

Expression of IFN- γ -inducible protein; monocyte chemotactic proteins 1, 3, and 4; and eotaxin in T_H1- and T_H2-mediated lung diseases

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Background: Chemokines are involved in the influx of leukocytes into the airways in inflammatory lung diseases. The differential cell recruitment characteristic of T_H1 versus T_H2 immune responses may be associated with differential chemokine expression.

Objective: We investigated the expression of chemokines; monocyte chemotactic proteins (MCPs) 1, 3, and 4; eotaxin; and IFN- γ -inducible protein 10 (IP-10) in both T_H1- and T_H2-mediated lung diseases.

Methods: By using immunocytochemistry and in situ hybridization, we examined the protein and mRNA expression, respectively, in bronchoalveolar lavage and biopsy samples in subjects with asthma, tuberculosis, sarcoidosis, and chronic bronchitis.

Results: Increased immunoreactivity and mRNA expression of IP-10 and of the MCPs was found in the bronchoalveolar lavage fluid and biopsy specimens of subjects with asthma and tuberculosis compared with that of control subjects ($P < .005$). IP-10, however, was particularly increased in subjects with sarcoidosis ($P < .001$). Eotaxin, on the other hand, was increased only in patients with asthma when compared with control subjects ($P < .005$).

Conclusion: This study demonstrates that MCP-1, MCP-3, and MCP-4 expression is not specifically associated with lung diseases characterized by a particular cytokine profile. In contrast, IP-10 is mostly expressed in T_H1-mediated diseases, and eotaxin expression seems to be specifically associated with lung diseases of a T_H2 cytokine profile. (J Allergy Clin Immunol 2001;107:664-70.)

Key words: Chemokines, eotaxin, monocyte chemotactic proteins, interferon gamma inducible protein, asthma, sarcoidosis, tuberculosis

Abbreviations used

ATS: American Thoracic Society
BAL: Bronchoalveolar lavage
IP-10: IFN- γ -inducible protein 10
MCP: Monocyte chemotactic protein

Chemokines are a family of cytokines involved in the extravasation of leukocytes to the site of inflammation.¹ Chemokines have been subdivided into 4 groups, C-X-C, C-C, C, and C-X₃-C, depending on the number and spacing of conserved cysteines.²⁻⁴ Chemokines are believed to be involved in the pathogenesis of several lung diseases, such as asthma, tuberculosis, sarcoidosis, and chronic bronchitis, by possibly recruiting leukocytes to the inflammatory site. Previous studies have mainly focused on the CXC and CC subfamilies. C-X-C chemokines, such as IL-8, monokine induced by IFN- γ , and IFN- γ -inducible protein 10 (IP-10), are predominantly involved in neutrophil and lymphocyte recruitment, and C-C chemokines, such as eotaxin and the monocyte chemotactic proteins (MCPs), are chemotactic for eosinophils, lymphocytes, and monocytes.¹⁻⁴

Two types of immune responses have been characterized on the basis of the cytokine produced. The T_H1 immune response mainly involves the production of IFN- γ , IL-2, and IL-12, whereas in the T_H2 response IL-4 and IL-5 are the predominant cytokines.⁵⁻⁹ Recently, it has been suggested that the expression of some chemokines could be preferentially associated with a T_H1 or a T_H2 immune response. The differential expression of chemokine receptors may dictate the action of chemokines and their involvement in mechanisms of polarized T_H1- and T_H2-mediated immune responses.¹⁰⁻¹²

To date, the expression of CC chemokines has only been investigated in T_H2-mediated allergic diseases, such as asthma and rhinitis. Studies have reported an increased expression of MCP-1, MCP-3, MCP-4, and eotaxin in bronchial biopsy specimens of patients with asthma and in patients with allergen-induced rhinitis.¹³⁻¹⁸ The expression of these CC chemokines in T_H1-mediated diseases, however, has not been investigated. More-

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over, the role of chemokines in the pathogenesis of lung diseases, such as tuberculosis and sarcoidosis, has not been well studied.

Therefore in the present study we analyzed the expression of CC and CXC chemokines in both T_H1 - and T_H2 -mediated diseases to verify whether differences in the cytokine microenvironment and cellular infiltrate correspond to a different profile of chemokines. We investigated and compared the expression of IP-10, MCP-1, MCP-3, MCP-4, and eotaxin in bronchial biopsy specimens and in bronchoalveolar lavage (BAL) fluid cells obtained from patients with asthma, sarcoidosis, and tuberculosis. Furthermore, the expression of the above chemokines was analyzed in bronchial biopsy specimens of patients with chronic bronchitis, which is considered to be a disease with a nonpolarized cytokine profile.

METHODS

Study subjects

Patients with atopic asthma ($n = 10$), pulmonary tuberculosis ($n = 10$), active sarcoidosis ($n = 10$), and chronic bronchitis ($n = 9$) and normal control subjects ($n = 10$) were entered into the study. Ethical approval for this study was obtained from the Montreal Chest Institute and the Calmette Hospital Ethics Committees. Written informed consent was obtained from all patients and control subjects before undergoing fiberoptic bronchoscopy.

Subjects with bronchial asthma

Diagnosis of asthma was based on American Thoracic Society (ATS) criteria.¹⁹ Asthmatic patients were recruited from the Montreal Chest Institute. All patients had documented airways reversibility and increased airways responsiveness to methacholine ($PC_{20} < 8$ mg/mL). They were all nonsmokers and atopic and had not received immunotherapy in the preceding 12 months. None of the patients had taken either oral or inhaled corticosteroids, and none had any exacerbations during the 12 weeks preceding the study.

Subjects with pulmonary tuberculosis

Patients with pulmonary tuberculosis were recruited from the Montreal Chest Institute after they had been referred for bronchoscopy on the basis of an abnormal chest radiograph and the possibility of a diagnosis of pulmonary tuberculosis. All subjects were nonatopic and had positive skin test responses for purified protein derivative (>10 mm). Active pulmonary tuberculosis was defined as microbiologically confirmed tuberculosis on the basis of positive cultures for *Mycobacterium tuberculosis* (sputum or BAL fluid) by using the Bactec system. Tuberculin testing was performed by using the Mantoux test.²⁰

Subjects with active pulmonary sarcoidosis

Patients with active pulmonary sarcoidosis were recruited from the Department of Pneumoinmunology, Calmette Hospital, Lille, France. The disease was classified as active according to clinical, radiologic, and histologic data.²¹ The criteria used to determine disease activity were as follows: (1) recently developed or increasing cough or dyspnea; (2) and/or systemic symptoms, such as cutaneous lesions, weakness, fever, or arthralgia; (3) and/or increasing opacities on chest x-ray film; and (4) the presence of noncaseating epithelioid cell granulomas seen in a lung or mediastinal lymph node biopsy specimen. None of these patients smoked; had any evidence of an atopic nature, as defined by the absence of clinical history and negative skin prick test responses to common aeroallergens; or had taken corticosteroids during the previous year.

Subjects with chronic bronchitis

Subjects with chronic bronchitis were recruited from the Department of Pneumoinmunology, Calmette Hospital, Lille, France. Chronic bronchitis was also defined according to the criteria of the ATS.¹⁹ Briefly, the patients were given a diagnosis of obstructive chronic bronchitis if they had a history of cough and sputum production on most of the days of the month for at least 3 months a year during the 2 years before the study. In addition, they had a documented lack of β_2 -induced airflow reversibility recorded as a less than 15% change in FEV_1 . All the subjects had a history of cigarette smoking, were nonatopic (ie, had negative skin test responses for common allergen extracts), and had no past history of asthma or allergic rhinitis. All subjects had been free of acute upper respiratory tract infections, and none had received glucocorticoids or antibiotics within the month preceding the study.

Control subjects

The normal control subjects were recruited from the Montreal Chest Hospital and were all nonsmokers, had no evidence of an atopic nature, and had never had any lung disease.

Study design

Fiberoptic bronchoscopy was performed according to ATS guidelines.¹⁹ The procedure was undertaken if the FEV_1 was greater than 60% of predicted value and platelets and clotting parameters were within normal limits. Subjects were premedicated with nebulized salbutamol (2.5 mg) 10 to 15 minutes before the procedure. At least 4 biopsy specimens were obtained per site in each patient. BAL was then performed by gently wedging the tip of a 6-mm fiberoptic bronchoscope into a segmental bronchus of the right middle lobe leading to the site of radiographic lung involvement and instilling 50 mL of normal saline solution into the distal airway and then having the subject aspirate a mean volume of 30 mL. BAL fluid samples were filtered, and cells were collected, concentrated, and resuspended in RPMI-1640 medium at a concentration of 10^6 cells/mL. Cells were then cytocentrifuged onto poly-L-lysine-coated slides, air-dried, and stored at -80°C until further use. For in situ hybridization, cytospin preparations were fixed in 4% paraformaldehyde, dried at 37°C overnight, and stored at -80°C until further use. Biopsy samples were fixed in 4% paraformaldehyde, washed in 15% PBS-sucrose, blocked in OCT medium, and stored at -80°C . Cryostat blocks were stored at -80°C until further use. For immunocytochemistry, the cytospin preparations were briefly fixed in a solution of acetone/methanol (60:40), air-dried, and stored at -80°C , whereas biopsy samples were immediately placed in PBS and within 30 minutes embedded in OCT medium and snap-frozen in isopentane precooled in liquid nitrogen.

Immunohistochemistry

Immunohistochemistry was performed on tissue and BAL preparations with the use of the alkaline phosphatase-antialkaline phosphatase technique, as previously described.²² Briefly, sections were incubated with the primary antibodies, mouse anti-human IP-10 diluted at 1:10, MCP-1 (1:2000), MCP-4 (1:50), eotaxin (1:50; Dr A. D. Luster, Harvard Medical School, Boston, Mass), and MCP-3 (1:1000, Cederlane).

In situ hybridization

In situ hybridization with digoxigenin-labeled complementary riboprobes for IP-10, MCP-1, MCP-3, MCP-4, and eotaxin was performed as previously described.²³ Briefly, biopsy and BAL samples were permeabilized with Triton X-100 with proteinase K at 37°C . The reaction was terminated by immersion of the preparation in 4% paraformaldehyde. For hybridization, digoxigenin-labeled riboprobe diluted in hybridization buffer was applied to each section.

TABLE I. Differential cell counts from BAL preparations of patients with asthma, tuberculosis, sarcoidosis, and chronic bronchitis and normal control subjects

	Asthma	Tuberculosis	Sarcoidosis	Control	Chronic bronchitis
Macrophages (%)	81.2	78.5	59	85.4	90
Lymphocytes (%)	10.9	11.3	39	9.6	7.5
Eosinophils (%)	3.8	0.0	0.1	0.0	0.5

TABLE II. Immunohistochemical assessment of distribution of chemokines in BAL cells and in bronchial biopsy specimens

	IP-10	MCP-1	MCP-3	MCP-4	Eotaxin
Normal control subjects					
BAL cells, n = 10 (%)	7.9 ± 0.5	7.4 ± 0.8	7.8 ± 1.3	7.3 ± 0.7	4.5 ± 0.8
Biopsy specimens, n = 7 (No. of positive cells per BM)		4.5 ± 0.8	2.6 ± 0.3	2.9 ± 0.4	1.2 ± 0.4
Asthma					
BAL cells, n = 10	19.3 ± 1.5*	10.2 ± 0.6§	15.5 ± 0.9‡	28.1 ± 2.0*	17.9 ± 1.4*
Biopsy specimens, n = 7		5.3 ± 0.8‡	12.7 ± 0.7‡	16.2 ± 1.7†	11.7 ± 1.1‡
Tuberculosis					
BAL cells, n = 10	19.6 ± 1.6*	20.2 ± 1.6*	19.3 ± 1.7†	13.5 ± 1.4‡	6.5 ± 1.1
Biopsy specimens, n = 8		11.3 ± 1‡	8.6 ± 0.6†	5.3 ± 0.6‡	3.1 ± 0.4
Sarcoidosis					
BAL cells, n = 10	44.8 ± 3.4*	7 ± 0.6	11.4 ± 1.6	7.6 ± 1.2	6.8 ± 0.8
Biopsy specimens, n = 7		5.7 ± 1.7	3.1 ± 0.5	3.6 ± 0.7	2.6 ± 1.3
Chronic bronchitis					
BAL cells, n = 9	9.7 ± 0.8	12.8 ± 0.9‡	16.2 ± 1.0	14.7 ± 1.4†	5.7 ± 0.7

BAL cells and airways section from patients and normal control subjects were stained with antichemokine antibodies, as described in the "Methods" section. A minimum of 1000 cells were counted in BAL preparations in at least 2 fields, and results from biopsy specimens were expressed as the number of positive cells per square millimeter of BM. Values are expressed as means ± SEM.

BM, Basement membrane.

**P* < .0005 versus control subjects.

†*P* < .001 versus control subjects.

‡*P* < .005 versus control subjects.

§*P* < .05 versus control subjects.

Posthybridization washing was performed in a decreasing concentration of standard saline citrate (4× standard saline citrate to 0.01× standard saline citrate) at 42°C. Slides were incubated in bovine serum and then with a sheep polyclonal antidigoxigenin antiserum conjugated with alkaline phosphatase, incubated with substrate solution, counterstained with hematoxylin, and mounted in glycerol gelatin. As a negative control, preparations were hybridized with sense probes under the same conditions.

Quantification and data analysis

Cytospin slides and bronchial biopsy sections were coded, and the number of positive cells for protein or mRNA was calculated. In biopsy specimens results were expressed as the number of positive cells per square millimeter. In BAL fluid a minimum of 1000 cells was counted, and the number of positive cells was expressed as a percentage of the total cells. BAL fluid differential cell counts were done by using conventional histology and expressed as the percentage of macrophages, lymphocytes, and eosinophils per total cell count (Table I). Statistical differences in the number of positive cells between groups were assessed by using the nonparametric Kruskal-Wallis ANOVA, and subsequent post hoc analyses were performed with the Mann-Whitney *U* test. A *P* value of less than .05 was considered statistically significant.

RESULTS

The immunoreactivity of all the chemokines (ie, MCP-1, MCP-3, MCP-4, eotaxin, and IP-10) examined in this study were significantly increased in BAL fluid

and biopsy specimens from asthmatic subjects compared with control subjects. There was an increased expression of MCP-1, MCP-3, MCP-4, and IP-10, but not eotaxin, in samples obtained from patients with tuberculosis compared with control subjects. In patients with sarcoidosis, only IP-10 immunoreactivity was significantly higher when compared with that of control subjects. We were unable to demonstrate any increase in the expression of eotaxin or IP-10 in patients with chronic bronchitis compared with that in control subjects; however, MCP-1 and MCP-4 were significantly increased. The numbers of MCP-1-, MCP-3-, and MCP-4-immunoreactive cells in both BAL fluid and biopsy specimens in the different groups of subjects with lung diseases, such as asthma, tuberculosis, sarcoidosis, and chronic bronchitis, and the normal control subjects are shown in Table II, and those of IP-10 and eotaxin are shown in Figs 1 and 2. The highest expression of IP-10 was demonstrated in subjects with sarcoidosis, and the highest expression of eotaxin was demonstrated in asthmatic subjects.

Similar results were obtained from in situ hybridization. The numbers of MCP-1, MCP-3, and MCP-4 mRNA-positive cells in the different subject groups are presented in Table III. The numbers of eotaxin mRNA-positive cells both in BAL fluid and biopsy specimens are shown in Figs 1 and 2, as well as the number of IP-

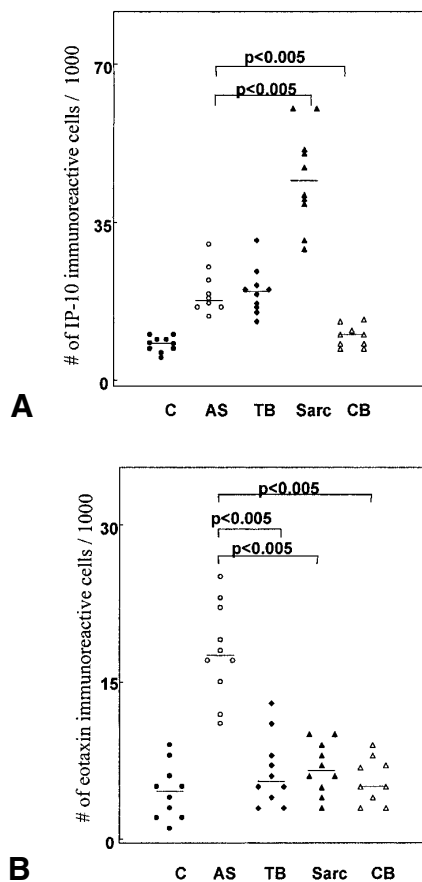


FIG 1. Individual values for IP-10 (**A**) and eotaxin (**B**) immunoreactivity in BAL fluid cells of normal subjects (*C*) and patients with asthma (*AS*), tuberculosis (*TB*), sarcoidosis (*Sarc*), and chronic bronchitis (*CB*). Results are expressed as the percentage of positive cells per 1000 cells. Horizontal bars represent median values.

10 mRNA-positive cells. The number of MCP-1 and MCP-3 mRNA-positive cells was highest in patients with tuberculosis compared with those in the other patient groups ($P < .005$). Eotaxin mRNA expression was highest in asthmatic tissue ($P < .005$) and expressed at low levels in normal control subjects and in patients with tuberculosis, sarcoidosis, and chronic bronchitis.

DISCUSSION

In this study we report the expression of CC and CXC chemokines both in allergic diseases, such as asthma, and in T_H1 -mediated diseases, such as tuberculosis and sarcoidosis. According to our results, eotaxin and IP-10 are the only chemokines among those we studied that are specific for a particular kind of immune response.

We investigated the expression of chemokines in several airways diseases, such as asthma, tuberculosis, sarcoidosis, and chronic bronchitis, each characterized by a different pattern of cytokine expression and a different inflammatory cell infiltrate. Asthma is a T_H2 -mediated disease in which activated $CD4^+$ T cells and

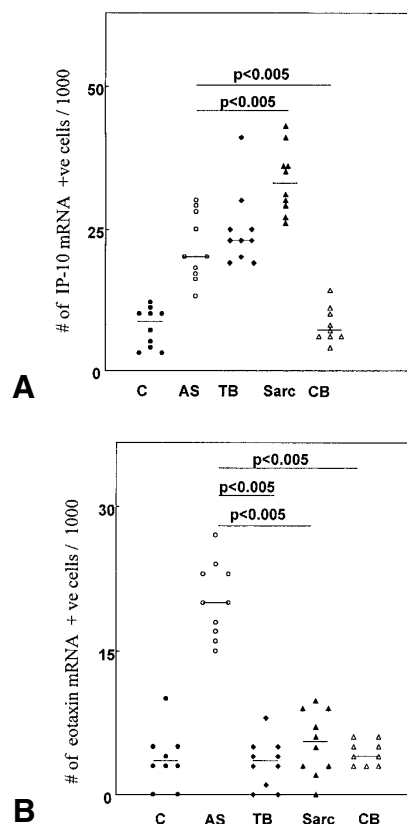


FIG 2. Individual values for IP-10 (**A**) and eotaxin (**B**) mRNA expression in BAL fluid cells of normal subjects (*C*) and patients with asthma (*AS*), tuberculosis (*TB*), sarcoidosis (*Sarc*), and chronic bronchitis (*CB*). Results are expressed as the percentage of chemokine mRNA-positive cells per 1000 cells. Horizontal bars represent median values.

eosinophils play a predominant role.^{9,24} Sarcoidosis and tuberculosis, on the other hand, are considered to be T_H1 -mediated diseases in which T cells and macrophages are the prominent cell types.²⁵⁻²⁷ Chronic bronchitis is associated mainly with neutrophils, macrophages, and $CD8^+$ cells²⁸⁻³⁰ and does not seem to be characterized by a particular cytokine profile. The mechanism responsible for the recruitment of leukocytes to the site of inflammation in these diseases involves the local generation of chemokines regulating their influx to the injured site. We examined the expression of the CC chemokines MCP-1, MCP-3, MCP-4, and eotaxin and the expression of the CXC chemokine IP-10 in the airways of subjects with the lung diseases mentioned above.

MCPs and eotaxin have mainly been implicated in T_H2 immune responses because they are chemoattractants for eosinophils, T lymphocytes, macrophages, and basophils, all of which are characteristic of allergic diseases.^{14-18,31} IP-10, on the other hand, has mainly been associated with T_H1 -mediated disorders because it is a chemoattractant for neutrophils and lymphocytes.³²⁻³⁴

TABLE III. Percentage of cells with positive hybridization signals for chemokines in BAL cells and bronchial biopsy specimens

	IP-10	MCP-1	MCP-3	MCP-4	Eotaxin
Normal control subjects					
BAL cells, n = 10	7.5 ± 1.1	6.1 ± 0.9	6.8 ± 0.8	8.8 ± 0.7	3.8 ± 0.9
Biopsy specimens, n = 10	1.42 ± 0.27	2.8 ± 0.21	2.52 ± 1	2.29 ± 1.2	2 ± 0.60
Asthma					
BAL cells, n = 10	21.6 ± 1.9*	5.3 ± 0.8‡	16.5 ± 1.1*	28 ± 1.8*	20.3 ± 1.2†
Biopsy specimens, n = 10	4.73 ± 0.39*	3.22 ± 0.50‡	7.69 ± 0.67*	8.67 ± 0.72*	7.9 ± 0.67*
Tuberculosis					
BAL cells, n = 10	24.8 ± 2.1*	17.6 ± 1.1*	17.5 ± 0.9*	12.6 ± 1.2‡	3.3 ± 0.8
Biopsy specimens, n = 10	4.43 ± 0.37	10.5 ± 1.4*	5.8 ± 0.43*	2.4 ± 0.49	1.9 ± 0.37
Sarcoidosis					
BAL cells, n = 10	33.4 ± 1.8*	7.2 ± 0.9	11.3 ± 1.7	6.3 ± 0.9	5.4 ± 1.1
Biopsy specimens, n = 10	8.7 ± 0.76*	2.2 ± 0.38	4.78 ± 0.55	1.8 ± 0.3	1.65 ± 0.33
Chronic bronchitis					
BAL cells, n = 9	8 ± 1.0	14.1 ± 0.7*	16.3 ± 1.1*	16.1 ± 1.6†	4.3 ± 0.4
Biopsy specimens, n = 10	3.9 ± 1.80	6.5 ± 0.83*	3.95 ± 0.39*	4.72 ± 0.52	1.86 ± 0.59

In situ hybridization was performed on BAL cells and airways sections from patients and normal control subjects, as described in the "Methods" section. A minimum of 1000 cells were counted in BAL preparations in at least 2 fields, and results from biopsy specimens were expressed as the number of positive cells per square millimeter of BM. Values are expressed as means ± SEM.

* $P < .0005$ versus control subjects.

† $P < .001$ versus control subjects.

‡ $P < .05$ versus control subjects.

In this study we found an increase in MCPs not only in asthma but also in T_H1 -mediated lung disorders. MCP-1, MCP-3, and MCP-4 were all increased in the airways of asthmatic subjects, in subjects with chronic bronchitis, and in subjects with tuberculosis, suggesting that MCPs target monocytes and T cells not only in T_H2 -type but also in T_H1 -type immune responses. Although we did not show a significantly increased expression of CC chemokines in patients with sarcoidosis, Car et al³⁵ have previously shown an increased expression of MCP-1 in the BAL fluid of such patients. MCP-1, in particular, has been shown to be abundant in mononuclear cell-rich inflammatory conditions, such as granulomatous inflammation, and in macrophage-rich regions of atherosclerotic lesions. Furthermore, Chensue et al³⁶ have reported a role of MCP-1 both in T_H1 and T_H2 antigen-induced granuloma formation. The fact that the expression of MCP-1, MCP-3, and MCP-4 is upregulated in diseases such as tuberculosis and chronic bronchitis in our study also suggests that these chemokines act nonspecifically to attract monocytes and T lymphocytes in any type of immune response. Furthermore, the constitutive expression of MCPs has been reported in the epithelium of normal tissue, suggesting a role for MCPs in baseline leukocyte trafficking through epithelial tissue as part of the natural host defense against pathogens, as suggested by Garcia-Zepeda et al.¹⁶

Eotaxin, also a CC chemokine, has been suggested to be specifically involved in T_H2 -type immune responses. Its expression is stimulated by IL-4, it exerts chemoattractant activity preferentially on eosinophils, and its receptor is mainly expressed by T_H2 -type cells.^{11,13,14} Consistent with previous findings, we observed an increased expression of eotaxin both at the protein and

mRNA levels in the BAL fluid and biopsy specimens of asthmatic patients only and not in subjects with tuberculosis, sarcoidosis, or chronic bronchitis. This finding may explain the abundance of eosinophils present in asthmatic airways and lacking in T_H1 -mediated diseases of the lung. In fact, studies have suggested a strong association among eosinophil numbers, their state of activation, and the severity of asthma. Eotaxin has been reported to be one of the most potent eosinophil chemoattractants, and this may be one mechanism for their selective recruitment into the airways.¹⁴ The increased expression of eotaxin may also contribute to basophil recruitment because it has been shown in vitro to have chemotactic activity on this cell type as well.³⁷ Although eotaxin seems to be the main contributor to eosinophil chemotaxis, the process governing eosinophil recruitment is likely to be the result of a combination of chemokines. Neutralizing antibodies against MCP-4 have been shown to abolish part of the chemoattractant activity of eosinophils found in the BAL fluid of asthmatic patients, suggesting that MCPs also contribute to the accumulation of inflammatory cells in the airways of asthmatic subjects.¹⁴ The fact that eotaxin expression is upregulated in asthma compared with other lung diseases supports the idea that this chemokine is specific for T_H2 immune responses. The major source of eotaxin in the airways seems to be the epithelium. Proinflammatory cytokines, such as IL-1 and TNF- α , have been reported to induce the expression of eotaxin by epithelial cells, and increased expression of this chemokine has been detected after allergen challenge in allergic disease.³⁸⁻⁴⁰

IP-10 is a member of the C-X-C subfamily, and in vitro, IP-10 induces chemotaxis of stimulated human T lymphocytes and natural killer cells.^{32,41} IP-10 is

believed to be more important in the T_H1 -type immune responses because its expression is stimulated by $IFN-\gamma$ and inhibited by IL-4.^{11,33} Its expression has been shown to be upregulated in delayed-type immune responses of the skin, in murine macrophages infected with *M tuberculosis*,¹² in atherosclerosis,⁴² and in viral meningitis.⁴³ Surprisingly, we have found increased expression of IP-10 not only in tuberculosis and sarcoidosis but also in asthma. Its expression, however, is highest in subjects with sarcoidosis compared with those with asthma, tuberculosis, and chronic bronchitis, confirming that IP-10 is mainly involved in the pathogenesis of T_H1 disease yet not selective for this type of inflammation.

The results of our study suggest that eotaxin is selectively increased in T_H2 -mediated airway diseases, and we support the concept that this chemokine plays a central role in the pathogenesis of asthma. On the other hand, the increased expression of MCPs in both T_H1 - and T_H2 -mediated diseases suggests that their expression is not associated with a particular cytokine profile and that they are nonspecifically expressed in airways inflammation. Similarly, IP-10 does not seem to be selectively expressed in T_H1 -mediated airway diseases because its upregulation was also seen in asthma. We therefore conclude that both IP-10 and eotaxin are differentially expressed in T_H1 versus T_H2 airways inflammation, whereas MCP-1, MCP-3, and MCP-4 are not.

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