood, there are clear, mechanistic differences between SAC- and LPS-driven IL-12 production [48, 49], including diametrically opposed effects induced by mitogen-activated protein kinase pathway inhibition (S. Keswani and C.L.K., unpublished data). It should also be noted that IFN-γ priming is necessary for maximal IL-12 production, whatever microbial or lymphocyte cell surface–associated stimulus is delivered [19]. The stimulus-specific kinetics of IL-12 suppression reported here thus suggest the likelihood of pathogen-specific susceptibilities in the wake of measles. In this light, long-term inhibition of IFN-γ–driven IL-12 expression may have direct relevance to disease due to Mycobacterium tuberculosis, a pathogen classically associated with measles-induced exacerbations [44]: IL-12 production during mycobacterial infection appears, itself, to depend on the production of IFN-γ [50].

Although the immunosuppression associated with measles is no doubt multifactorial, the specific defect in IL-12 production demonstrated here provides a unifying mechanism for many of the phenomena observed. Specifically, susceptibility to superinfection, ablation of delayed-type hypersensitivity responses, suppression of NK cell activity, and polarization toward type 2 cytokine production all are consonant with a failure of IL-12 activity. The accompanying compromise in IL-10 production may also be relevant to type 2 cytokine polarization, as genetic deletion of IL-10 augments the type 2 polarization effected by IL-12 deprivation in murine models [51]. Defective IL-12 also is consistent with the development of successful MV-specific antibody and CTL responses: CTL and overall antibody responses develop normally in the absence of IL-12, and genetic deletion of IL-12 is compatible with viral clearance [52–55]. However, a lack of IL-12 would not explain the lymphoproliferative defects seen during measles, although a mechanistic understanding of this phenomenon may be close [9, 56, 57].

IL-12 suppression has been reported in infected children. Severe disease due to respiratory syncytial virus or Plasmodium falciparum leads to acute defects in IL-12 production [58, 59]; chronic deficiencies are seen with chronic HIV infection [42]. The prolonged impairment of IL-12 seen with measles demonstrated here is novel and surprising, however: measles is an acute infection marked by limited viral replication after the onset of clinical disease in normal hosts. Although the kinetics of IL-12 inhibition parallel the time course of increased susceptibility to secondary infections and excess delayed mortality seen after measles, they raise mechanistic questions. The cytokine responses reported here mimic those seen after in vitro ligation of CD46 on monocytes [22]. Viral antigens, including the hemagglutinin ligand for CD46 [24, 25], may persist in lymphoid tissue long after acute disease [60], which would allow for long-term, hemagglutinin-mediated IL-12 suppression in the absence of viremia or even viral replication. The mechanism underlying in vivo IL-12 inhibition by measles remains undetermined, however. IL-12 activity is tightly controlled by multiple mechanisms, including cytokines, complement activation products, and endogenous pharmacoactive substances [19], any of which might theoretically be altered chronically by the insult of measles. Infection may also lead to secondary effects on nutritional status or subclinical coinfection with other pathogens. Precedence does exist for a direct, long-term resetting of IL-12 productive capacity. Chougnet et al. [42] have shown that maternal HIV infection leads to persistent inhibition of IL-12 production, even in HIV-uninfected children. Studies are currently underway to address the mechanisms that underlie the prolonged in vivo IL-12 suppression of measles.

Despite a failure of IL-12 production, IL-12 responsiveness is maintained. Reconstitution of IL-12 in vitro significantly enhanced IFN-γ production and MV-specific CTL activity, even in patients who lack detectable baseline CTL responses. We suggest caution while considering the potential therapeutic implications of these latter findings. Down-modulation of IL-12 by MV may not be entirely contrary to the interests of the host. MV-induced dampening of cellular immunity, through inhibition of IL-12 and IFN-α/β [61] production, may serve to limit damage resulting from the clearance of virus from epithelial cells of the gut and lung. Furthermore, postmeasles encephalitis, an autoimmune disease of unclear etiology, occurs in 0.1% of those infected with natural measles. Given the prominent pathogenic role of IL-12 in many organ-specific autoimmune diseases [19], suppression of IL-12 by MV may protect from this complication. There is still much to be learned from this ancient scourge.

Acknowledgments

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