Macrophage Inflammatory Protein–1α (Not T Helper Type 2 Cytokines) Is Associated with Severe Forms of Respiratory Syncytial Virus Bronchiolitis

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It has been suggested that the pathogenesis of respiratory syncytial virus (RSV) infection is related to the development of T helper (Th) type 2 cytokine responses. The presence of Th1 and Th2 cytokines and the chemokines macrophage inflammatory protein (MIP)–1α and monocyte chemotactic protein (MCP)–1 were assessed by ELISA in nasopharyngeal secretions of infants with RSV infection. Infants with mild bronchiolitis had increased Th1 cytokines and reduced Th2 cytokines, compared with infants with upper respiratory tract illness alone. Severe bronchiolitis was characterized by a more balanced Th1-Th2 response that did not differ from that of infants with upper respiratory tract illness alone. In contrast, MIP-1α was markedly increased in infants with severe bronchiolitis. MIP-1α and MCP-1 levels also were inversely related to oxygen saturation (P < .005). Thus, the severity of RSV bronchiolitis appears to be related more to chemokine release than to Th2 cytokine production.

Respiratory syncytial virus (RSV) is the most important cause of bronchiolitis in infancy, but our knowledge of the pathogenesis of RSV bronchiolitis is incomplete. Further investigation of the pathogenic mechanisms of bronchiolitis might be of assistance in the development of safe and effective vaccines against RSV infection and might also help in determining the nature of the link between bronchiolitis and asthma.

It is currently believed that the pathogenesis of human asthma may be related to a predominance of Th helper (Th) type 2 lymphocytes or their products over Th1-like lymphocytes [1]. Th2-like cytokines, such as interleukin (IL)–4, IL-5, and IL-13, are important in stimulating the production of IgE antibody [2], in causing the migration and activation of eosinophils [3], and in activating eosinophils and basophils [4]. Th1-like cytokines, such as interferon (IFN)–γ, oppose the actions of the Th2-like cytokines. Therefore, one proposed pathogenic mechanism in RSV-induced wheezing is that a predominant Th2-like cytokine response in the airway at the time of RSV infection produces pathologic changes similar to those observed in asthma [5–7].

Nevertheless, in a previous study [8], we found that the IFN-γ:IL-4 ratio in respiratory secretions actually was greater in infants with wheezing at the time of RSV infection than in infants with uncomplicated upper respiratory tract illness (URTI) alone. These findings suggest that skewing of Th lymphocyte responses at the time of RSV infection toward overproduction of IFN-γ might underlie the pathogenesis of RSV-induced obstructive airway disease. We undertook the present study, in part, to determine whether other Th2-like cytokines (specifically, IL-5 and IL-13) were involved in the pathogenesis of bronchiolitis. We also wished to determine whether specific cytokines were related to the persistence of eosinophilia in the peripheral blood of infants at the time of bronchiolitis, especially because the persistence of eosinophilia appears to predict which infants with bronchiolitis will go on to have repeated wheezing through school ages [9, 10]. In addition, we evaluated the presence of the chemokines macrophage inflammatory protein (MIP)–1α, monocyte chemotactic protein (MCP)–1, and RANTES in respiratory secretions and their possible role in the pathogenesis of RSV infection.

Patients and Methods

Study population The study population comprised groups of infants and children <24 months old recruited from the well child clinics and Emergency Department (subjects with URTI alone or mild bronchiolitis that did not require hospitalization) or inpatient areas of the Children’s Hospital of Buffalo. These subjects were assigned a diagnosis of URTI alone (absence of crackles or wheezing on auscultation of the chest, oxygen saturation >97% on room air, and normal chest radiograph when obtained), “nonhypoxic bronchiolitis” (defined for the purposes of this study as wheezing on auscultation with oxygen saturation >95% on room air), or “hypoxic bronchiolitis” (wheezing on auscultation and oxygen saturation <95% on room air or, in the absence of wheezing, hyper-
inflation on chest radiograph and oxygen saturation ≥95% on room air).

Hypoxia was assessed at the time that secretion samples were obtained, with subjects breathing ambient air without oxygen supplementation. Subjects with recurrent wheezing were not included nor were those with prior histories of chronic lung disease or congenital heart disease. Another group of 18 infants who were intubated because of acute respiratory failure caused by RSV infection had secretion samples obtained simultaneously from the nasopharynx and also via the endotracheal tube. These samples were analyzed separately to determine the correlation of mediator concentrations in samples obtained from the lower respiratory tract with those obtained from the upper respiratory tract. Finally, 8 asymptomatic infants with no history of recent respiratory disease served as uninfected control subjects.

RSV infection was confirmed in all symptomatic subjects by detection of viral antigen in nasopharyngeal secretions (NPS) samples by commercial direct immunofluorescence assay. All samples were obtained within the first 5 days of respiratory illness and within 24 h after the onset of wheezing. Demographic features of the study subjects are summarized in table 1.

**NPS samples.** Samples of NPS were obtained by passing size 5 French feeding tubes into the nasopharynx and applying gentle suction. Secretions then were rinsed into collecting traps with 3 mL of viral transport medium. After centrifugation to precipitate cells, 1 mL of the supernatant was frozen at −70°C and was saved for subsequent cytokine or chemokine analysis. The other 2 mL was used for viral culture.

**Cytokine and chemokine analysis.** Concentrations of IFN-γ (Genzyme), IL-4 (R&D Systems), IL-5 (Genzyme), and IL-13 (Endogen) were determined by ELISA. The lower limits of detection for these assays were as follows (all in picograms per milliliter): IFN-γ, 15.6; IL-4, 31.2; IL-5, 8; and IL-13, 7. Quantities of MIP-1α and MCP-1 (R&D Systems) and RANTES (Biosource) also were determined by ELISA. The lower limits of detection for these assays were <5 pg/mL for MIP-1α and MCP-1 and <3 pg/mL for RANTES.

Analysis of all cytokines and chemokines was done on single specimens obtained from different groups of RSV-infected subjects. However, sample size limitations prevented simultaneous determinations of both cytokines and chemokines from the same samples. Subjects were recruited for the study of cytokine content until admission of both cytokines and chemokines from the same sample.

We report concentrations of cytokines and chemokines as picograms of the detected cytokine or chemokine per milliliter of the mixture of secretions and transport medium, without correction to the concentration of any substance in the mixture. Previous investigations have demonstrated that quantities of total protein, albumin, and IgA in secretions vary during the course of a respiratory infection [11]. Therefore, although the volume of secretions obtained from different subjects probably was not constant, the correction of measured quantities of cytokines or chemokines to the concentration of these proteins would not provide an accurate correction for the different volumes of secretions obtained.

**Peripheral blood eosinophil counts.** Eosinophil counts were determined at the time the child initially presented for acute RSV-related illness to a patient care area. Total eosinophil counts were determined by multiplying the total white blood cell count by the percentage of eosinophils seen on differential counts.

**Statistical analysis.** Statistical analysis was done with the assistance of the statistical biostatistician with the StatView 5.0.1 program (Abacus Concepts). Differences in demographic factors were compared by Fisher’s exact test. Because cytokine and chemokine data showed skewing from the normal distribution, statistical analyses were completed after logarithmic (base 10) transformation of data, which established a normal distribution. Differences between illness groups were assessed by use of analysis of variance. Post hoc comparisons were made by Scheffé test. Data correlations were determined by using the transformed data values to calculate the coefficient of correlation. Values of zero were converted to 1 before logarithmic transformation for statistical analysis. Data are presented in the figures and tables as the mean ± SE of the log_{10} values of individual cytokines and chemokines or of their ratios. To enable comparisons with other studies, we also provide the geometric mean values after transformation back from the log_{10} value.

**Table 1. Characteristics of study patients with respiratory syncytial virus infection, by illness group.**

<table>
<thead>
<tr>
<th>Study, patient characteristics</th>
<th>URTI alone</th>
<th>Nonhypoxic bronchiolitis</th>
<th>Hypoxic bronchiolitis</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cytokine study</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of subjects</td>
<td>9</td>
<td>27</td>
<td>41</td>
<td></td>
</tr>
<tr>
<td>Age, mean months</td>
<td>5.4</td>
<td>5.0</td>
<td>4.9</td>
<td>NS</td>
</tr>
<tr>
<td>Boys:girls</td>
<td>16:23</td>
<td>16:11</td>
<td>28:13</td>
<td>.02</td>
</tr>
<tr>
<td>Race</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>21 (54)</td>
<td>12 (45)</td>
<td>20 (49)</td>
<td>NS</td>
</tr>
<tr>
<td>Black</td>
<td>16 (41)</td>
<td>10 (37)</td>
<td>14 (34)</td>
<td>NS</td>
</tr>
<tr>
<td>Hispanic</td>
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<td>4 (15)</td>
<td>5 (12)</td>
<td>NS</td>
</tr>
<tr>
<td>Asian</td>
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<td></td>
</tr>
<tr>
<td>No. of subjects</td>
<td>24</td>
<td>34</td>
<td>36</td>
<td></td>
</tr>
<tr>
<td>Age, mean months</td>
<td>5.2</td>
<td>5.1</td>
<td>4.6</td>
<td>NS</td>
</tr>
<tr>
<td>Boys:girls</td>
<td>14:10</td>
<td>19:15</td>
<td>22:14</td>
<td>.11</td>
</tr>
<tr>
<td>Race</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>11 (46)</td>
<td>14 (41)</td>
<td>15 (42)</td>
<td>NS</td>
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<tr>
<td>Black</td>
<td>10 (42)</td>
<td>15 (44)</td>
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<td>NS</td>
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<tr>
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<td>2 (8)</td>
<td>5 (15)</td>
<td>5 (14)</td>
<td>NS</td>
</tr>
<tr>
<td>Asian</td>
<td>1 (4)</td>
<td>0</td>
<td>2 (6)</td>
<td>NS</td>
</tr>
</tbody>
</table>

**NOTE.** Data are no. (%) of children, except where noted. NS, not significant; URTI, upper respiratory tract illness.
Table 2. Cytokine content of nasopharyngeal secretions from infants and children with respiratory syncytial virus infection, by illness group.

<table>
<thead>
<tr>
<th>Cytokine</th>
<th>URTI</th>
<th>Nonhypoxic bronchiolitis</th>
<th>Hypoxic bronchiolitis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. of children</td>
<td>Cytokine level</td>
<td>No. of children</td>
</tr>
<tr>
<td>IFN-γ</td>
<td>36</td>
<td>0.81 ± 0.15</td>
<td>18</td>
</tr>
<tr>
<td>IL-4</td>
<td>36</td>
<td>1.87 ± 0.11</td>
<td>18</td>
</tr>
<tr>
<td>IL-13</td>
<td>15</td>
<td>0.99 ± 0.22</td>
<td>10</td>
</tr>
<tr>
<td>IL-5</td>
<td>9</td>
<td>0.13 ± 0.13</td>
<td>9</td>
</tr>
</tbody>
</table>

NOTE. Cytokine levels are mean log_{10} pg/mL ± SE. IFN, interferon; IL, interleukin; URTI, upper respiratory tract illness.

a vs. URTI group.  
P _p .002
b vs. nonhypoxic bronchiolitis group.  
P _p .048
c vs. URTI group.  
P _p .004
d vs. URTI group.  
P _p .025

4 was reduced in the group with nonhypoxic bronchiolitis (14.5 pg/mL; _P_ = .004). In the group with hypoxic bronchiolitis, the mean IL-4 concentration was 52.7 pg/mL, a value that tended to be higher than that for the nonhypoxic bronchiolitis group (_P_ = .057) but did not differ significantly from that of the group with URTI alone (_P_ = .43).

The geometric mean concentration of IL-13 for the group with URTI alone was 9.8 pg/mL. The mean concentration of IL-13 for the group with nonhypoxic bronchiolitis was slightly lower (3.6 pg/mL) but did not differ significantly from that of the group with URTI alone (_P_ = .24). The mean concentration of IL-13 in the group with hypoxic bronchiolitis was 1.4 pg/mL, which was significantly lower than that of the group with URTI alone (_P_ = .025) but not than that of the group with nonhypoxic bronchiolitis (_P_ = .46).

Geometric mean concentrations of IL-5 in the 3 illness groups were 1.4, 3.4, and 2.0 pg/mL, respectively. No significant differences among these values were detected.

We determined the IFN-γ:IL-4 concentration ratios for each illness group, to determine whether there was a skewing of cytokine responses toward a Th1 or Th2 bias. As shown in figure 1, the log of the IFN-γ:IL-4 ratio was markedly higher for subjects with nonhypoxic bronchiolitis than for those with URTI alone (_P_ < .0001). In contrast, this ratio was lower in subjects with hypoxic bronchiolitis than in those with nonhypoxic bronchiolitis (_P_ = .006). However, the ratio for subjects with hypoxic bronchiolitis did not differ from that for subjects with URTI alone (_P_ = .17). Taken together, these observations indicate that mild bronchiolitis is associated with an increase in the production of IFN-γ relative to that of IL-4, but that more severe forms of bronchiolitis are characterized by a more balanced cytokine response that is similar to that seen in subjects with URTI alone.

Relationship of cytokines to degree of hypoxia. We also determined the relationship of the concentration of each cytokine in NPS to the degree of hypoxia, as determined by simultaneous measurement of oxygen saturation. There was no significant relationship between the concentration of any of the individual cytokines measured or of the ratio of IFN-γ to any of the Th2-like cytokines to the oxygen saturation in a given subject at the time of illness (each _P_ > .11; data not shown).

Ratios of Th1- and Th2-like cytokines and eosinophilia during RSV bronchiolitis. Previous studies demonstrated that infants who maintain eosinophilia during RSV bronchiolitis are more likely to have asthma at ages 6 and 7 years [9, 10]. We wondered whether the pattern of cytokine expression in NPS would be associated with peripheral blood eosinophilia during bronchiolitis. There was a positive correlation (_r_ = .489; _P_ = .01) of IFN-γ:IL-4 ratios in NPS samples with peripheral blood eosinophil counts. This relationship was attributable predominantly to an inverse relationship of quantities of IL-4 with

![Figure 1. Ratio of interferon (IFN-γ) to interleukin (IL-4) in nasopharyngeal secretions of subjects with upper respiratory tract illness (URTI) alone or bronchiolitis at the time of respiratory syncytial virus infection. Horizontal bars, mean of log_{10} of individual IFN-γ:IL-4 ratios.](https://academic.oup.com/jid/article-abstract/184/4/393/808146)
eosinophil counts \((r = -0.522; \ P = 0.006; \text{figure } 2, \text{ bottom})\) rather than to a relationship of IFN-\(\gamma\) concentrations with eosinophil counts \((r = 0.181; \ P = 0.38; \text{figure } 2, \text{ top})\). There was no significant association of ratios of either IFN-\(\gamma\):IL-13 \((r = 0.38; \ P = 0.69)\) or of IFN-\(\gamma\):IL-5 \((r = -0.02; \ P = 0.95)\) with peripheral blood eosinophil counts (data not shown).

**Other analyses of cytokines.** The secretions of the 8 healthy control infants without symptoms of respiratory illness lacked detectable quantities of IFN-\(\gamma\), IL-5, and IL-13 and had a geometric mean concentration of IL-4 of 18 pg/mL. Cytokine concentrations did not differ significantly when analyzed by sex or ethnicity and did not correlate significantly with patient age, either for the overall study population or for individual illness groups (data not shown). In most cases, we could not determine whether the RSV infection represented primary or secondary infection. We assumed that infants \(\leq10\) months old probably were undergoing primary infection. The group means of individual cytokine concentrations did not differ between infants \(\leq10\) months old and older infants (each \(\ P > 0.29\)).

**Association of \(\beta\)-chemokines with RSV disease form.** To determine the role of the \(\beta\)-chemokines MIP-1\(\alpha\), MCP-1, and RANTES in the pathogenesis of RSV disease, we simultaneously determined the presence of these chemokines in NPS samples of subjects with various forms of illness. The data are summarized in table 3. Quantities of MIP-1\(\alpha\) were similar in subjects with URTI alone or with nonhypoxic bronchiolitis (geometric mean, 147 vs. 136 pg/mL; \(P = 0.72\)). However, subjects with hypoxic bronchiolitis had more MIP-1\(\alpha\) in secretions than did subjects in the URTI group (317 vs. 147 pg/mL; \(P = 0.01\)) or in the nonhypoxic bronchiolitis group (317 vs. 133 pg/mL; \(P = 0.0088\); figure 3). Quantities of MCP-1 did not differ significantly among illness groups. The MCP-1 content of NPS samples from subjects with hypoxic bronchiolitis was slightly but not significantly greater than that among subjects with bronchiolitis without hypoxia (64 vs. 39 pg/mL; \(P = 0.09\); figure 3). Quantities of RANTES in NPS samples did not differ between subjects in the URTI group or in the group with nonhypoxic bronchiolitis \((P = 0.56)\) or between the groups with URTI alone or with hypoxic bronchiolitis \((P = 0.68)\). However, concentrations of RANTES were slightly greater in subjects with bronchiolitis and hypoxia than in subjects with bronchiolitis without hypoxia (102 vs. 34 pg/mL; \(P = 0.39\); figure 3).

**Relationship of chemokines to degree of hypoxia.** The findings described above suggest that increased concentrations of MIP-1\(\alpha\) in respiratory secretions might be associated with more severe forms of illness due to RSV. We also determined the correlation between the concentration of various chemokines in secretions and the simultaneous oxygen saturation values. Subjects with nonhypoxic and hypoxic bronchiolitis were included in this analysis.

As shown in figure 4, the quantities of MIP-1\(\alpha\) in secretions were significantly and inversely related to the degree of oxygenation \((r = -0.404; \ P = 0.0005)\). A significant and inverse re-
Table 3. Chemokine content of nasopharyngeal secretions of infants and children with respiratory syncytial virus infection, by illness group.

| Chemokine   | URTI                  | No. of children | Chemokine level |  | No. of children | Chemokine level |  | No. of children | Chemokine level |
|-------------|-----------------------|-----------------|-----------------|  |                |                  |  |                |                  |
| MIP-1α      |                       | 24              | 2.17 ± 0.1      |  | 34              | 2.13 ± 0.1      |  | 36              | 2.50 ± 0.1      |
| MCP-1       |                       | 13              | 1.57 ± 0.17     |  | 23              | 1.59 ± 0.18     |  | 17              | 1.81 ± 0.09     |
| RANTES      |                       | 21              | 2.10 ± 0.17     |  | 32              | 1.53 ± 0.19     |  | 33              | 2.01 ± 0.13     |

NOTE. Chemokine levels are mean log10 pg/mL ± SE. MCP, monocyte chemotactic protein; MIP, major inflammatory protein; URTI, upper respiratory tract illness.

* vs. URTI group and vs. nonhypoxic bronchiolitis group.

Interrelationships of chemokines. Individual concentrations of a given chemokine correlated well with those of the other chemokines: MIP-1α and MCP, \( r = .522 (P = .045) \); MIP-1α and RANTES, \( r = .551 (P = .032) \); and MCP-1 and RANTES, \( r = .655 (P = .007) \).

Other chemokine analyses. Secretions from healthy infants without symptoms of respiratory illness contained no detectable MIP-1α, 12.4 pg/mL MCP-1, and 9.3 pg/mL RANTES. Concentrations of chemokines did not differ significantly when analyzed by sex or ethnicity and did not correlate significantly with patient age either for the overall study population or by individual illness group (data not shown). Chemokine concentrations did not differ between the groups of subjects ≤10 months old (those presumably undergoing primary infection) and older subjects (each \( P > .34 \)).

Correlation of results in upper and lower respiratory tract secretions. Secretion samples were obtained simultaneously from the upper and lower respiratory tracts of 18 infants who were intubated as a result of respiratory failure due to RSV infection. There was a strong correlation between the concentrations of mediators measured in NPS samples with the corresponding concentrations in the lower respiratory tract. The correlation coefficients were as follows: IFN-γ, \( r = .866 \); IL-4, \( r = .957 \); MIP-1α, \( r = .813 \); RANTES, \( r = .684 \); and MCP-1, \( r = .462 \). With the exception of MCP-1 (\( P = .22 \)), all \( P \) values were <.04. No IL-5 or IL-13 was detected in the lower respiratory tract secretions of these intubated infants. Thus, mediator release in the upper respiratory tract usually appeared to accurately reflect that in the lower respiratory tract.

Discussion

Our results indicate that, unlike atopic asthma, the development of bronchiolitis at the time of RSV infection is not associated with a predominance of Th2-like cytokines in the respiratory tract. The development of mild nonhypoxic forms of RSV bronchiolitis appears to be associated with a shift toward an increased production of IFN-γ and reduced Th2-like cytokines, compared with findings in persons with URTI alone. This is similar to the response in the mouse model of RSV bronchiolitis, where IFN-γ is markedly increased and IL-4 and IL-5 are unchanged [12]. On the other hand, the development of severe (hypoxic) bronchiolitis in the present study was associated with a more balanced cytokine response, with an increase in IL-4 relative to the group with nonhypoxic bronchiolitis. However, this relative shift toward Th2 cytokines cannot be assumed to be the cause of more severe disease, since the concentrations of each cytokine were no greater in subjects with hypoxic bronchiolitis than in subjects with URTI alone. In addition, neither the concentrations of Th1 nor Th2 cytokines bore any significant relationship to individual values of oxygen saturation.
Figure 4. Relationship of quantities of macrophage inflammatory protein (MIP)–1α, monocyte chemotactic protein (MCP)–1, and RANTES concentrations in nasopharyngeal secretions to the degree of hypoxia, as determined by pulse oximetry done at the time that samples were obtained from infants with respiratory syncytial virus bronchiolitis. Points represent log₁₀ of chemokine concentrations vs. log₁₀ of oxygen saturation values.

In contrast to the lack of an association of any cytokine with the development of severe forms of illness, the chemokine MIP-1α was found in higher concentrations in secretions of subjects with more severe hypoxic bronchiolitis than in those of subjects in the other illness groups. Measured quantities of MIP-1α also were related directly to the degree of hypoxia. The MCP-1 and RANTES concentrations also tended to be higher in the subject group with hypoxic bronchiolitis than in the group with non-hypoxic bronchiolitis, but the differences were less striking than those for MIP-1α. Furthermore, in a previous study from our institution [13], the chemokine eotaxin was correlated inversely with the degree of hypoxia observed in infants with RSV bronchiolitis. Although further study is indicated, these findings suggest that an enhanced release of MIP-1α and perhaps other chemokines might secondarily augment the inflammatory response and therefore increase the severity of illness in RSV bronchiolitis.

Some support for a pathogenic role for MIP-1α in RSV infection comes from studies in a mouse model. Mice with a targeted gene deletion for MIP-1α have little inflammatory response in the lung after RSV infection [14], which suggests that this chemokine plays an important role in determining the magnitude of the inflammatory response after RSV infection. Our findings suggest that overproduction of MIP-1α might also result in exaggerated inflammatory responses and enhanced severity of disease in humans. MIP-1α induces IgE formation from human B cells [15] and activates eosinophils and basophils [16, 17].

In previous studies, MIP-1α and RANTES were increased in secretions of infants and older children with virus-induced wheezing, compared with healthy control subjects [18]. Other studies found MIP-1α and RANTES in upper and lower respiratory tract secretions of infants with RSV bronchiolitis [19, 20]. In particular, the MIP-1α (but not RANTES) content of lower respiratory secretions was associated with evidence of eosinophil degranulation in the respiratory tract [20]. Because these studies did not include subjects with nonwheezing URTI, it was not possible to determine the relationship of excessive chemokine release to the development of lower respiratory disease. The present study confirms the previous observations and extends them by demonstrating that hypoxic bronchiolitis is associated with the presence of higher concentrations of MIP-1α (and somewhat greater concentrations of MCP-1 and RANTES) in secretions than those found in subjects with milder forms of bronchiolitis or URTI alone. The presence of only negligible amounts of cytokines and chemokines in asymptomatic, presumably uninfected, infants suggests that these mediators are induced in the respiratory tract by RSV infection.

Other studies of cytokine production in RSV infection have appeared. An earlier study from our institution [8] found that healthy uninfected infants have low levels of IL-4 and undetectable quantities of IFN-γ in NPS, whereas IFN-γ predominates over IL-4 in NPS of infants with bronchiolitis. Three other studies evaluated mitogen-stimulated secretion of IFN-γ and IL-4 by peripheral blood lymphocytes obtained from infants with RSV-related URTI alone or lower respiratory tract disease. In one study [21], cells from infants with URTI alone produced predominantly IL-4, whereas cells from infants with bronchiolitis produced predominantly IFN-γ. In another study, IFN-γ production was lower in mononuclear cells from infants with more severe illness [22]. In general, the results of these 2 studies are consistent with our findings that IFN-γ was increased in secretions of subjects with mild bronchiolitis, compared with those of subjects with URTI alone, but was reduced in cases of more severe bronchiolitis. In a third study, the production of IFN-γ by lymphocytes exceeded that of IL-4, regardless of illness severity. None of these studies measured cytokine release in respiratory secretions. In 2 studies, concentrations of IFN-γ exceeded those of IL-4 in the plasma of...
infants with bronchiolitis [6, 23]. The results of these earlier reports, together with those of the present study, collectively fail to show a predominance of Th1- or Th2-like cytokines in severe RSV bronchiolitis and suggest that other factors may be more important in determining the pathogenesis of RSV-induced lower respiratory disease.

A study by Martinez et al. [24] identified 2 forms of virus-induced wheezing in early life. A milder form appears to occur in nonatopic infants, who have smaller or more reactive airways, than in nonwheezing infants of the same age. These infants usually are treated as outpatients for their wheezing episodes and usually do not experience recurrences of wheezing beyond their third birthday. A more severe form of bronchiolitis occurs in infants who are more likely to be hospitalized initially, who are more likely to have wheezing that persists through age 6 years, and who are more likely to show evidence of atopy. Although direct studies are required of this hypothesis, we believe that our nonhypoxic group of infants is similar to the group of infants with milder wheezing illnesses described by Martinez et al. [24], whereas our subjects with greater release of chemokines and with hypoxia constitute the second group (with more severe illness) described by Martinez et al.

It is possible that, in our study, hypoxia caused greater chemokine release rather than enhanced chemokine release, resulting in hypoxia. However, we are unaware of any evidence that suggests that hypoxic conditions can enhance chemokine release from any type of cell.

In summary, our findings suggest that wheezing at the time of RSV infection is not associated with a predominance of either Th1- or Th2-like cytokines in the airway. We suggest that chemokines, MIP-1α in particular, may be more important determinants of the development of severe forms of illness.

References