Review Articles

Advances in Immunology

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Editors

ASTHMA

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ASTHMA is a complex syndrome with many clinical phenotypes in both adults and children. Its major characteristics include a variable degree of airflow obstruction, bronchial hyperresponsiveness, and airway inflammation. For many patients, the disease has its roots in infancy, and both genetic factors (atopy)[1,2] and environmental factors (viruses,[3] allergens,[4] and occupational exposures[5]) contribute to its inception and evolution. To comprehend the pathogenetic mechanisms underlying the many variants of asthma, it is essential to identify factors that initiate, intensify, and modulate the inflammatory response of the airway and to determine how these immunologic and biologic processes produce the characteristic airway abnormalities.

In this regard, immune responses mediated by IgE antibodies in the lung have come to the forefront. Indeed, the association of asthma with allergies has long been recognized, but until recently the mechanism of this association was a puzzle. Recent noteworthy advances have been made in defining allergic airway inflammation and relating it to the clinical manifestations of asthma. This review deals with the epidemiology of asthma, the initiation of the disorder, and the main features of IgE-mediated asthma.

IMMUNOHISTOPATHOLOGY OF ASTHMA

Evidence that inflammation was a component of asthma was initially derived from findings at autopsy in patients with fatal asthma. Their airways showed infiltration by neutrophils and eosinophils, degranulated mast cells, sub-basement-membrane thickening, loss of epithelial-cell integrity, and occlusion of the bronchial lumen by mucus. Hyperplasia and hypertrophy of bronchial smooth muscle and hyperplasia of goblet cells were also present. These findings were considered to be characteristic of fatal asthma, but not necessarily of other forms of the disease.

More recent studies have found substantial inflammation in bronchial-biopsy specimens from patients with asthma, even those with mild disease. These inflammatory changes can occur throughout the central[6] and peripheral[6,7] airways and often vary with the severity of the disease.[8,9] Although not observed uniformly, denudation of the airway epithelium, deposition of collagen beneath the basement membrane, mast-cell degranulation, and infiltration of the airway by lymphocytes and eosinophils have been found in patients with mild-to-moderate asthma (Fig. 1). Many of the cells in the airway appear to be activated, implying that by releasing preformed or newly synthesized mediators, they have a direct role in asthma.

Further evidence of an inflammatory response in asthma is the presence of cytokines that mediate inflammation and chemotactic chemokines in bronchoalveolar-lavage fluid or pulmonary secretions.[10] Since these cytokines and chemokines are elaborated by resident and inflammatory cells in airways and have many effects on these cells, a variety of autocrine, paracrine, and endocrine networks could participate in asthma (Table 1). Some cytokines initiate inflammatory responses by activating transcription factors, which are proteins that bind to the promoter region of genes. Transcription factors involved in asthmatic inflammation include nuclear factor-κB, activator protein-1, nuclear factor of activated T cells, cyclic AMP response-element binding protein, and various members of the family of signal transduction-activated transcription (STAT) factors. These transcription factors act on genes that encode inflammatory cytokines, chemokines, adhesion molecules, and other proteins that induce and perpetuate inflammation. Corticosteroids modulate immunoinflammatory responses in asthma by inhibiting these transcription factors.[11]

The ability of cytokines to induce the expression of adhesion molecules such as intercellular adhesion molecule 1, vascular-cell adhesion molecule 1, and endothelial-leukocyte adhesion molecule provides a mechanism for the adhesion of inflammatory cells to the endothelium and the migration of these cells from the circulation into the lamina propria, the epithelium, and in many cases, the airway lumen itself.[12]

ALLERGIC INFLAMMATION IN ASTHMA

Epidemiologic and clinical observations have linked IgE antibodies to the severity of asthma[13] and the initial and sustained responses of the airway to allergens.[14] To initiate the synthesis of IgE, inhaled allergens must

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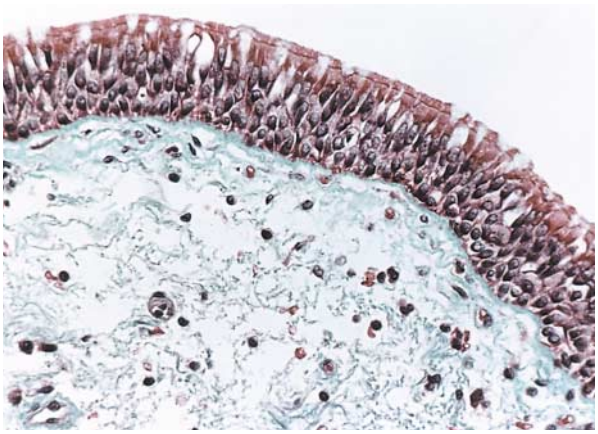
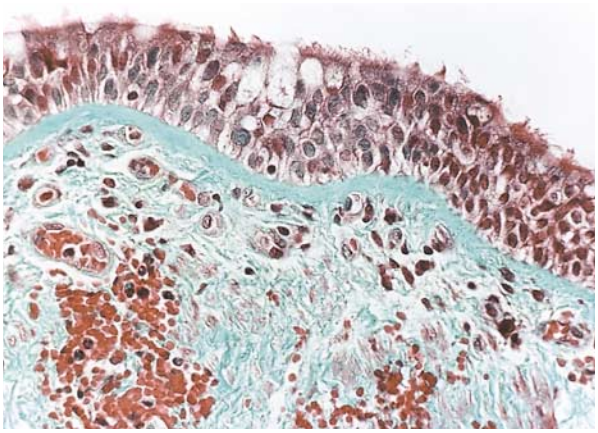


A



B

Figure 1. Specimen of Bronchial Mucosa from a Subject without Asthma (Panel A) and a Patient with Mild Asthma (Panel B) (Hematoxylin and Eosin, ×40).

In the subject without asthma, the epithelium is intact; there is no thickening of the sub-basement membrane, and there is no cellular infiltrate. In contrast, in the patient with mild asthma, there is evidence of goblet-cell hyperplasia in the epithelial-cell lining. The sub-basement membrane is thickened, with collagen deposition in the submucosal area, and there is a cellular infiltrate. Photographs courtesy of Nizar N. Jarjour, M.D., University of Wisconsin.

encounter dendritic cells that line the airway. These dendritic cells then migrate to draining lymph nodes, where they present processed antigen to T and B cells.[15] Interactions among these cells elicit responses that are influenced by cytokines and the presence or absence of costimulatory molecules. For example, a switch by B cells to the production of a particular immunoglobulin isotype requires two signals. For a switch to the synthesis of IgE, the first signal is delivered by interleukin-4 or interleukin-13 when these cytokines bind to receptors on B cells; the receptors for interleukin-4 and interleukin-13 share a common α chain and use the same signal-transduction pathway (STAT-6).[16] The second signal is delivered when CD40 on B cells binds to its ligand on T cells. Additional interactions between other pairs of ligands and receptors (between CD28 and B7 and between $\alpha_L\beta_2$ integrin and intercellular adhesion molecule 1) may complement or up-regulate the T cell–dependent activation of B cells that follows the binding of CD40 to its ligand (Fig. 2).[17]

Once synthesized and released by B cells, IgE antibodies briefly circulate in the blood before binding to high-affinity IgE receptors (FcϵRI) on the surface of mast cells in tissue or peripheral-blood basophils, and low-affinity IgE receptors (FcϵRII, or CD23) on the surface of lymphocytes, eosinophils, platelets, and macrophages. Whether the binding of IgE to its low-affinity receptors activates cells and contributes to inflammation is unclear. Soluble FcϵRII receptors, however, appear to be important in regulating IgE synthesis.[18] Molecular bridging of FcϵRI receptors, which occurs when allergen interacts with receptor-bound IgE molecules, causes activation of the cell and the release of preformed and newly generated mediators.[18] Interestingly, basophils and mast cells can secrete interleukin-4 and interleukin-13 and express the CD40 ligand; however, since the release of cytokines depends on cross-linking of IgE by allergen, these cells most likely amplify rather than induce the synthesis of IgE.[17]

Mast Cells

Mast cells arise in the bone marrow, enter the circulation as CD34+ mononuclear cells that are positive for stem-cell factor and FcϵRI, travel to mucosal and submucosal sites in the airway, and undergo tissue-specific maturation.[19] The cross-linking of mast-cell–bound IgE by allergen induces the activation of membrane and cytosolic pathways that cause the release of preformed mediators such as histamine and initiates the synthesis of arachidonic acid metabolites.[20]

There are at least two subpopulations of mast cells: mast cells with tryptase and mast cells with both tryptase and chymase. Although the role of these enzymes is not fully defined, inhibitors of tryptase have been shown to modulate the response of the airway to allergen.[21] Mast cells also contain proteoglycans with diverse biologic properties or functions, ranging from being supporting structures for various proteins (i.e., remodeling) to exerting effects on the differentiation and proliferation of cells, the adhesion and motility of cells, and tissue morphogenesis. Mast cells produce several cytokines, including interleukin-1, interleukin-2, interleukin-3, interleukin-4, interleukin-5, granulocyte–macrophage colony-stimulating factor, interferon-γ, and tumor necrosis factor α.[22] The potential for the extracellular release of these cytokines raises the possibility that mast cells contribute to both acute and chronic allergic inflammation.

The response of the airway to inhaled allergen provides insights into immunologic mechanisms that con-

TABLE 1. CYTOKINES THAT MAY HAVE A ROLE IN THE PATHOGENESIS OF ASTHMA.*

CYTOKINE	PRIMARY SOURCES	PRIMARY TARGETS	EFFECTS OR FUNCTION
Basic fibroblast growth factor	Endothelial cells	Fibroblasts, matrix	Production of fibroblasts, matrix formation
Granulocyte colony-stimulating factor	Monocytes, fibroblasts, epithelial cells	Neutrophil precursors	Maturation and differentiation of target cells
Granulocyte–macrophage colony-stimulating factor	Activated macrophages and T cells	Eosinophils, neutrophils, macrophages	Proliferation, differentiation, activation, and prolonged survival of target cells; enhanced cytokine production; degranulation of eosinophils
Interferon-α	Monocytes, macrophages	Virus-infected cells	Inhibition of viral replication
Interferon-β	Monocytes, macrophages	Virus-infected cells	Inhibition of viral replication
Interferon-γ	CD4+ Th1 cells, lymphocytes, natural killer cells, some CD8+ T cells	Macrophages	Differentiation of macrophages; activation of macrophages, leading to the expression of Fcγ receptors, MHC class I and II molecules, nitric oxide synthase, interleukin-1, tumor necrosis factor
		CD4+ T cells	Shift in cytokine profile from Th2 type to Th1 type; increased expression of interleukin-2 receptors
		CD8+ T cells	Increased cytotoxicity of CD8+ T cells
		Natural killer cells	Activation of natural killer cells
Interleukin-1	Monocytes, macrophages	CD4+ Th2 cells	Production of cytokines
		CD8+ T cells	Cellular cytotoxicity; production of cytokines
		B cells	Differentiation of B cells; proliferation of B cells; production of immunoglobulin
Interleukin-2	CD4+ T cells	T cells	Clonal expansion of antigen-specific cells; differentiation and expression of cytokines; maturation of CD8+ T cells
Interleukin-3	T cells	Hematopoietic stem cells	Proliferation and differentiation of target cells
Interleukin-4	CD4+ Th2 cells	B cells	Growth and activation of B cells; production of MHC class II molecules, interleukin-6, tumor necrosis factor, CD23; class switching to IgE; enhancement of IgE, IgG1, and IgG4 and inhibition of IgM, IgG2, and IgG3 production
		Th1 cells	Inhibition of differentiation of Th1 cells and production of interferon-γ
		Th2 cells	Differentiation of Th2 cells
		CD8+ T cells	Differentiation of CD8+ T cells; production of interleukin-5
		Natural killer cells	Inhibition of proliferation
Interleukin-5	CD4+ T cells, CD8+ T cells	Eosinophils	Proliferation, chemoattraction, adhesion, activation, enhanced survival, and degranulation of eosinophils
Interleukin-6	Monocytes, macrophages	B cells	Maturation of B cells into plasma cells; class switching to IgG1 and IgA
		Monocytes, macrophages	Inhibition of lipopolysaccharide; production of interleukin-1 and tumor necrosis factor α
Interleukin-7	Bone marrow stromal cells	Pre-B cells	Proliferation of progenitors
		T cells	Proliferation of activated T cells
Interleukin-8	Macrophages	Neutrophils	Directed migration of neutrophils to endothelium but inhibition of adhesion of these cells
Interleukin-9	CD4+ T cells (especially Th2)	B cells	Enhancement of response to interleukin-4†
Interleukin-10	CD4+ Th0 cells, Th1 cells, Th2 cells, CD8+ T cells	Monocytes	Differentiation to macrophages
		Macrophages	Inhibition of the expression of MHC class II molecules and many adhesion molecules; inhibition of interferon-γ and tumor necrosis factor production, resulting in switching of T-cell differentiation from Th1 to Th2; inhibition of interleukin-4 and interferon-γ by Th2 cells
Interleukin-11	Bone marrow stromal cells	B cells and plasma cells	Similar to those of interleukin-6
Interleukin-12	Monocytes, macrophages	Natural killer cells	Activation of natural killer cells
		Th0 cells	Production and proliferation of interleukin-2
		Th1 cells	Production of interferon-γ and tumor necrosis factor α
		Th2 cells	Inhibition of production of interleukin-4, 5, and 10
Interleukin-13	CD4+ Th2 cells	B cells	Similar to those of interleukin-4
		Monocytes	Enhancement of production of MHC class II molecules and integrins; inhibition of production of interleukin-1 and tumor necrosis factor
Interleukin-14	Activated T cells	Activated B cells	Expansion of B-cell clones and suppression of immunoglobulin secretion
Interleukin-15	Monocytes, macrophages	T cells, natural killer cells	Proliferation and increased cytotoxicity of target cells; expression of intercellular adhesion molecule 3
Interleukin-16	CD8+ T cells	CD4+ T cells	Chemoattraction, growth factor
Interleukin-17	CD4+ memory cells	CD4+ T cells	Proliferation and activation of autocrine factors
Interleukin-18	Macrophages	Activated B cells	Similar to those of interleukin-12; inhibition of IgE production by increasing interferon-γ
Macrophage colony-stimulating factor	Monocytes, fibroblasts, epithelial cells	Multipotential hematopoietic precursors	Differentiation of monocytes
Platelet-derived growth factor	Alpha granules of platelets, monocytes, macrophages	Fibroblasts and smooth-muscle cells	Proliferation of target cells; chemoattractant for fibroblasts; active in wound healing, atherogenesis, and airway remodeling
Stem-cell factor (also called c-*kit* ligand)	Bone marrow stroma, fibroblasts	Mast cells	Chemoattraction; along with interleukin-3, stimulation of growth; induction of histamine release

*Th1 denotes type 1 helper T, MHC major histocompatibility complex, Th2 type 2 helper T, and Th0 precursor of Th1 and Th2.

†Interleukin-2 is required for this response.

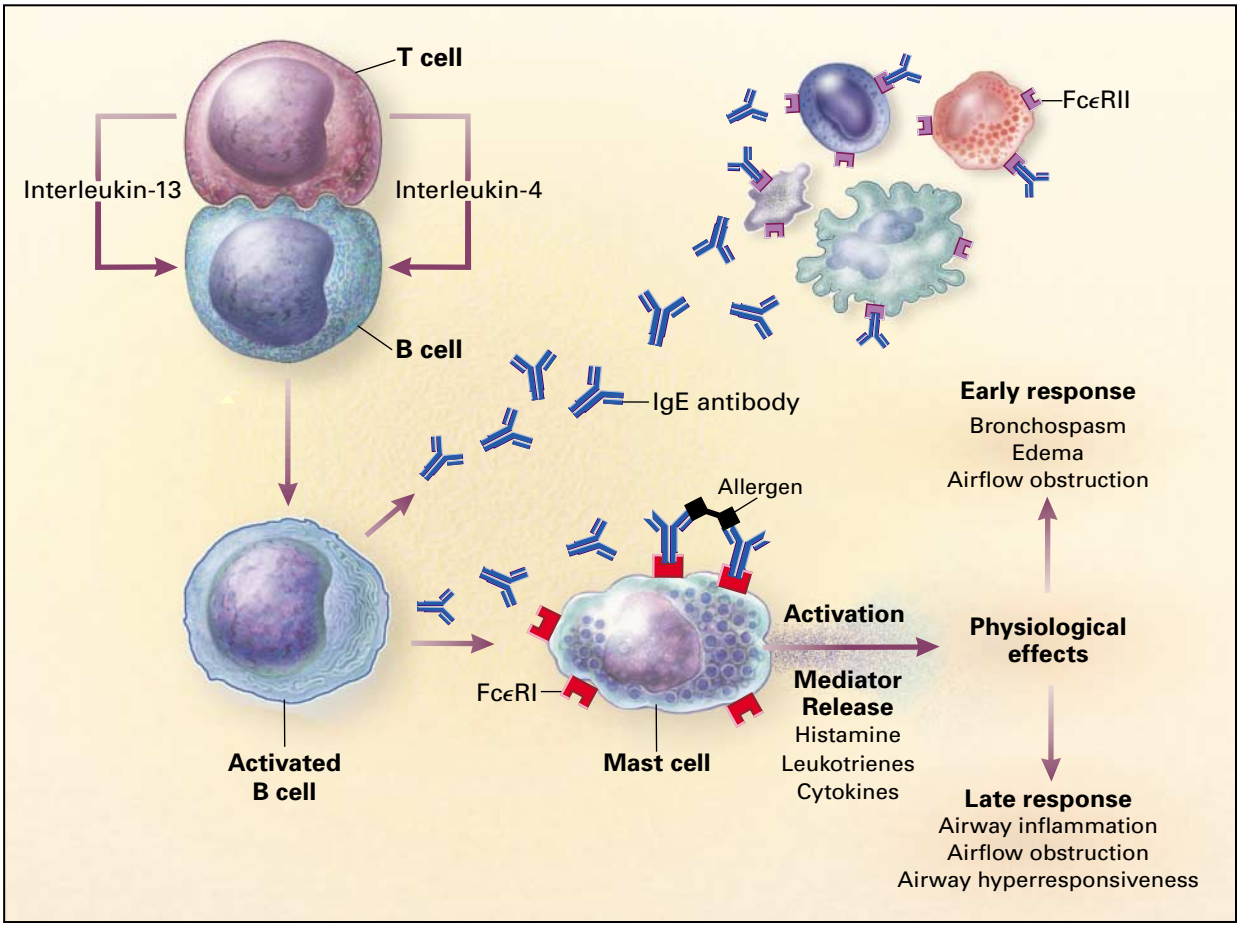


Figure 2. Interactions between CD4 T Cells and B Cells That Are Important in IgE Synthesis.

Interleukin-4 and interleukin-13 provide the first signal to B cells to switch to the production of the IgE isotype. The second signal is provided by accessory pairs of molecules, such as $\alpha_L\beta_2$ integrin and intercellular adhesion molecule 1 and CD40 and its ligand. The engagement of allergen by the complex of the T-cell receptor and CD3 on major-histocompatibility-complex (MHC) class II B cells results in the rapid expression of the CD40 ligand. Once formed, IgE antibody circulates in the blood, eventually binding to both high-affinity IgE receptors (FcεRI) and low-affinity IgE receptors (FcεRII, or CD23). After subsequent encounters with antigens, binding of the high-affinity IgE receptors produces the release of preformed and newly generated mediators. Once present in various tissues, mediators may produce various physiological effects, depending on the target organ.

tribute to the pathogenesis of asthma.[23] In patients with asthma, inhaled allergen precipitates acute obstruction of the airway by initiating the release from mast cells of histamine and leukotrienes, which cause constriction of smooth muscles. This early-phase reaction usually resolves within an hour. Four to six hours later, a prolonged late-phase reaction with obstruction of airflow may develop as a result of cytokines and chemokines generated by resident inflammatory cells (e.g., mast cells, macrophages, and epithelial cells) and recruited inflammatory cells (lymphocytes and eosinophils).

Eosinophils

Eosinophilopoiesis begins in the bone marrow and is regulated by interleukin-3, interleukin-5, and granulocyte–macrophage colony-stimulating factor; interleukin-5 induces terminal differentiation of immature eosinophils.[24] The mature eosinophil has dense intracellular granules that are sources of inflammatory proteins, including major basic protein, eosinophil-derived neurotoxin, peroxidase, and cationic protein. Major basic protein, in particular, can directly damage airway epithelium, intensify bronchial responsiveness, and cause degranulation of basophils and mast cells. These effects increase the severity of asthma. The eosinophil is a rich source of leukotrienes, particularly the cysteinyl leukotriene C_4, which contracts airway smooth muscle, increases vascular permeability, and may recruit more eosinophils to the airway.[25]

A number of cytokines regulate the function of eosinophils and other cells in asthma (Table 1). Interleu-

kin-5 stimulates the release of eosinophils into the circulation and prolongs their survival. Challenge of the airway with allergen increases the local concentration of interleukin-5, which correlates directly with the degree of airway eosinophilia.[26] In mice lacking the gene for interleukin-5, eosinophilia does not occur after challenge by an antigen.[27] Direct administration of interleukin-5 to the airway in humans causes mucosal eosinophilia and an increase in bronchial responsiveness.[28] Whether interleukin-5 alone is sufficient to cause eosinophilic inflammation is not established in humans; in mice, it is not sufficient to induce an asthma-like state.[24] A recent study involving an antibody against interleukin-5 in humans has indicated a dissociation between the concentration of eosinophils in peripheral blood and sputum and the late asthmatic responses and bronchial hyperresponsiveness that follow challenge of the airway with allergen.[29]

To participate in the allergic inflammatory response, the eosinophil must migrate from the circulation to the airway.[30,31] The first step in this process is the phenomenon of cell rolling, which is mediated by P-selectin on the surface of eosinophils (Fig. 3). Cell rolling activates eosinophils and requires the participation of the β_1 and β_2 classes of integrins on the eosinophil surface. Eosinophils and lymphocytes express the β_1 integrin $\alpha_4\beta_1$ integrin (also referred to as very late antigen 4, or VLA4), which binds to its ligand, vascular-cell adhesion molecule 1. Adhesion of the eosinophil to vascular-cell adhesion molecule 1 decreases the threshold for activation of the cell by mediators.[32] The interactions between the β_2 integrins on eosinophils and intracellular adhesion molecule 1 on vascular tissue appear to be important for the transendothelial migration of eosinophils.[33] The β_1 and β_2 integrins are constitutively expressed on the surface of eosinophils, but their state of activity is regulated by a variety of cytokines and chemokines.

The chemokines RANTES, macrophage inflammatory protein 1α, and the eotaxins are central to the delivery of eosinophils to the airway (Table 2).[9,34,35] These chemoattractants are produced by epithelium, macrophages, lymphocytes, and eosinophils.[9] Chemokines have been detected on cells and in airway tissue from patients with asthma. Berkman et al.[36] found that the constitutive expression of messenger RNA (mRNA) for RANTES was greater in the airway of patients with asthma than in normal subjects. Holgate et al.[37] detected RANTES, macrophage inflammatory protein-1α, and monocyte chemotactic protein 1 in the airway of normal subjects and patients with asthma within four hours after airway challenge with allergen. At 4 hours, there was a positive correlation between RANTES concentrations and the number of eosinophils in the air space, and the concentrations of all three chemokines returned to base-line values within 24 hours. Ying et al.[38,39] performed immunohistochemical studies of airway-biopsy specimens from normal subjects, allergic patients with asthma, and patients with nonallergic asthma and found that epithelial cells, endothelial cells, and macrophages were the primary sources of eotaxin, eotaxin-2, RANTES, and monocyte chemotactic proteins 3 and 4. Moreover, significant correlations were found between the degree of staining of eosinophils for EG2, a monoclonal antibody against the cleaved form of eosinophil cationic protein, and the concentrations of eotaxin. Collectively, the characteristics of many of the chemokines that act through the CCR3 receptor on the eosinophil, such as eotaxin, suggest that they are important in attracting eosinophils to the airway in asthma.

Lymphocytes

Mucosal-biopsy specimens obtained from patients during an episode of asthma after the inhalation of allergen contain lymphocytes, many of which express surface markers of activation.[40] In mice, there are two types of helper CD4+ T cells. In simple terms, type 1 helper T (Th1) cells produce interleukin-2 and interferon-γ, which are essential for cellular defense mechanisms. In contrast, type 2 helper T (Th2) cells produce cytokines (interleukin-4, 5, 6, 9, and 13) that mediate allergic inflammation. Furthermore, there is reciprocal inhibition, in that Th1-type cytokines inhibit the production of Th2-type cytokines and vice versa. CD8+ T cells may also be classified in a similar fashion according to their cytokine profiles (Tc1 and Tc2).[41] These observations in rodents raise the possibility that allergic (asthmatic) inflammation results from a Th2-mediated mechanism.

Figure 3 (facing page). The Role of Eosinophils in Allergic Inflammation.

Inhaled antigen activates mast cells and Th2 cells in the airway. They in turn induce the production of mediators of inflammation (such as histamine and leukotrienes) and cytokines including interleukin-4 and interleukin-5. Interleukin-5 travels to the bone marrow and causes terminal differentiation of eosinophils. Circulating eosinophils enter the area of allergic inflammation and begin migrating to the lung by rolling, through interactions with selectins, and eventually adhering to endothelium through the binding of integrins to members of the immunoglobulin superfamily of adhesion proteins: vascular-cell adhesion molecule 1 (VCAM-1) and intercellular adhesion molecule 1 (ICAM-1). As the eosinophils enter the matrix of the airway through the influence of various chemokines and cytokines, their survival is prolonged by interleukin-5 and granulocyte–macrophage colony-stimulating factor (GM-CSF). On activation, the eosinophil releases inflammatory mediators such as leukotrienes and granule proteins to injure airway tissues. In addition, eosinophils can generate granulocyte–macrophage colony-stimulating factor to prolong and potentiate their survival and contribution to persistent airway inflammation. MCP-1 denotes monocyte chemotactic protein, and MIP-1α macrophage inflammatory protein.

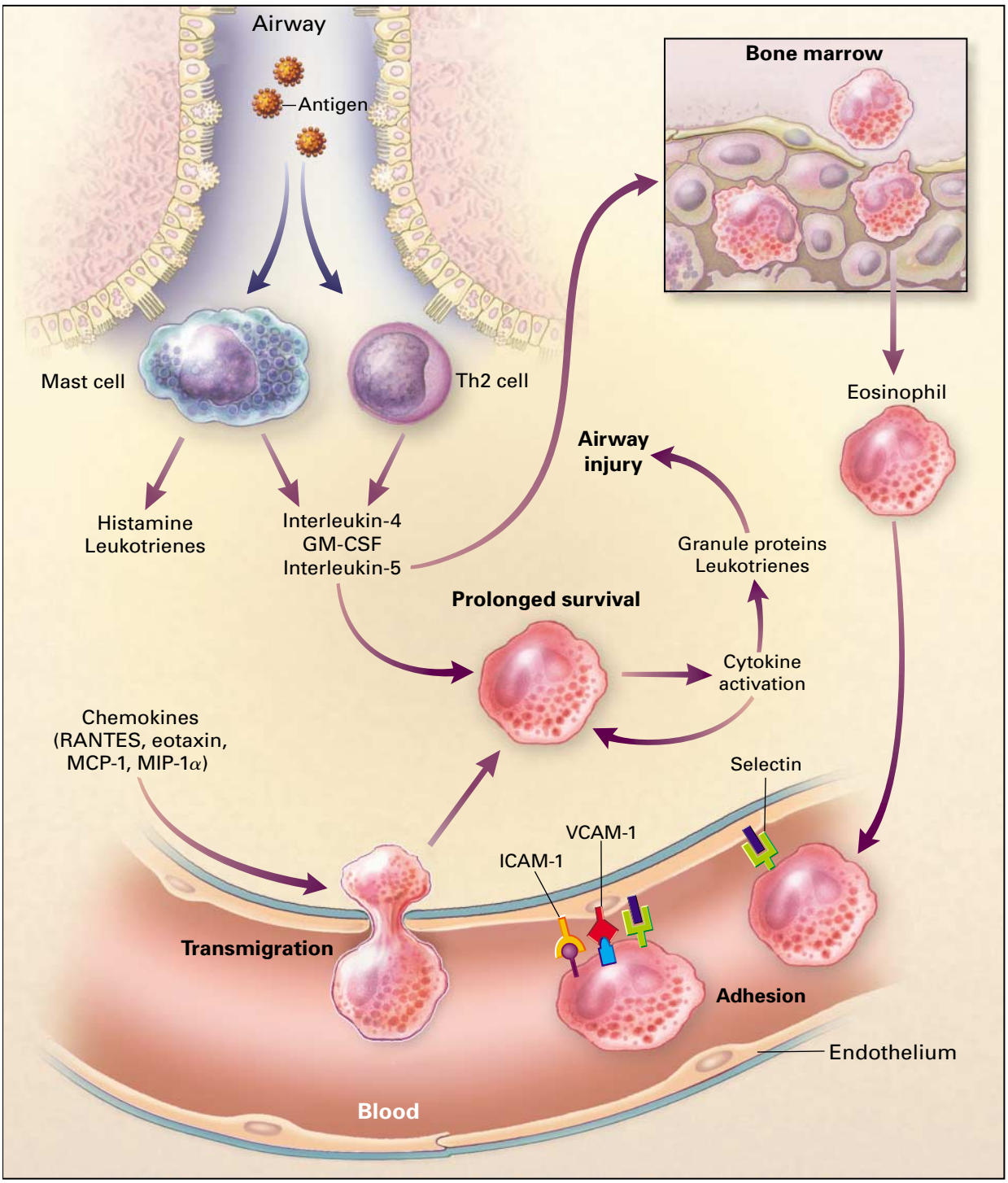


TABLE 2. HUMAN CHEMOKINES OF POTENTIAL IMPORTANCE IN ASTHMA.*

CHEMOKINE	SOURCES	TARGET CELLS	CHEMOKINE RECEPTORS	EFFECTS OR FUNCTION
CXC (α) chemokines				
GRO-α, GRO-β, GRO-γ	Monocytes, macrophages	Neutrophils	CXCR2	Chemotaxis, activation
	Fibroblasts	Basophils		Chemotaxis, activation
	Endothelial cells	Fibroblasts		Growth factor
	Epithelial cells	Endothelial cells		
	Neutrophils	T cells		Inhibition of angiogenesis (GRO-β, GRO-γ)
	T cells			Chemotaxis
Interleukin-8	Monocytes, macrophages	Neutrophils	CXCR1	Chemotaxis, activation
	T cells	Basophils	CXCR2	Chemotaxis, inhibition of histamine release
	B cells	T cells		Chemotaxis, inhibition of interleukin-4 synthesis
	Eosinophils	B cells, B-cell chronic lymphocytic leukemia		Chemotaxis, inhibition of growth and IgE production
	Epithelial cells			
	Endothelial cells	Endothelial cells		Promotion of angiogenesis
	Fibroblasts	Keratinocytes		Chemotaxis, expression of HLA-DR
CC (β) chemokines				
Eotaxin	Epithelial cells	Eosinophils	CCR3	Chemotaxis
	Endothelial cells	Basophils		Chemotaxis, activation
	Eosinophils	Th2 cells		Chemotaxis
	Fibroblasts			
	Macrophages			
Eotaxin-2	Monocytes, basophils, resting T cells	Eosinophils	CCR3	Chemotaxis, activation
MCP-1	Monocytes	Monocytes	CCR2	Chemotaxis
	Eosinophils	Basophils	CCR10	Activation
	Epithelial cells	T cells		Chemotaxis, inhibition of Th1-type cytokines, differentiation of CD4+ T cells to Th1 phenotype
	Endothelial cells			
	Fibroblasts			Chemotaxis, activation
	Mast cells	Natural killer cells		Chemotaxis, activation
MCP-2	Fibroblasts	T cells, eosinophils, basophils	CCR2	Chemotaxis, activation
	Natural killer cells			Chemotaxis, activation
	Dendritic cells			Chemotaxis
MCP-3	Monocytes	Monocytes	CCR1	Chemotaxis
	Epithelial cells	Eosinophils	CCR3	Chemotaxis
		Basophils	CCR10	Chemotaxis, activation
		Natural killer cells		Chemotaxis, activation
		Dendritic cells		Chemotaxis
MCP-4	Epithelial cells	Eosinophils	CCR2	Chemotaxis, activation
	Endothelial cells	Monocytes	CCR3	Chemotaxis
		Basophils		Chemotaxis, activation

MDC	Macrophages	Monocyte-derived dendritic cells	CCR4	Chemotaxis
	Monocyte-derived dendritic cells	Natural killer cells		Chemotaxis
	Thymus	Monocytes		Chemotaxis
	Lung	Eosinophils		Chemotaxis, shape change
MIP-1α	Mononuclear cells	T cells	CCR1	Chemotaxis, differentiation of CD4+ T cells to Th1 phenotype
	Endothelial cells	B cells	CCR5	Increased production of IgE and IgG4
	Smooth-muscle cells	Natural killer cells		Chemotaxis, activation
		Basophils		Activation
		Dendritic cells		Chemotaxis
MIP-1β	Mononuclear cells	T cells	CCR5	Chemotaxis
	Endothelial cells	Natural killer cells	CCR8	Chemotaxis, activation
		Dendritic cells		Chemotaxis
		Megakaryocytes		Increased production of IgE and IgG4
		B cells		
MIP-3α	Lymphocytes	Mononuclear cells	CCR6	Chemotaxis
	Monocytes	T cells		Chemotaxis
MIP-3β	Lymphoid tissue	Activated T cells	CCR7	Chemotaxis
				Adhesion to endothelial intercellular adhesion molecule 1
	Activated B cells			Chemotaxis
PARC	Lung	T cells	Not determined	Chemotaxis
RANTES	T cells	Eosinophils	CCR1	Chemotaxis
	Monocytes	T cells	CCR3	Chemotaxis
	Platelets	Monocytes	CCR5	Chemotaxis
	Eosinophils	Basophils		Chemotaxis
	Epithelial cells	Natural killer cells		Chemotaxis, activation
	Endothelial cells	B cells		Increased production of IgE and IgG4
	Dendritic cells			
TARC	Lymphoid tissue	T cells	CCR4	Chemotaxis
	Mononuclear cells	Th2 cells	CCR8	Chemotaxis
C (γ) chemokines				
Lymphotactin	CD8+ T cells	Lymphocytes	XCR1	Chemotaxis
	γ/δ Epithelial T cells	Activated natural killer cells		Chemotaxis, activation
	Natural killer cells			
CX3C chemokines				
Fractalkine	Endothelial cells	Monocytes	CX3CR1	Chemotaxis, adhesion to endothelium-bound fractalkine
		T cells		Chemotaxis, adhesion to endothelium-bound fractalkine

*Adapted from Nickel et al.[34] GRO denotes growth-related oncogene, CXCR receptor for CXC chemokine, DC-CK1 dendritic-cell chemokine, CCR receptor for CC chemokine, Th2 type 2 helper T, MCP monocyte chemotactic protein, Th1 type 1 helper T, MDC macrophage-derived chemokine, MIP macrophage inflammatory protein, PARC pulmonary- and activation-regulated chemokine, TARC thymus- and activation-regulated chemokine, XCR receptor for XC chemokine, and CX3CR receptor for CX3C chemokine.

A number of observations support this hypothesis. Recently, high concentrations of mRNA for GATA-3, a transcription factor that is confined to Th2 cells, were found in bronchial-biopsy specimens from patients with asthma.[42] In patients with asthma, more of the cells from bronchoalveolar-lavage fluid contain mRNA for interleukin-3, interleukin-4, interleukin-5, and granulocyte–macrophage colony-stimulating factor than do cells from bronchoalveolar-lavage fluid obtained from normal subjects.[43] The interleukin-4 and interleukin-5 were found predominantly in T cells.[44] In contrast, the number of cells containing mRNA for interferon-γ was similar in the two groups. The concentration of interleukin-5 protein is higher in bronchoalveolar-lavage fluid from patients with allergic or nonallergic asthma than in samples from patients who have other lung diseases such as hypersensitivity pneumonia and sarcoidosis.

Bronchial-biopsy specimens from patients with symptomatic allergic asthma or nonallergic asthma contain increased concentrations of mRNA for interleukin-4 and interleukin-5.[45] Thus, it seems that the bronchial mucosa in patients with asthma contains an excess of activated Th2 cells irrespective of the allergic sensitization of the patient, but whether this means that the immunopathology of allergic and nonallergic asthma is similar is unknown. In evaluating these results, it is important to acknowledge that interleukin-4 can contribute to allergic inflammation by mechanisms other than the regulation of IgE synthesis.

The idea that allergic inflammation in asthma arises from an imbalance between Th1 and Th2 cells has focused attention on the Th1-type cytokine interferon-γ. Since interferon-γ inhibits the synthesis of IgE and the differentiation of precursor cells to Th2 cells, a lack of interferon-γ would induce the Th2-type cytokine pathway to promote allergic inflammation. The evidence from in vivo studies of asthma, however, conflicts with this hypothesis. For example, the amount of interferon-γ is elevated in the serum of patients with severe asthma during the acute phase of an attack,[46] in supernatants from cultures of unstimulated and stimulated bronchoalveolar-lavage-fluid cells,[47] and in the lavage fluid itself after challenge with an allergen.[48] Furthermore, interferon-γ increases not only the expression of CD69, HLA-DR, and intercellular adhesion molecule 1 (all of which are markers of cell activation) on eosinophils but also the viability of eosinophils.[49] These and other data[50,51] suggest that interferon-γ contributes to the activation of eosinophils and thus is likely to augment inflammation. For these reasons, the classification of allergic inflammation in asthma as a Th2-mediated disease is too simplistic.

AN IMBALANCE BETWEEN Th1 AND Th2 CELLS AND THE ORIGINS OF ASTHMA

Although we question the importance of an imbalance between Th1 cells and Th2 cells in patients with established asthma, the possibility that this imbalance contributes to the cause and evolution of atopic diseases, including asthma, is intriguing. Largely as a result of Th2-trophic factors from the placenta, the population of T cells in the cord blood of newborn infants is skewed toward a Th2 phenotype.[2] The extent of the imbalance between Th1 cells and Th2 cells (as indicated by diminished production of interferon-γ) during the neonatal phase may be useful in predicting the subsequent development of allergic disease, asthma, or both.[2,52,53] To reduce the risk of asthma and allergies in childhood, some have suggested that infants at high risk for these conditions should be exposed to stimuli that up-regulate Th1-mediated responses, so as to restore the balance during a critical time in the development of the immune system and the lung.

The increasing prevalence of asthma in Western countries has led to the "hygiene hypothesis."[54,55] The basic tenet of this hypothesis is that the immune system of the newborn infant is skewed toward Th2 cells and needs timely and appropriate environmental stimuli to create a balanced immune response (Fig. 4). Factors that enhance Th1-mediated responses and that are associated with a reduced incidence of allergy, asthma, or both include infection with *Mycobacterium tuberculosis*,[56] measles virus,[57] and hepatitis A virus[58]; increased exposure to infections through contact with older siblings[54]; attendance at a day-care facility during the first six months of life[59]; and a reduction in the production of interferon-γ as a result of decreased exposure to environmental endotoxin or to polymorphisms of the major endotoxin receptor (CD14) that diminish the response to endotoxin.[60] Restoration of the balance between Th1 cells and Th2 cells may be impeded by frequent administration of oral antibiotics, with concomitant alterations in gastrointestinal flora (Fig. 4).[55] Immune "imprinting" may actually begin in utero through the transplacental transfer of allergens and cytokines.[61] Although these observations have generated intense interest, conflicting results have prevented researchers from drawing firm conclusions about the validity of the hygiene hypothesis.[62]

The relevance to asthma of allergic sensitization is supported by the evolution of the disease in later childhood. Indeed, many children with asthma have positive skin-prick tests to extracts of protein from house-dust mites,[63] cockroaches,[64] pets (especially cat dander),[65] and the fungi alternaria.[4] In 6-year-old children with asthma, sensitization to alternaria was associated with a significantly reduced frequency of remission of asthma by the age of 11 years (9 percent in those who were sensitized vs. 39 percent in those who were not sensitized).[4] Thus, it appears that the genetic background sets the stage for a cytokine imbalance that promotes the formation of IgE and that the allergens in the local environment dictate the specificity of the antibody response. Finally, sensitization to certain allergens, such as cockroach and alternaria

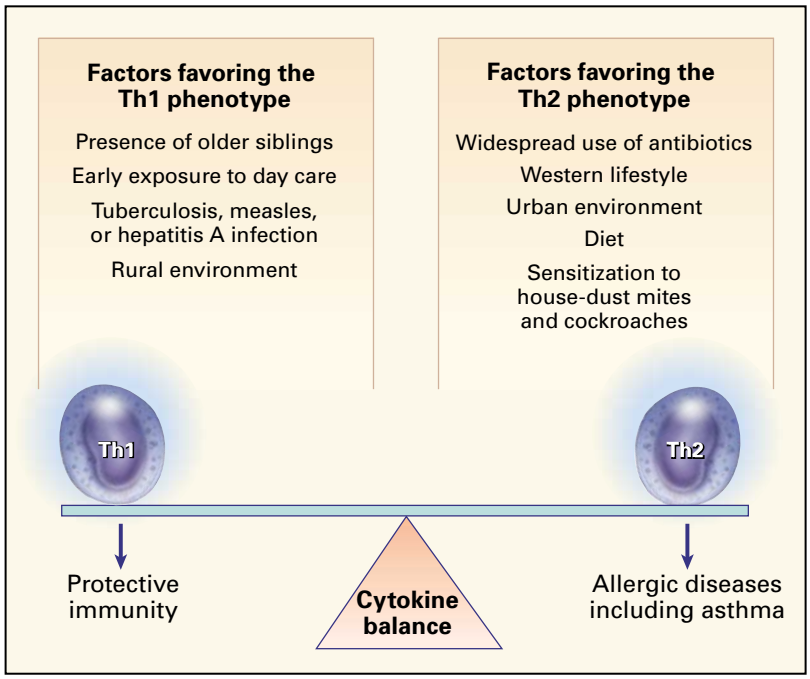


Figure 4. The Importance of Establishing a Balance between Th1-Type and Th2-Type Cytokine Responses.
Numerous factors, including alterations in the number or type of infections early in life, the widespread use of antibiotics, adoption of the Western lifestyle, and repeated exposure to allergens, may affect the balance between Th1-type and Th2-type cytokine responses and increase the likelihood that the immune response will be dominated by Th2 cells and thus will ultimately lead to the expression of allergic diseases such as asthma.

allergens, may increase the risk of asthma-related morbidity,[64] respiratory arrest during exacerbations of asthma,[66] and, perhaps, of the development of asthma.[4,63]

AIRWAY REMODELING IN ASTHMA

The rate of decline in lung function with age is greater in adults with asthma than in those without asthma,[67,68] and the ability to reverse the impairment in pulmonary function in many patients with asthma depends on the early recognition and treatment of the condition.[69-71] Remodeling entails thickening of the airway walls, with increases in submucosal tissue, the adventitia, and smooth muscle.[72-74] These features differ in asthma and chronic obstructive pulmonary diseases,[72] in allergic and nonallergic asthma,[75] and with the severity of asthma.[72] The precise mechanisms underlying the remodeling process are under intense study. Recent observations in children with asthma (age, 5 to 12 years)[59] suggest that preventing the progressive loss of lung function in childhood may require recognition and treatment of the disease during the first five years of life.[76] Whether there is a mechanistic link between this loss of airway function and structural remodeling of the airway in early life is not yet known.

THERAPY

In the past decade, the treatment of asthma has emphasized long-term suppression of airway inflammation plus relief of symptoms with quick-acting bronchodilators (primarily aerosolized beta-agonists).[77] Inhaled corticosteroids are the most effective agents available for the symptomatic control of asthma and improvement in pulmonary function,[78] but their potential side effects when used in escalating doses have led to the use of adjunctive therapies.[79,80] Concomitant treatment with long-acting beta-agonists,[81-83] theophylline,[84] and leukotriene antagonists[85] have all been shown to help control asthma while minimizing the doses of inhaled corticosteroids that are needed.

Nevertheless, whether used alone or in combination with other therapies, corticosteroids do not consistently abrogate airway inflammation in patients with asthma.[86-88] For this reason, other approaches that modulate IgE-associated immunologically mediated inflammatory responses are in use or under development. Conventional allergen immunotherapy can be effective in many,[89] but not all,[90,91] patients. DNA vaccines and other molecular methods of down-regulating antigen-specific Th2-mediated responses are

currently being studied.[92,93] Agents directed at diminishing the production of IgE through effects on interleukin-4 or on IgE itself have also been evaluated. One such compound is a soluble recombinant interleukin-4 receptor that can be delivered in nebulized form.[94] Single-dose studies of this agent in patients with moderate asthma have demonstrated its short-term safety and efficacy after the withdrawal of inhaled corticosteroids.[94] Another compound, a recombinant humanized monoclonal antibody that forms complexes with free IgE (rhuMAB-E25, or omalizumab), blocks the interaction of IgE with mast cells and basophils. In early clinical investigations, this antibody attenuated the early-phase and late-phase airway-obstructive response to challenge by allergen and suppressed the accumulation of eosinophils in the airway.[95,96] Subsequent evaluations have shown that regular intravenous administration of this preparation to patients with moderate or severe allergic asthma can control symptoms better than does a placebo and permit a clinically significant reduction in the dose of oral and inhaled corticosteroids.[97] The efficacy of these therapies emphasizes the important contribution of allergic inflammatory mechanisms in the pathophysiology of asthma in many patients.

CONCLUSIONS

Our concept of the mechanisms of asthma has changed dramatically in the past decade. With the recognition that immunologically mediated responses, in particular those involving IgE-dependent mechanisms, are integrally linked to the development of airway inflammation and hence the inception, persistence, and severity of disease, treatment of asthma is now being directed principally toward these factors. Moreover, more focused research into these immunologically mediated inflammatory mechanisms has facilitated the development of new and exciting therapies. The next decade holds promise for further advances as the genes associated with asthma are discovered, their function is defined, and the results lead to more specific therapeutic approaches in individual patients. In view of the rising prevalence of asthma, these advances hold hope for millions of affected patients.

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