THE EOSINOPHIL

Friend or Foe?

Donald Kennerly, M.D., Ph.D.

Internal Medicine Grand Rounds

University of Texas Southwestern Medical Center

January 3, 1991

. . . The presence of high numbers (of eosinophils) associated with parasite infections has led to the widespread assumption that this cell is involved in the defense against such organisms whereas the high numbers found in allergic conditions such as asthma have been interpreted to indicate the protection of the body from the nasty mediators released from mast cells and basophils. . . .

. . . The hypothesis of the eosinophil being a protective cell against mast cell and basophil mediators, however, has been completely revised . . .

P. Venge (Agents and Actions, 1990, 29:1)

OVERVIEW

The eosinophil was first described approximately 110 years ago by Ehrlich. Since that time there has been considerable revision of the prevailing view of its role in health and disease. In this area, as in a startlingly large number of other fields, it appears that authors in the late 19th century were probably more accurate in their perspective than were many authors writing during the 1970's and early 1980's.

This Grand Rounds seeks not to provide a comprehensive description of the many diseases in which the eosinophil is seen in abundance (Appendices 1-5), but rather to review current knowledge about the cell itself and to provide an example of a hypersensitivity state in which the eosinophil appears to play a major role -- asthma.

A significant misconception evolved out of flawed interpretation of data obtained in the early 1970s regarding the role of the eosinophil in allergic processes (and has been promulgated in even several recent reviews). Specifically, it was suggested that the prominence of the eosinophil's presence in the tissues and secretions of patients with asthma and allergic rhinitis was a reflection of its principal role in dampening allergic inflammation by the destruction of inflammatory mediators produced by mast cells (Figure 1). As the result of the work by a number of laboratories we have learned that the eosinophil contains potent toxic molecules and can generate large quantities of lipid mediators that together almost certainly contribute in a *proinflammatory* fashion to the pathophysiologic evolution of these illnesses. Indeed, the currently prevailing view of the eosinophil is that it is a potent effector cell that is able to importantly participate in host defense in helminthic infections, but which contributes importantly to destructive inflammation when it is called into action in hypersensitivity states.

HISTORY

In 1879 Dr. Paul Ehrlich described the existence of eosinophils. In his experiments he determined that a small fraction of circulating granulocytes avidly bound the dye eosin (a highly negatively charged fluorescein derivative) in such a way that the granules of this cell exhibited intense staining -- hence the name *eosinophils* (eosin loving). We now know that the intensity of this staining is due to the presence in the large secretory granules of eosinophils of highly positively charged proteins (described in detail in a subsequent section). Ehrlich's staining procedure for eosinophils allowed the demonstration of eosinophil infiltration in the tissues of a number of disease processes. For example, the presence of eosinophils in asthma was demonstrated in 1889 and the important presence of eosinophils associated with trichinosis was shown in 1898. Paul Ehrlich, known for his identification of and brilliant proposals regarding the role of mast cells and basophils, stunningly accurately portended in 1900 the role of eosinophils and tissue-derived chemotactic stimuli in a variety of pathologic processes involving eosinophil infiltration.

An increasing number of descriptive findings were published during the next half century, but after 1960 the ability to employ the emerging tools of biochemistry and cell biology led to increasingly rapid progress. From 1960 to 1975, laboratories described the existence of lymphocyte/T-cell factor(s) that promoted the growth and differentiation of eosinophils. The purification of the major basic protein from rodent eosinophils and it's demonstration of important cytotoxicity towards helminths and normal cells was also demonstrated during this period. Although initial attempts at *in vitro* growth of eosinophils had been successful, the ability to generate significant numbers for study could not be accomplished.

Although Ehrlich envisioned the eosinophil as an important proinflammatory cell in host defense and hypersensitivity reactions, increasing attention during the 1970s focused on the hypothesis that the eosinophil was important in down-regulating inflammation induced by mast cells (Figure 1). That view gained support by the early demonstrations that the eosinophil contained enzymes capable of destroying inflammatory mediators released by mast cells: histaminase (capable of destroying histamine, a prominent mast cell-derived mediator), aryl sulfatase B (an enzyme with minimal capacity to destroy the as yet structurally uncharacterized slow reacting substance of anaphylaxis, SRS-A) and phospholipase D [PLD; capable of destroying platelet activating factor (PAF)]. The emergence of the concept that increases in mast cell cAMP by pharmacologic agents could block exocytosis coupled with the observation that eosinophils could be stimulated to synthesize PGE_1 and PGE_2 (which can increase cellular cAMP), it was natural to propose that eosinophils either

FIGURE 1

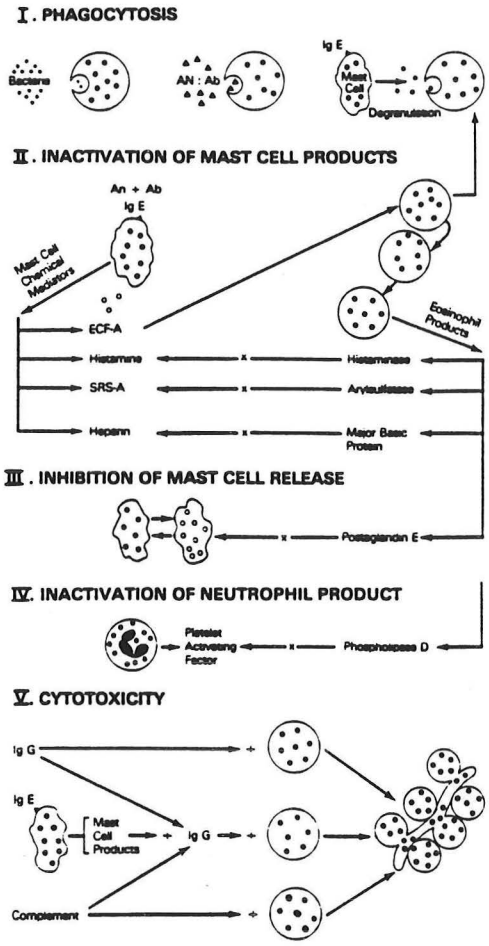


Fig. 35-5. Schematic representation of the major functional capabilities of the eosinophil (see text for discussion).

destroyed mast cell-derived mediators or "turned off" mast cells. It was not until the full cytotoxic potential of eosinophil granule proteins and the ability of eosinophils to synthesize impressive amounts of PAF and LTC_4 in response to stimulation became known that a proinflammatory role for eosinophils began to take hold. Quantitative arguments regarding the limited capacity of these mediator destructive pathways (discussed in more detail in subsequent sections) led increasingly to acceptance of the view that the eosinophil contributes to inflammation and does not dampen allergic inflammation to any great extent. With an increasing appreciation that asthma involves a vigorous cellular inflammatory response, the eosinophil has taken on, as predicted by Ehrlich, a role as an important proinflammatory cell likely involved in generating and/or perpetuating pathologic inflammation.

CELL BIOLOGY OF THE EOSINOPHIL

Morphology

The eosinophil is a circulating granulocyte that is slightly larger than a neutrophil, being 12-17 microns in diameter and, as described earlier, contains abundant large granules staining intensely with eosin. Even in poorly stained specimens, eosinophils are reasonably easy to discern by the large size and refractile nature of their granules compared to those of neutrophils and basophils. The nucleus is nearly always bilobed

Staining of the eosinophil is accomplished successfully using a Wright, Giemsa or Hansel stains, but several peculiarities of staining deserve description. When examining urine to assess possible existence of eosinophiluria, low pH often impairs the ability to stain eosinophils using the Wright procedure encouraging the use of other procedures. In blood smears, staining is easily accomplished, but considerable variation in the frequency of eosinophils is observed in different parts of the slide depending on the thickness of the area being examined. This difficulty accounts for much of the variability often observed in the frequency of the eosinophils on the differential analysis of repeated CBCs that cannot be explained by intercurrent administration of systemic glucocorticoids or epinephrine. Because of this difficulty, many clinical laboratories offer an *absolute eosinophil count*, obtained by vital staining and counting in a hemacytometer, rather than simply utilizing the less desirable method of obtaining the product of WBC count and the frequency of eosinophils on differential analysis.

Electron microscopy of the eosinophil reveals the striking presence of an abundance of large granules containing an electron dense core and more electron lucent matrix (schematic illustration in Figure 2). Close examination of this core demonstrates a lattice structure which has been demonstrated to consist of precipitated major basic protein (MBP; Figure 3). Other toxic cationic proteins in eosinophil secretory granules are principally located in the granule matrix. In addition to these large granules, small, fairly electron dense granules can also be demonstrated. Lipid bodies (accumulations of lipid without a surrounding membrane) are also observed fairly frequently in eosinophils. As will be described in greater detail subsequently, the partially activated or "primed" eosinophils have a lower density (density 1.070-1.082) and can be distinguished morphologically from normodense eosinophils (density 1.082-1.095) usually by the presence of one or more vacuoles and fewer granules.

FIGURE 2

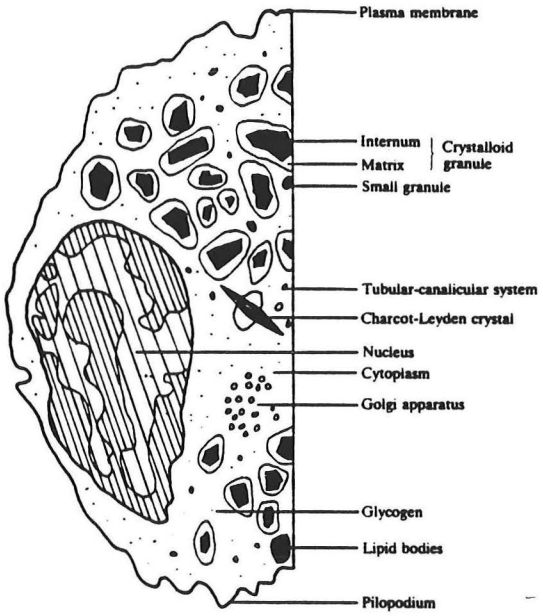
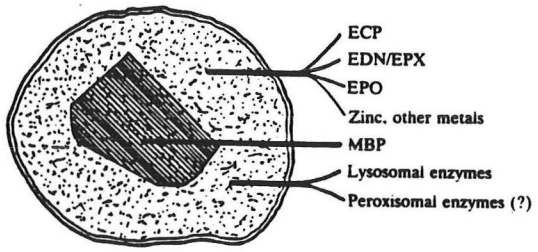


FIGURE 3

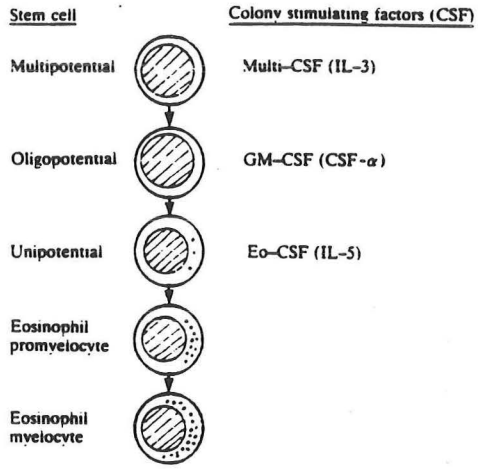


Eosinophil Growth and Differentiation

Facilitating our knowledge with regard to the role of the eosinophil has been an increasing understanding of the factors that regulate both the production of bone marrow precursors and promote eosinophil differentiation in and release from the bone marrow. Specifically, the purification, cloning, sequencing and expression of a variety of colony stimulating factors (CSFs) and interleukins (ILs) have permitted a number of critical experiments that have led to the identification of those factors of greatest importance in the production of mature eosinophils (Figure 4).

Eosinophils are derived from the bone marrow as a result of the replication and differentiation of the CFU_{eo} which appear to be independent of the neutrophil

FIGURE 4



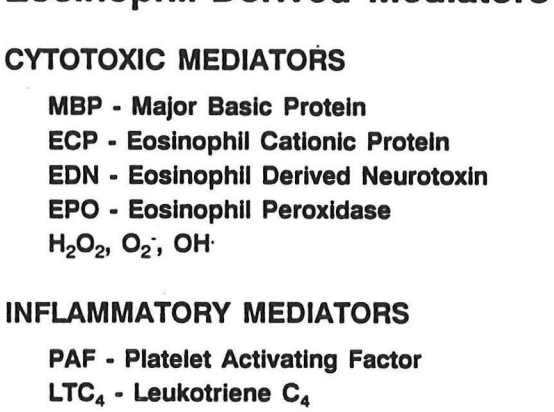
lineage. Stimuli which increase the presence of eosinophil precursors in the bone marrow include GM-CSF and IL-3. Differentiation of eosinophils is dramatically enhanced by the presence of IL-5 which both increases the number of late eosinophil precursors and increases their differentiation. Studies in humans suggest that it takes approximately 4-5 days for differentiation of eosinophils in the bone marrow. Eosinophils spend a very brief period circulating in the intravascular pool (approximately 8 to 12 hours) prior to entering peripheral tissues. It is estimated that the tissue to vascular ratio of eosinophils is approximately 100:1. The existence of a marginal pool of eosinophils in normal individuals is controversial, although data from patients with the hypereosinophil syndrome suggest the existence of complex kinetics that may involve a marginal pool. Although the bulk of the nonmarrow eosinophils reside in the tissues, one cannot view the eosinophil as a resident cell inasmuch as its lifetime in this compartment is estimated to be brief (approximately 2-10 days). Because eosinophils do not recirculate via the lymphatics and no specific mechanism appears to exist with regard to their removal, it is postulated that eosinophils simply lose viability in the periphery with local dissipation of their products.

Preformed Eosinophil Mediators

The eosinophil is rich in a number of highly positively charged small proteins that are contained within its secretory granules (Figure 5). The most abundant of these include major basic protein (MBP), eosinophil cationic protein (ECP), eosinophil peroxidase (EPO), and eosinophil derived neurotoxin (EDN). These cationic proteins have a variety of different functions which are described below and are unique to the eosinophil. Together they have the potential to inflict very serious injury upon either

normal tissues or invading organisms.

FIGURE 5



Major Basic Protein (MBP) - Figure 6 summarizes the principal characteristics and functional properties know of MBP. It was the first toxic eosinophil protein to the characterized and consists of a single chain of 117 amino acids with the sequence illustrated in Figure 7. As a result of post translational modification, the molecular weight varies, but averages 14 kD. Of considerable interest is that 16% of the amino acids are arginine and that the molecule is intensely cationic with a pI = 11.6. MBP tends to form oligomers/polymers as a result of the spontaneous formation of disulfide bonds with the several free cysteine residues on its surface. Many of the activities of MBP are increased or decreased somewhat when it is subjected to reduction/alkylation. MBP is disproportionately represented in hypodense eosinophils (50^+ pg/cell) whereas in normodense cells, it is present in quantities similar to those of the other eosinophil toxic proteins described subsequently (10-20 pg/cell). As

FIGURE 6

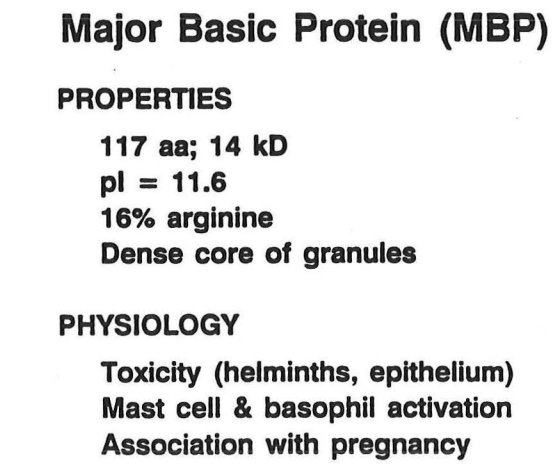


FIGURE 7

FIG. 1. The amino acid sequence of MBP as determined by analyses of the intact protein and from isolated peptides (▬). Sequence obtained from end terminal sequence of the intact protein (⁙⁙). Sequence obtained from peptides generated by cleavage at tryptophan residues by cyanogen bromide (▬▬). Sequence obtained from acetylation of the amino terminus, cleavage at aspartic acid and dilute acid, and analysis of the carboxy-terminal peptide. (From Wasmoen TL, Bell MP, Loegering DA, et al. J Biol Chem 1988;263:12559-63.)

described previously, MBP is present in the electron dense core of the large secretory granules of the eosinophil (Figure 3). The minor presence of MBP in basophils is likely due to secondary uptake by basophils of eosinophil-derived MBP suggesting that MBP in inflammatory sites is the exclusive responsibility of the eosinophil. Curiously, MBP is significantly synthesized by one other cell -- the placental X cell. In fact, circulating levels of MBP increase dramatically during pregnancy and a significant transient increase in plasma MBP occurring just prior to parturition is intriguing. Aside from pregnancy, MBP appears to be essentially exclusively derived from eosinophils.

The recent development of antibodies to human MBP has permitted assessment of the immunohistochemical presence of MBP in a variety of pathological processes including asthma, atopic dermatitis, urticaria, parasitic infection, bullous pemphigoid, the hypereosinophillic syndrome and the eosinophilia-myalgia syndrome, just to mention just a few. This technology is of considerable importance because in several disorders in which eosinophils had not been considered to be involved (because of their relative absence by conventional histologic staining), MBP has been found to be abundantly present (atopic dermatitis and urticaria).

From a functional perspective, major basic protein is able to cause a number of important effects. The ability of MBP to cause toxicity to a variety of cells has been the focus of considerable study. When added to a variety of different parasites *in vitro*, it has been shown to cause toxicity in the low ug/ml range -- levels that would be well within those expected to be achieved *in vivo* in sites of inflammation. In

addition to toxicity to a number of parasites, MBP has also been shown to have considerable toxicity against normal mammalian cells. Pulmonary epithelial dysfunction, damage and denudation seen associated with asthma (*vide infra*) may well be the result of MBP inasmuch as MBP can cause these effects at low ug/ml concentrations (demonstrated to be present in the sputum of asthmatics). At increasing concentrations, MBP causes cilial dysfunction, ciliostasis of the respiratory epithelium in tracheal ring preparations, separation of epithelial cells from the basement membrane and cell death. While MBP has been shown to be toxic to certain tumor cells and present surrounding some malignant neoplasms, a role in immune surveillance has not been established. Electron microscopic studies of MBP-induced toxicity toward parasites shows rapid membrane blebbing of the tegumentary layer suggesting that its effects are on the surface membrane of cells.

MBP also causes a variety of responses in addition to cytotoxicity. Noncytotoxic degranulation of mast cells and basophils with resultant release of inflammatory mediators occurs at modest concentrations of MBP. Neutralization of heparin occurs *in vitro* as the consequence of MBP's positive charge, but *in vivo*, MBP causes an increase in the clotting time by mechanisms that are yet to be defined.

No specific counter-regulatory molecules result in the modulation of MBP-induced phenomenon although heparin with its intense negative charge can complex with MBP. Mast cells and basophils secrete heparin and other intensely negatively charged proteoglycans that may modulate MBP availability and/or activity.

Eosinophil Cationic Protein (ECP) - The properties and major activities of ECP are summarized in Figure 8. It is a family of molecules varying from 16 to 21 kd probably derived from a single gene as the result of post translational modification. Like MBP, it is highly cationic with a pI of greater than 11. Unlike MBP however, ECP is present primarily in the matrix of the eosinophil secretory granule surrounding the dense core (Figure 3). N-terminal sequence analysis demonstrates considerable homology of ECP with pancreatic ribonuclease (RNase) as discussed in the section dealing with EDN.

Like MBP, ECP is a potent toxic agent to a variety of cells. It causes the formation of pores in the surface of cells allowing passage of small molecules and may be the mechanism of its cytotoxicity. In some parasite toxicity assays, ECP has been shown to be 8 to 10 fold more potent than MBP. In addition to local toxicity to host cells and parasites, intrathecal injection of ECP in very small amounts in experimental animals causes a peculiar neurotoxic reaction first observed in 1932 (termed the Gordon phenomenon). This reaction develops over the course of several

Eosinophil Cationic Protein (ECP)

PROPERTIES

- **16-21 kD**
- **pI > 11**
- **Homology with EDN and RNase**
- **Granule matrix (EG-2 vs EG-1)**

PHYSIOLOGY

- **Marked toxicity (parasites, epithelium)**
- **Membrane pores**
- **Mast cell activation**
- **Neurotoxin (Gordon phenomenon)**
- **Plasminogen activation**
- **Reduced lymphoctye proliferation**

FIGURE 8

days to a week and involves forelimb ataxia, muscle weakness and renders the animal unable to stand. Relatively selective loss of Purkinje cells has been demonstrated. The relevance of this phenomenon to human pathology is uncertain.

In addition to cytotoxicity, ECP can activate mast cells, but not basophils. ECP's effects on coagulation include: *i*) a decrease the clotting time by activating Factor XII and *ii*) neutralization of heparin activity (probably as the result of ionic interaction). ECP has also been shown to cause a reduction in lymphocyte proliferation in both one way MLR and during mitogen-induced activation.

The presence of ECP in pathologic reactions can be interestingly assessed by the use of two monoclonal antibodies. EG-1 reacts with ECP that is either contained in the secretory granule or has been released into the extracellular environment. EG-2, however, reacts selectively with ECP that has been released as the result of eosinophil secretion.

Eosinophil Derived Neurotoxin (EDN) - Figure 9 demonstrates that, like the other eosinophil granule proteins, EDN is a cationic protein of 19 kD with a pI greater than 11. It ability to induce the Gordon phenomenon (described above) is its prominent characteristic resulting in its name. EDN is localized to the matrix of eosinophil granules and can be quantitated directly by radioimmunoassay.

Although EDN can also induce direct toxicity to parasites, it appears to be less active than ECP and MBP and has not been shown to be able to cause stimulation

Eosinophil-Derived Neurotoxin (EDN)

PROPERTIES

18.6 kD
pI > 11
Matrix of granules

PHYSIOLOGY

Moderately toxic
Neurotoxic (Gordon phenomenon)
RNase homology and activity

FIGURE 9

of mast cells or basophils. Perhaps it functions in a "backup" capacity with respect to the other cytotoxic eosinophil proteins, but it may have important functions that remain to be described. Of interest is that EDN has considerable N-terminal sequence homology to pancreatic ribonuclease (RNase). While both ECP and EDN both have RNase activity in *in vitro* assays, EDN is considerably more active. The contribution of RNase activity to inflammation foci is at best uncertain. RNase activity does not contribute to EDN's or ECP's abilities to cause the Gordon phenomenon inasmuch as intrathecal injection of pancreatic RNase fails to induce an neurologic changes.

Eosinophil Peroxidase (EPO) - Unlike the other cationic proteins present in the eosinophil granule, EPO is a heterodimeric molecule with a total molecular weight of approximately 70 kd (Figure 10). Like the other proteins, it is highly positively charged with a pI > 11. EPO is abundant in the matrix of eosinophil secretory

Eosinophil Peroxidase (EPO)

PROPERTIES

Heterodimer; 67-77 kD
pI > 11
Matrix of granules
Distinct from PMN myeloperoxidase
Reaction: Halide$^-$ + H_2O_2 ---> HOHa

PHYSIOLOGY

H_2O_2-dependent toxicity
Adheres to parasites and MC granules
Mast cell activation (± toxicity)
Inactivation of leukotrienes

FIGURE 10

granules (approximately 15 pg/cell). Although EPO is distinct from neutrophil myeloperoxidase, it is similarly able to catalyze the formation of hypohalous acids in the presence of hydrogen peroxide (H_2O_2) and appropriate halide ions (Figure 10). EPO is not only capable of generating hypochlorous acid (HOCl), but can also vigorously form hypobromous acid (HOBr) and hypoiodous acid (HOI) in the presence of very low concentrations of Br^- and I^-, respectively. Toxicity to parasites and mammalian cells by these compounds is due to oxidative injury cased by hypohalous acids, not due to acidity. Considerably greater EPO-induced toxicity is observed when I^- or Br^- are included at very low concentrations (uM range). Because eosinophils are not very effective as phagocytes, the ability of EPO to generate toxic hypohalous acid-mediated destruction of bacteria is viewed to be modest compared to neutrophils. The ability to generate toxicity depends on the genesis of H_2O_2) which is produced by the respiratory burst of activated eosinophils and other inflammatory cells.

The highly positively charged nature of EPO makes it able to adhere tightly to the surface of parasites and target cells, rendering them sensitive to the subsequent generation of H_2O_2 by eosinophils or other cells. EPO binds tightly to mast cell granules (which contain highly negatively charged proteoglycans). This is of importance because the EPO/mast cell granule complex is even more potent than free EPO in stimulating mast cell activation. Although EPO-induced mast cell exocytosis is H_2O_2 dependent, it is curiously a Ca^{+2}-dependent noncytotoxic process at low concentrations of EPO but at higher concentrations of MBP, ECP and EPO, mast cells can release inflammatory mediators as the result of direct cytotoxicity.

EPO has been shown *in vitro* to cause H_2O_2-dependent destruction of leukotrienes and may represent at least one eosinophil mechanism that might contribute to down regulation of inflammation although its quantitative relevance in leukotriene metabolism is not firmly established.

Other Eosinophil Enzymes - A variety of other enzymes have been shown to be present and/or released as a result of exocytotic stimuli and several are of particular historical interest with regard to the evolution of our understanding of the participation of the eosinophil in disease processes.

Eosinophil lysophospholipase has the properties described in Figure 11. It is localized to the plasma membrane and has been shown to crystallize to form hexagonal bipyramids identical to Charcot Leyden crystals (CLC) and the CLC protein has identical electrophoretic properties of the eosinophil lysophospholipase. Since the CLC protein is present in significant quantities in the plasma membrane

Eosinophil Lysophospholipase (CLC)

FIGURE 11

PROPERTIES

CLC protein - hexagonal bipyrimids
Plasma membrane associated
Present also in basophils
lyso-PC ---> G-P-choline + FA

PHYSIOLOGY

Destroys endogenous lyso-PL

of basophils, it cannot be viewed as specific for eosinophils. The vigorous ability of eosinophils to destroy lysophospholipids by the abundant presence of lysophospholipase may reflect a protective mechanism against the lytic properties of lysophospholipids. The enzyme producing lysophospholipids, phospholipase A_2 (PLA_2), has been shown to be increased in some inflammatory foci including synovial fluid in rheumatic conditions. Thus, since lysophospholipids may be increased in the milieu in which the eosinophil is called upon to act and, indeed, because the eosinophil forms them endogenously as the result of the PLA_2-mediated release of arachidonic acid for the synthesis of inflammatory eicosanoids (*vide infra*), the rich presence of lysophospholipase in the plasma membrane of eosinophils may be in an attempt at self-preservation in a hostile environment.

Aryl sulfatase B -- an enzyme that catalyzes removal of sulfate residues -- was discovered to cause slow destruction of SRS-A activity. These observations came prior to structural knowledge regarding the sulfidopeptide leukotrienes. Aryl sulfatase B was localized to the small secretory granules of eosinophils. An additional "mediator destroying" enzyme, histaminase, was shown in early studies to be present in eosinophils. These two early observations (at a time when the toxic activities of MBP were only beginning to emerge) led to advancement of the hypothesis that the eosinophil was principally involved in down regulating the inflammatory effects of mast cells. Additionally, phospholipase D (PLD) was shown to be associated with eosinophil granules. Because PLD was shown to be able to abolish the activity of PAF (again prior to the knowledge of the structure of PAF) *in vitro*, PLD activity was taken as support that the eosinophil down regulated the proinflammatory activity of mast cell-derived mediators. Unfortunately, subsequent studies demonstrating the almost trivial activity of eosinophil histaminase (compared to the amount of histamine present in mast cell granules), the genesis of LTC_4 by eosinophils (not its destruction by aryl sulfatase B) and a principal role of PAF acetylhydrolase in PAF destruction

(rather than PLD) were only slowly able to erode the popular hypothesis that eosinophils were responsible for reducing mast cell-initiated inflammation. This evolution serves as an important reminder to investigators that the *existence* of sought after enzymatic activities in any given cell is not alone sufficient to fully support the importance of a particular enzyme or pathway in the principal functional activities of the cell of interest. Although quantitative assessments of these pathways are often difficult to accomplish, they are critical to a thorough evaluation of the role of any given process.

The presence of PLD in the eosinophil granule may be of critical importance to the signaling mechanisms involved in eosinophil activation. Recent studies by a number of laboratories indicate that receptor stimulation causes activation of PLD-mediated hydrolysis of phosphatidylcholine (PC) (reviewed in the JBC by Exton, 1990). Indeed, Billah's group has recently demonstrated C5a-induced activation of PLD activity in eosinophils. The importance of this pathway lies with the ability of PLD to catalyze the formation of phosphatidic acid (PA). Although the role of PA as a second messenger is controversial, it is converted to both 1,2-diacylglycerol (DG; a second messenger that is well known to activate protein kinase C) and lyso-PA (that in recent reports has been shown to be able to liberate Ca^{+2} from intracellular sites and to be mitogenic). Our own studies in mast cells suggest that phospholipase D (PLD) activation prominently takes place seconds after Fcϵ-RI stimulation and its presence in secretory granules (unpublished observations) suggest that it may be important in the signal transduction and/or directly by promoting the membrane fusion process necessary to exocytosis.

Newly synthesized mediators

Products of the respiratory burst - Like neutrophils, activation of eosinophils causes a respiratory burst with the reduction of molecular oxygen by NADPH to form superoxide (HO_2) and its anion (O_2^-), hydrogen peroxide (H_2O_2) and hydroxyl radicals ($OH^\cdot$). These agents can cause direct oxidative toxicity to a variety of cellular constituents. In the presence of EPO and halide anions, H_2O_2 can be converted to highly toxic hypohalous acids (*vide supra*) which themselves are potent oxidative toxins.

Arachidonic Acid Products - The eosinophil can produce a variety of eicosanoid products, principally of the lipoxygenase lineage, in response to appropriate stimulation (opsonized particles, PAF, fMLP and ECF-A, for example). The principal inflammatory eicosanoid is the sulfidopeptide leukotriene LTC_4 produced by activation of the 5-lipoxygenase pathway. LTC_4 can be converted, by sequential

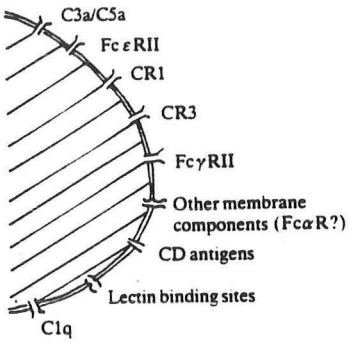
removal of amino acids of the glutathione side chain, to LTD_4 and LTE_4. LTC_4 is a potent bronchoconstrictive agent that is able to effectively increase mucus secretion -- effects that are likely important in the pathophysiology of asthma. Of less certain importance, but considerable interest, is the presence of 15-lipoxygenase activity in eosinophils which results in the formation of 15-HPETE and 15-HETE. The latter two mediators have been suggested potentially to be important in modulating eosinophil activation, particularly with regard to LTC_4 synthesis, although this concept is still evolving. Of historical interest was the early demonstration of the ability of the eosinophil to synthesize PGE_2 and PGE_1 -- agents that were thought to be important in down regulating mast cells by increasing cellular cAMP. As with other observations that suggested that eosinophils were responsible for dampening the effector functions of mast cells, it ultimately became clear that the amounts of PGE_2 and PGE_1 that were generated by eosinophils were modest and likely did not significantly alter the function of mast cells.

Platelet Activating Factor (PAF) - Stimulation of eosinophils results in a dramatic increase in the formation of PAF. This is a potent proinflammatory agent that is able to cause bronchoconstriction, chemotaxis of granulocytes and the development of nonspecific bronchial hyperreactivity in subjects treated by inhalation challenge [the diverse effects of this inflammatory mediator were reviewed in my last Grand Rounds protocol (1989)]. Of particular interest in this context is that PAF is not only generated by eosinophils, but it is able to cause, at low concentrations, eosinophil chemotaxis, priming and secretion. From a quantitative perspective, a stimulated eosinophil can produce sufficient PAF to cause an average PAF concentration of 100 nM within a sphere with a diameter of 10 cell diameters -- a concentration easily capable of causing profound eosinophil effects. Thus, the potential for causing a self stimulating positive feedback loop is considerable and demands an investigation into what regulatory forces prevent unregulated eosinophil recruitment, activation and subsequent inflammation.

Receptor Repertoire

Eosinophils possess a variety of cell surface receptors demonstrated either *i*) by virtue of their physical presence using appropriate monoclonal antibodies or ligand binding assays or *ii*) by their functional presence as measured by cellular responses as the result of adding an appropriate ligand which are assumed to interact with specific receptors (Figure 12). The eosinophil can interact with a number of antibody molecule Fc receptors including those for IgG, IgE and, more recently, sIgA. For IgE and IgG, interactions are through low affinity receptors (Fc-gamma-R2; Fcϵ-R2) although the nature of the interaction of the sIgA has not been characterized.

FIGURE 12



Interaction of eosinophils with particles bearing free immunoglobulin molecules of the appropriate isotype sets into motion a variety of responses. For small particles coated with IgG, "primed" eosinophils (*vide infra*) take up the particles by phagocytosis. For large particles, eosinophils become tightly adherent to the particle and are activated to synthesize LTC_4, have a respiratory burst and undergo exocytosis of preformed granules directed toward the Ig bearing surface. Because eosinophils express relatively low affinity Fc receptors and are principally activated by interaction with surfaces that are highly substituted by immunoglobulin suggests that the eosinophil's role is to destroy opsonized particles to which a humoral immune response has been generated. This is in contrast to the mast cell and basophil which require only a divalent crosslinking of IgE receptors to achieve exocytosis. Thus, the mast cell represents a full "armed" cell that requires only exposure to soluble proteins from organisms, while the eosinophil is an effector cell that most effectively interacts with cells/pathogens that have: *i*) induced sufficient inflammation to recruit eosinophils and *ii*) have been coated with either specific antibodies or bear C3 or $C3b_i$ as the result of activation of the alternative pathway of complement (*vide infra*).

In addition to antibody receptors, the eosinophil interacts effectively with complement components. Specifically, eosinophils possess CR1 and CR3 receptors which bind the ligands C3b and $C3b_i$. When C3 is activated by either the direct or alternative pathway of complement activation, C3b becomes covalently linked to the surfaces of cells by a displacement reaction involving membrane proteins and a internal thioester in C3. $C3b_i$ is an inactive proteolytic fragment of C3b that remains covalently linked to the cell surface. Thus, pathogens or cells that elicit a humoral immune response can activate eosinophils either by interaction directly with eosinophil Fc receptors or, should the immune response involve either IgM or appropriate IgG subtypes, complement receptors in addition. Immune complexes that activate complement can activate eosinophils. In addition to CR1 and CR3 receptors,

eosinophils respond to C5a (a small soluble fragment of C5 released by complement activation termed an anaphylatoxin as the result of its ability to cause anaphylactic reactions when administered to animals) by both chemotaxis and exocytosis.

Although studies have demonstrated the presence of IL-5 receptors on eosinophils, other receptors have also been implicated as the result of cellular responses to physiologically relevant agents: fMLP (formyl-methionyl-leucyl-phenylalanine; a bacterial product causing chemotaxis of PMNs and activation of all granulocytes); IL-3; granulocyte/macrophage colony stimulating factor (GM-CSF) and mast cell-derived eosinophil chemotactic factors of anaphylaxis (ECF-A; tetrapeptides that are more important in priming eosinophils *in vivo*, but may not be important in eosinophil chemotaxis).

The expression of cell surface receptors on eosinophils is not static. Eosinophil priming (*vide infra*) results in a very significant increase in the surface expression of a number of these receptors. For example, one study has shown that unstimulated normodense eosinophils have fewer than 100 IL-5 receptors/cell but after priming, eosinophils express more than 10 fold more receptors/cell.

Eosinophil Priming

Cell activation may be defined by a variety of functional end points and in the case of the eosinophil, exocytosis or priming or both. The priming phenomenon (termed activation by some investigators) is usually defined as the enhanced ability of a population of eosinophils to *i*) cause cytotoxicity when they are subsequently added to an antibody coated target cell or *ii*) release granule contents when they are exposed to IgG coated beads. The spectrum of effects of eosinophil priming are summarized in Figure 13. The eosinophil's ability to deliver highly toxic molecules to the surface of invading organisms (such as during helminthic infection) depends in part on appropriate opsonization but, in addition to that requisite, the state of readiness of an eosinophil to respond is important and depends upon priming. Since

FIGURE 13

EOSINOPHIL ACTIVATION

Density shift (hypodense)
Enhanced ADCC
Increased Fc receptors (IgG, IgE, ?sIgA)
Increased IL-5 receptors
Increased C3b receptors

priming involves the increased surface expression of both complement and Fc receptors, it is easy to imagine that priming would be important to the effectiveness and extent to which an eosinophil can form a tight interaction with a target cell/organism and to be activated to undergo exocytosis.

Although priming can be described from the functional or quantitative perspectives described above, a morphologic association is also useful. Primed eosinophils (eosinophils that are obtained from patients with the hypereosinophil syndrome or those obtained in peritoneal exudates of experimental animals) also demonstrate morphologic differences. As shown in Figure 14 distinctly different densities are observed in resting eosinophils (normodense; density > 1.082) compared to those obtained from hypereosinophil syndrome patients (hypodense; density <1.082). In addition to being more functionally active [increased surface expression of a variety of receptors and enhanced ability to express antibody dependent cellular cytotoxicity (ADCC)], hypodense eosinophils examined by electron microscopy have somewhat fewer granules and demonstrate the presence of small vacuoles.

FIGURE 14

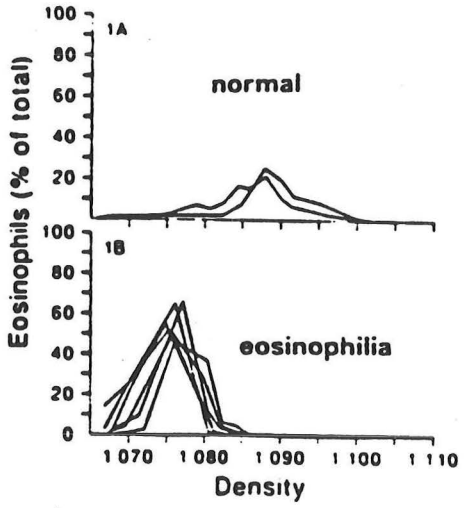


Figure 15 represents a partial list of agents that can cause eosinophil priming. It is not surprising that, for the most part, these represent soluble agents that will be present in inflammatory foci -- serving to fully prepare the eosinophil for its effector role once it encounters an appropriate target. Figure 16 illustrates the concentration dependent ability of IL-5 to cause enhanced release of EDN from eosinophils exposed to a variety of agents covalently substituted on the surface of sepharose beads.

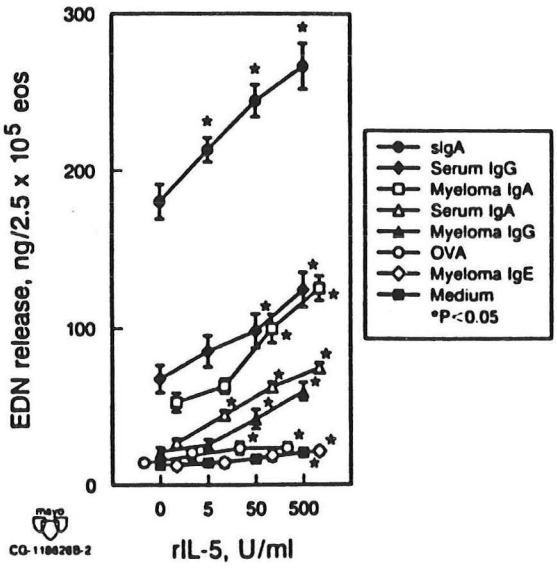
FIGURE 15

EOSINOPHIL ACTIVATION

- INDUCERS OF "PRIMING" -

IL-3
IL-5
GM-CSF
TNF
PAF (low concentrations)
ECF-A
Histamine
Parasite-derived factors
fMLP

FIGURE 16



Eosinophil Exocytosis

The ability of ligands to induce exostosis is essential to delivering the contents of the secretory granules to appropriate targets. As described above, interaction with large surfaces substituted with IgG, IgE, sIgA or C3 fragments can induce exocytosis -- particularly in primed eosinophils (summarized in Figure 17). However, in addition to the short range "hand to hand combat" that the eosinophil wages by virtue of its tight association with opsonized cells (± associated complement), the eosinophil can be induced to undergo exocytosis by soluble agents and, in so doing, contribute to a

FIGURE 17

EOSINOPHIL SECRETION

- SURFACE-DEPENDENT -

IgG
C3b
$C3b_i$
sIgA
IgE

less target-directed increase in the inflammatory milieu. Figure 18 lists physiologically relevant soluble agents known to cause eosinophil exocytosis.

Of interest is that preferential release of some granule constituents has been reported -- IgG sepharose induces a selective release of the eosinophil cationic protein (ECP) while IgE sepharose tends to cause preferential release of eosinophil peroxidase (EPO). Whether this is an *in vitro* artifact or occurs as the result of selective secretion of granules particularly enriched in one or more eosinophil granule proteins is uncertain. The relevance of these differences to the evolution of pathological process is unknown.

FIGURE 18

EOSINOPHIL SECRETION

- SOLUBLE ACTIVATORS -

C5a
PAF (higher concentrations)
----
fMLP
Substance P
GM-CSF
IL-5

Of focal importance are the many roles of PAF in eosinophil function. At low concentrations (low nanomolar range) PAF is a potent chemotactic agent and can prime eosinophils while at higher concentrations (mid to high uM range; likely to be achieved in many inflammatory sites) it can cause exocytosis and indeed, the

additional synthesis by eosinophils of PAF. Thus, the potential to develop a positive feedback loop exists that seems likely to be of considerable importance in amplifying and/or perpetuating eosinophil-mediated inflammatory reactions. [Eosinophil stimulation ----> PAF synthesis ----> eosinophil chemotaxis, priming and stimulati ----> etc.). The inability to intervene in this self sustaining reaction may, in some way, be important to the chronic inflammation associated with some allergic conditions in which the eosinophil is important (asthma, atopic dermatitis, chronic urticaria).

Eosinophil Chemotaxis

The presence of eosinophils in sites of allergic inflammation and of helminthic infection has been noted histologically for nearly 100 years when it was first suggested that circulating eosinophils were recruited into the tissues by chemotactic factors. Compounds causing eosinophil chemotaxis are structurally and physiologically diverse (Figure 19) and likely represent important effector roles of eosinophils in inflammation developing as the result of a variety of immune and nonimmune processes. Significant concern exists regarding the physiologic relevance *in vivo* of ECF-A and histamine as chemotactic agents.

FIGURE 19

Eosinophil Chemotaxis

PAF
C5a
Tissue from disease states

Histamine
ECF-A

Agents documented to cause chemotaxis *in vivo* as well as *in vitro* include PAF and C5a. PAF causes chemotaxis of both neutrophils and eosinophils in the low nanomolar range -- a concentration that seems quite likely to be generated by the many cells capable of synthesizing PAF upon appropriate stimulation (for details, please see my 1989 Grand Rounds protocol). The anaphylatoxin, C5a (a proteolytic cleavage product C5 during complement activation), is a potent chemotactic agent for eosinophils and, like PAF, can cause the accumulation eosinophils when injected intracutaneously. Although not well characterized, extracts of a variety of helminthic parasites can also cause vigorous eosinophil migration. 5-HPETE, 5-HETE and

LTB_4 cause eosinophil migration at low concentrations *in vitro*, but the roles *in vivo* of 5-HETE and 5-HPETE are uncertain inasmuch as studies *in vivo* are lacking. Injection of LTB_4 into skin sites principally causes PMN infiltration.

Probably the most controversial chemotactic agents examined to date are the tetrapeptide eosinophil chemotactic factors of anaphylaxis (ECF-A; ALA-GLY-SER-GLU and VAL-GLY-SER-GLU) released during mast cell exocytosis. Although these agents have been shown *in vitro* in Boyden Chambers to cause eosinophil chemotaxis, their role *in vivo* has never been satisfactorily demonstrated. Since these peptides appear important in the priming of eosinophils, it seems likely that a role in eosinophil activation rather than chemotaxis is likely for these mast cell-derived tetrapeptide "ECFs".

Histamine, which is also capable of priming eosinophils, has in some reports (involving *in vitro* assessments) been described to be chemotactic, but has also failed to cause *in vivo* eosinophil infiltration. Thus, like ECF-As, histamine may have a role to enhance chemotaxis induced by other agents by priming eosinophils.

Summary

Taken together, data support the view that the eosinophil is a potent effector cell involved in tissue inflammation. It can respond to low concentrations of chemotactic stimuli generated by both immune inflammation (C5a from antigen-specific antibody-induced complement activation; PAF produced by IgE sensitized macrophages and type II pneumocytes, mast cells and eosinophils) and nonimmune inflammation (PAF from injured endothelial cells; C5a generated by the alternative pathway of complement activation). The ability of eosinophils to respond both chemotactically and biochemically (priming and/or exocytosis) to a diverse series of mediators released by a variety of resident and inflammatory cells support the involvement of the eosinophil in a number of processes in which it has been shown to be present histologically or in which tissue deposition of MBP or ECP has been demonstrated. Its specific role once it has been recruited into an area of inflammation has shifted from the view that it opposes inflammation to one where it is seen as a potent killer cell producing a wide array of redundant cytotoxic compounds capable of reeking havoc on invading cells and/or normal tissue in disease states characterized by eosinophil infiltration.

ROLE OF THE EOSINOPHIL IN ASTHMA

Overview

As described above, the last 10 to 15 years has seen a very significant change in our view of the role of the eosinophil in allergic inflammation. This section seeks to use asthma as a model of human allergic inflammation to examine the participation of the eosinophil. Unfortunately, most animal models of chronic asthma are seriously flawed and there are thus far no human diseases associated with eosinophil deficiency (or serious dysfunction) in which to assess the impact of the loss of eosinophil to the evolution of diseases in which the eosinophil is proposed to be important.

We currently view asthma as a disease of self perpetuating cellular inflammation (please see my Grand Rounds during 1987) and that the eosinophil plays an important role in this process. Observations that support this view are discussed subsequently and include: 1) eosinophils are nearly uniformly present in increased numbers in both the tissues and sputum of asthmatic airways; 2) eosinophils are preferentially increased in bronchoalveolar lavage fluids during antigen challenge of patients manifesting a late asthmatic reaction (the LAR is associated with the development of nonspecific bronchial hyperreactivity -- the *sine qua non* of asthma); 3) eosinophil chemotactic factors are generated as the result of stimuli known to be important in asthma by a variety of resident cells; 4) eosinophils secrete or synthesize potent mediators capable of eliciting *in vitro* many of the pathologic findings associated with asthma; 5) eosinophil-derived mediators are found in the tissues and/or sputum of asthmatics correlated with the severity of disease or as the result of experimental antigen challenge; 6) release of eosinophil-derived mediators occurs *in vitro* in response to secretory agonists observed to either be present in the airways of asthmatics or produced by cells in response to agents known to be acting in asthma; 8) the severity of asthma has often been found to be correlated with the degree of tissue and/or sputum eosinophilia; 9) medications causing a pronounced decrease in the presence and/or activity of eosinophils reduce the severity of asthma.

Eosinophils in Asthma - Observational Studies

Studies performed in the late 19th century demonstrated the presence of eosinophils in the sputum and airways of patients with asthma. Although the vascular compartment represents a relatively small portion of the total eosinophils in humans (approximately 1%), a number of early investigators noted a correlation of the degree of vascular eosinophilia and the severity of asthmatic symptoms or, more recently, nonspecific bronchial hyperreactivity.

Eosinophils in Asthma - Challenge Studies

With the development of appropriate techniques for bronchoscopy of patients during bronchospasm induced either either naturally or experimentally, increasing insight has developed with regard to the processes taking place during allergen-induced asthma. Figure 20 summarizes the results obtained from a number of studies in which individuals with allergic sensitivity (IgE mediated reactions contributing to asthma) were challenge with relevant aeroallergens. During bronchoscopy saline is introduced after either placebo or antigen inhalation challenge and the presence of cells and/or mediators in the resulting bronchoalveolar lavage fluid assessed. In virtually all patients one can demonstrate an antigen-dependent rapid increase in the presence of mast cell mediators such as histamine, LTC_4 and PGD_2. This is followed by the influx into the airways of neutrophils. Influx of eosinophils and the detection of eosinophil specific products (MBP and ECP) tends to be delayed and less transient than that of neutrophils and is more impressively observed in patients who have late asthmatic responses (LARs). The association of BAL eosinophilia associated with the LAR is not a trivial observation inasmuch as a number of studies have shown that the genesis of a LAR is associated with the development of a significant increase in nonspecific bronchial hyperreactivity (NSBH; the sensitivity of the airway to bronchoconstriction to agents such as methacholine or histamine) which is considered by many to be the best clinical indicator of both the presence and severity of asthma. Thus, association of the antigen-dependent appearance of eosinophils and a LAR is at least consistent with the hypothesis that the eosinophil is causally involved in developing NSBH. As expected, pharmacologic blockade of the LAR is associated with a reduction or failure to observed antigen dependent eosinophil infiltration of the airways.

FIGURE 20

BAL and Antigen Challenge

CELLULAR CONSTITUENTS

- **NEUTROPHILS: 2-8 hours**
- **EOSINOPHILS: 4-24 hours (more during LAR)**

INFLAMMATORY MEDIATORS

- **MBP**
- **ECP**
- **LTC_4**
- **Histamine**
- **PGD_2**
- **Albumin**

Eosinophil Chemotaxis in Asthma

A previous section described agents inducing eosinophil chemotaxis either *in vitro* or *in vivo* (Figure 19). Probably the most important chemotactic factor generated *in vivo* during allergic inflammation is PAF. Because PAF exerts its inflammatory activities in the nanomolar concentration range and is fairly quickly destroyed by an ubiquitous PAF acetyl hydrolase activity (catalyzing the removal of the acetate group at the 2 position resulting in the formation of inactive lyso-PAF), its presence in biologic fluids has been extremely difficult to study. Although flawed from a number of perspectives, the presence of lyso-PAF (1-alkyl,2-lyso-PC) has been used as a tentative indication of the previous presence of PAF that has been destroyed by PAF acetylhydrolase. Preliminary observations suggest that lyso-PAF is present in increased quantity as the result of antigen challenge. Both the type II pneumocyte and mast cell have both been demonstrated to generate PAF as the result of antigen specific IgE dependent activation (by the low affinity and high affinity receptors respectively present on these cells). The recent demonstration of antigen-specific activation of eosinophils by the presence of IgE on low affinity Fc receptors (Fcϵ-R2) provides a direct mechanism for eosinophil activation by antigen. Coupled with (and introduced in a previous section) the ability of eosinophils themselves to produce PAF in response to stimulation, antigen-dependent eosinophil activation of resident eosinophils and their release of PAF generation may be critical in initiating and perpetuating IgE-dependent inflammation in asthma.

Eosinophil Activation in Asthma

In addition to being recruited into an area of active inflammation (in this case in an asthmatic airway challenged with antigen), it is important to evaluate whether recruited eosinophils become activated in the tissues and airways of asthmatics. In previous sections, activators of eosinophils were discussed in greater detail (Figures 15, 17 and 18). Relevant to the focus on asthma, a number of these agonists are likely to be present as the result of antigen challenge and/or chronic antigen-independent asthma.

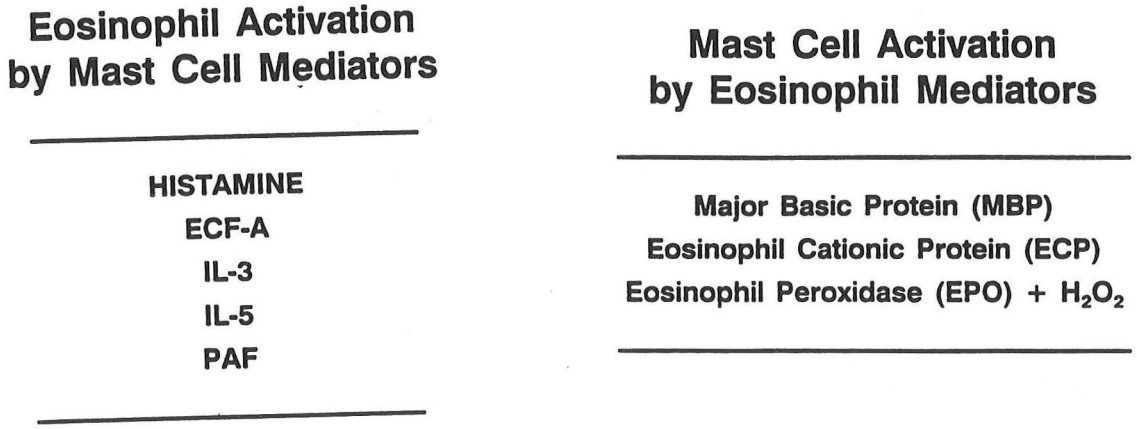
From the mast cell, ECF-A and large amounts of histamine are released in as the result of IgE-dependent mast cell activation -- an event that likely effectively primes resident and recruited eosinophils. Although its release into the extracellular environment is more controversial, mast cells synthesize PAF as the result of antigen stimulation -- a potent activator of eosinophil priming and secretion. Perhaps most excitingly, mast cells have recently been demonstrated to vigorously synthesize and release a number of inflammatory cytokines including IL-5 and GM-CSF which are

potent "primers" of eosinophils and modest secretagogues.

In addition to mast cell products, endothelial cells generate PAF in response to a variety of nonspecific stimuli likely to be present in asthma. Recently, Kay's group has demonstrated the increased presence of $CD4^+$ T cells in biopsy specimens from patients with asthma. The separate observation that circulating T cells from asthmatic patients synthesize increased amounts of IL-5 as the result of IL-2 exposure (compared to nonasthmatic patients) suggests that T cells may indeed have important roles in both antigen dependent and independent asthma in addition to the sythesis and regulation of IgE synthesis.

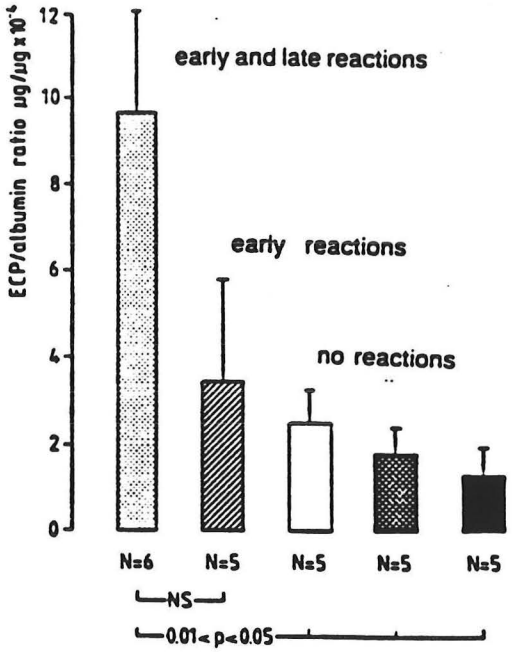
In addition to the ability to activate eosinophils either directly or indirectly as a result of exposure to relevant antigen in IgE mediated asthma, it is quite likely that non-specific stimuli can result in the elaboration of many of these inflammatory mediators capable of activating eosinophils. It is particularly noteworthy that the elaboration of eosinophil products such as EPO, MBP, and ECP can cause dramatic stimulation of mast cell exocytosis with resultant genesis of mediators which can reciprocally activate the eosinophil (Figure 21). This interaction creates the potential for a self sustaining positive feedback loop that may underlie nonIgE-dependent asthma ("intrinsic" asthma). The well known ability of nonspecific stimuli to cause asthmatic exacerbations (URI, irritants, neuropsychiatric, etc.) lends support to the notion that much of asthma need not be the result of an IgE dependent process, but rather the result of a self perpetuating inflammatory reaction that may importantly involve the eosinophil. In addition to the ability of $CD4^+$ T cells to participate in eosinophil activation, these cells release a family of compounds that cause mast cell activation (histamine releasing factors; HRFs) that may modulate these interactions.

FIGURE 21



In addition to these theoretical consideration, biopsy specimens from most patients with asthma demonstrate the robust presence of eosinophil-derived MBP detected by immunofluorescence. Additionally, Figure 22 demonstrates that patients with a dual asthmatic response (early + late asthmatic reactions) as the result of antigen challenge show a dramatic increase in eosinophil-derived ECP in their BAL compared to placebo challenged asthmatics and nonasthmatic patients. In addition to airway findings, plasma MBP and ECP levels have been shown to increase as the result of antigen challenge. Thus, the eosinophil is not only present in the airways, but evidence indicates that it is releasing its granule proteins in the airway and/or bronchial wall.

FIGURE 22



Effect of Eosinophil Products on Airways

The biochemical properties and biologic effects of eosinophil-derived mediators were described in a previous section. Of greatest concern are 3 important effects of the eosinophil derived-mediators on human airways. First, eosinophils elaborate a series of toxic molecules capable of causing dramatic epithelial damage. Major basic protein (MBP) and eosinophil cationic protein (ECP) at low ug/ml concentrations causes a number of toxic effects on the bronchial epithelium *in vitro*. With increasing concentration there is a time dependent ciliostasis, morphologic changes of the epithelial cells (rounding up), detachment from the lamina propria and cell death.

The ability of these toxic eosinophil-derived to cause these responses *in vitro* is particularly noteworthy in light of biopsy studies of patients with even mild to moderate asthma that demonstrate both the presence of MBP and epithelial injury or loss in the epithelium. Studies of the concentration of MBP in the sputum of asthmatics demonstrates that during an exacerbation, levels in the low to mid ug/ml range are, in fact, achieved. Associated with the loss of bronchial epithelium is the removal of an important barrier and clearance mechanism for reducing the exposure of subepithelial immune and inflammatory cells to inhaled antigens. Additionally, epithelial loss exposes type C fibers to the inflammatory contents of the sputum and stimulation may cause antidromic conduction with resultant secretion of inflammatory neuropeptides (substance P and neurokinin A for example) from the neurons of the nonadrenergic noncholinergic (NANC) system. Finally, some data suggest that epithelial cells generate a relaxing factor, the loss of which may result in an increase in bronchomotor tone and perhaps contribute to nonspecific bronchial hyperreactivity.

Second, eosinophil products can contribute to the bronchoconstriction and mucus hypersecretion typical of asthma. The significant elaboration by activated eosinophils of PAF and LTC_4 may be very important along these lines inasmuch as both platelet activating factor and LTC_4 have been shown to be potent bronchoconstrictors and LTC_4 is a powerful stimulator of mucus secretion *in vitro*.

Finally, as described above and, perhaps most importantly, the eosinophil product, PAF, likely acts as a critical proinflammatory mediator as a result of its ability to recruit and/or activate a variety of cells including platelets, eosinophils, neutrophils and mast cells. Other sections describe in detail the richness and likely importance of the cellular inflammatory interactions that take place in chronic asthma. Figure 21 demonstrates the potential role not only of PAF, but also of other mediators from mast cell and eosinophils to create self-sustaining inflammation. Clinical studies involving PAF inhalation have shown not only bronchospasm, but also the generation (and in some cases, long lasting presence) of nonspecific bronchial hyperreactivity. Although there exists some controversy about this finding, PAF is the only mediator thus far shown to be capable of generating non-specific bronchial hyperactivity when administered alone. The proinflammatory potential of this mediator coupled with its likely involvement in asthma has led to vigorous efforts by pharmaceutical firms to develop PAF receptor antagonists -- a task that has turned out to be more difficult than the development of a variety of other classes of antagonists, but one that holds considerable promise in the therapy of asthma in the future.

The Eosinophil's Role in Responsiveness to Pharmacologic Therapy

An important test of the viability of the hypothesis that eosinophils are important in the genesis and/or perpetuation of asthma is whether agents that impair eosinophil activation and/or chemotaxis have an impact on the presence or activity of asthma. Although data obtained using eosinophil depleted rodents (using an anti-eosinophil antibody) are of interest, they primarily deal with acute responses to antigen inhalation and do not adequately reflect the impact on the chronic illness we know as asthma. No specific pharmacologic agents for eosinophils exist for human use, but most clinicians are well aware of the profound eosinopenia generated by the systemic administration of glucocorticoids. Thus, while glucocorticoids will affect a variety of processes, the parallel decline in circulating eosinophils, sputum MBP and clinical improvement of clinical asthma are consistent with the proposed important role for eosinophils.

The ability of glucocorticoids to cause pronounced effects on a variety of essential components of allergic inflammation (GM-CSF, IL-3, IL-4 and IL-5 synthesis, eosinophil chemotaxis and eosinophilopoiesis, for example) encourages an emerging and exciting hypothesis regarding the nature of the defect in individuals who have the propensity for developing disorders characterized by chronic inflammation. Specifically, it is proposed by Sternberg and her colleagues that the inadequate corticotropin releasing factor response during acute inflammation seen in certain strains of rats (with the predicted associated modest and inadequat rise in circulating glucocorticoids) lies at the heart of the documented increased sensitivity of these strains of rats to the development of chronic arthritis induced by adjuvants or other nonspecific stimuli. By analogy, the age old question as to what difference accounts for the differential development of allergic rhinitis *vs* asthma in atopic individuals may, at least in part, be the result of inadequate counter regulatory forces for allergic inflammation in patients with asthma. Dr. Sullivan at UT Southwestern is vigorously examining this hypothesis using a variety of model systems.

The anti-allergic compound cromolyn (and it's more recently developed congener, nedocromil) are less potent than systemic glucocorticoids and also affect a variety of inflammatory cells, but both cause impairment of eosinophil chemotaxis and activation *in vitro* and reduce symptoms and bronchial hyperreactivity in asthmatic patients.

It may well be possible in the near future to examine relatively specifically the impact of eosinophils to the pathophysiology of asthma. Specifically, the identification, purification, cloning and expression of IL-5 along with the development of high affinity monoclonal antibodies to IL-5 will permit the evolution of important

new information regarding the eosinophil in asthma and, indeed, in a variety of processes in which eosinophils may importantly contribute in humans. Because the principal activity of IL-5 appears to be in the growth, differentiation and priming of eosinophils, interfering with its actions would be of considerable clinical value both from investigative and therapeutic perspectives. Since the IL-5 receptor is present in very small numbers even on primed eosinophils (10^3 - 10^4/cell) and because it appears to be of very high affinity (K_d of 1-5 x 10^{-11}), it seems quite likely that both the circulating concentration of IL-5 and the total number of molecules per person will be exceedingly small. Thus, the use the existing murine anti-IL-5 mAB may be useful (since human anti-mouse antibodies may not develop with the very small amounts of mAB that would be required to neutralize IL-5). Although receptor antagonists and/or a humanized version of the murine monoclonal antibody would represent the most elegant and perhaps therapeutically better alternatives, the murine anti-IL-5 is in hand and could be used to reduce the presence and activation human eosinophils -- perhaps initially in the idiopathic hypereosinophil syndrome but, assuming safety is demonstrated, also in asthma.

Coda

Taken as a whole, while there are unfortunate gaps in our knowledge regarding the role of the eosinophil in asthma, data substantively support a proinflammatory role for the eosinophil over the hypothesis that it down-regulates the activity of mast cells and their mediators. Further, the relative importance of eosinophils compared to other inflammatory cells is yet to be firmly established and encourage increased research effort to dissect out the role of the eosinophil in order to best assess the value of pursuing development of pharmacologic agents capable of either *i*) blunting the recruitment and activation of eosinophils or *ii*) antagonizing the effects of released eosinophil mediators.

THERAPEUTIC POSSIBILITIES

Disorders Characterized by Nonproductive Eosinophil-Mediated Damage

Although the focus of this Grand Rounds has been to update the reader on the cell biology of the eosinophil and its potentially important role in asthma, it should be apparent that excessive activity of this cell may be critical in a number of other disorders. Appendices 1-5 list some of the states in which tissue and/or circulating levels of eosinophils are increased. The role of agents currently available to the clinician (systemic and topical glucocorticoids, ß-agonists and cromolyn) has already been discussed from a mechanistic perspective but, aside from anti-IL-5, the potential for other agents, not yet approved by the FDA, deserves mention. Nedocromil (Tilade^R) is similar to, but more effective than, cromolyn and is actively being used for therapy of allergic diseases outside the United States. It seems likely that approval will come in the near future. The H_1 histamine receptor antagonist, cetirizine, appears to have modest activity in reducing the activity of eosinophils. The ability of this agent to impact asthma, however, has not been established. Like nedocromil, approval by the FDA would appear to be in the near future.

In the more distant future, a number of agents appear to have promise. PAF antagonists are receiving abundant attention from the pharmaceutical industry and massive efforts related to the development, testing and assessment of clinical efficacy are under way. Unfortunately, the agents closest to clinical availability appear not to be as potent as one would hope for. Thus, the ability to block the effects of PAF-induced inflammation (eosinophils, platelets, neutrophils and endothelial cells) may be elusive for a time. LTD_4 receptor antagonists and 5-lipoxygenase inhibitors are in clinical trials, but initial findings suggest that their ability to interfere with chronic asthma (as opposed by acute challenge models) may be limited. As described in a previous section, the ability to fairly selectively reduce IL-5 mediated production and activation of eosinophils may be the most hopeful (albeit most distant, from a time perspective) approach to intervening in eosinophil-mediated inflammation.

Diseases in which Eosinophil-Mediated Inflammation May Be Beneficial

The eradication of helminths is difficult and currently available anti-helminthic agents often induces significant, and frequently limiting, human toxicity. During the chronic phase of helminthic of infection, parasites are careful not to kill the host, and most have devised a variety of clever ways to defuse the full force of immune and nonimmune inflammatory mechanisms that could be developed by the host. Although efforts are currently under way to develop vaccines capable of generating specific

immunity to certain helminthic infections, it may turn out that enhancing nonspecific mechanisms may be important. Although completely speculative, augmented eosinophil activity achieved by the brief administration of IL-5 (perhaps even associated by passive administration of specific anti-helminth IgG) might result in exceeding a critical threshold for antibody dependent eosinophil mediated cytotoxicity of helminths resulting in parasite eradication. Should serious adverse reactions develop using this approach (as well might occur) rapid reversal of increased eosinophil activity could likely be accomplished by the use of systemic glucocorticoids.

REFERENCES

Ackerman SJ, Loegering DA, Venge P, et al. 1983. Distinctive cationic proteins of the human eosinophil granule: major basic protein, eosinophil cationic protein, and eosinophil-derived neurotoxin. J. Immunol. 131: 2977.

Agosti JM, Altman LC, Ayars GH, Loegering DA, Gleich GJ, Klebanoff SJ. 1987. The injurious effect of eosinophil peroxidase, hydrogen peroxide, and halides on pneumocytes in vitro. J. Allergy Clin. Immunol. 79:496.

Anwar ARE, and Kay AB. 1977. Membrane receptors for IgG and complement (C4, C3b and C3d) on human eosinophils and neutrophils and their relation to eosinophilia. J. Immunol. 119:976.

Auriault C, Capron M, and Capron A. 1982. Activation of rat and human eosinophils by soluble factor(s) released by *Schistosoma mansoni* schistosomula. Cell Immunol. 66:59.

Ayars GH, Altman LC, Gleich GJ, Loegering DA, Baker CB. 1985. Eosinophil- and eosinophil granule-mediated pneumocyte injury. J. Allergy Clin. Immunol. 76:595.

Baigelman W, Chodosh S, Pizzuto D, Cupples LA. 1983. Sputum and blood eosinophils during corticosteroid treatment of acute exacerbations of asthma. Am. J. Med. 75:929.

Bass DA, Grover WH, Lewis JC, et al. 1980. Comparison of human eosinophils from normals and patients with eosinophilia. J Clin. Invest. 66:1265.

Basten A, Boyer MH, and Beeson PB. 1970. Mechanism of eosinophilia. I. Factors affecting the eosinophil response of rats to *Trichinella spiralis*. J. Exp. Med. 131:1271.

Basten A, and Beeson PB. 1970. Mechanism of eosinophilia. II. Role of the lymphocyte. J Exp Med. 131:1288.

Bruijnzeel PL. 1989. Contribution of eosinophil-derived mediators in asthma. Int. Arch. Allergy Appl. Immunol. 90(suppl)1:57-63.

Bruijnzeel PL, Koenderman L, Kok PTM, et al. 1986. Platelet-activating factor (PAF-acether) induced leukotriene C_4 formation and luminol dependent chemiluminescence by human eosinophils. Pharmacol Res Commun. 18(suppl):61.

Burrows B, Hasan FM, Barbe RA, Halonen M, Lebowitz MD. 1980. Epidemiologic observations on eosinophilia and its relation to respiratory disorders. Am. Rev. Resp. Dis. 122:709.

Butterworth AE. 1984. Cell-mediated damage to helminths. Adv Parasitol. 23:143.

Butterworth AE, Vadas MA, Wassom DL, et al. 1979. Interactions between human eosinophils and schistosomules of *Schistosoma mansoni*. II. The mechanisms of irreversible eosinophil adherence. J Exp Med. 150:1456.

Butterworth AE, Wassom DL, Gleich GJ, et al. 1979. Damage to schistosomula of *Schistosoma mansoni* induced directly by eosinophil major basic protein. J. Immunol. 122:221.

Butterworth AE. 1984. Cell-mediated damage to helminths. Adv Parasitol. 23:143.

Capron A, Capron M, Grangette C, Dessaint JP. 1989. IgE and inflammatory cells. Ciba Foundation Symposium. 147:153-60; discussion 160-70.

Capron M, Grangette C, Torpier G, Capron A. 1989. The second receptor for IgE in eosinophil effector function. Chemical Immunol. 47:128-78.

Capron M, Tomassini M, Torpier G, Kusnierz JP, MacDonald S, Capron A. 1989. Selectivity of mediators released by eosinophils. Int. Arch. Allergy Appl. Immunol. 88:54-8.

Capron M, Bazin H, Jospeph M, and Capron A. 1981. Evidence for IgE-dependent cytotoxicity by rat eosinophils. J Immunol. 126:1764.

Capron M, Kusnierz JP, Prin L, et al. 1985. Cytophilic IgE on human blood and tissue eosinophils. Int Arch Allergy Appl Immunol. 77:246.

Capron A, Dessaint JP, Capron M. 1990. Fc epsilon RII and IgE-dependent activation of inflammatory cells. Research in Immunol. 141:99-105.

Capron M, Spiegelberg HL, Prin L, et al. 1984. Role of IgE receptors in effector function of human eosinophils. J. Immunol. 132:462.

Carlson MG., Peterson CGB, and Venge P. 1985. Human eosinophil peroxidase: purification and characterization. J. Immunol. 134:1875.

Clark RAF, Gallin JI, and Kaplan AP. 1975. The selective eosinophil chemotactic activity of histamine. J. Exp. Med. 142:1462.

Dale DC, Hubert RT, and Fauci A. 1976. Eosinophil kinetics in the hypereosinophilic syndrome. J. Lab. Clin. Med. 87:487.

Gleich GJ, Schroeter Al, Marcoux JP, et al. 1984. Episodic angioedema associated with eosinophilia. N. Eng. J. Med. 310:1621.

Denburg JA, Telizyn S, Messner H, et al. 1985. Heterogeneity of human peripheral blood eosinophil-type colonies: evidence for a common basophil-eosinophil progenitor, Blood. 66:312.

DeSimone C, Donelli G, Meli D, et al. 1982. Human eosinophils and parasitic diseases. II. Characterization of two cell fractions isolated at different densities. Clin. Exp. Immunol. 48: 249.

Dessaint JP, Capron A. 1989. IgE and inflammatory cells: the cellular networks in allergy. International Archives All & Applied Immunol. 90(suppl)1:28-31.

Djukanovic R, Roche WR, Wilson JW, Beasley CR, Twentyman OP, Howarth RH, Holgate ST. 1990. Mucosal inflammation in asthma. Am. Rev. Resp. Dis. 142:434-57.

Dresch C, Johnson GR and Metcalf D. 1977. Eosinophil colony formation in semisolid cultures of human bone marrow cells, Blood. 49:835.

Durack DT, Ackerman SJ, Loegering DA, and Gleich GJ. 1981. Purification of human eosinophil-derived neurotoxin. Proc. Nat. Acad. Sci. (USA) 78:5165.

Enright T, Chua S, Lim DT. 1989. Pulmonary eosinophilic syndromes. Ann. Allergy. 62:277-83.

Fauci AS, Harley JB, Roberts WC, et al. 1982. The idiopathic hypereosinophilic syndrome. Clinical, pathophysiologic and therapeutic considerations. Ann. Int. Med. 97:78.

Fernandez HN, Henson PM, Otahi A, and Hugli TE. 1978. Chemotactic response to human C3a and C5a anaphylatoxins. I. Evaluation of C3a and C5a leukotaxis in vitro and under simulated in vivo conditions. J. Immunol. 120:10

Filley WV, Holley KE, Kephart GM, Gleich GJ. 1982. Identification by immunofluorescence of eosinophil granule major basic protein in lung tissue of patients with bronchial asthma. Lancet. 2:11

Flavahan NA, Aarhus LL, Rimele TJ, Vanhoutte PM. 1985. Respiratory epithelium inhibits bronchial smooth muscle tone. J. Appl. Physiol. 58:834.

Flavahan NA, Vanhoutte PM. 1985. The respiratory epithelium releases a smooth muscle relaxing factor. Chest. 87(suppl): 189s.

Fredens K, Dahl R., and Venge P. 1982. The Gordon phenomenon induced by the eosinophil cationic protein and eosinophil protein X. J. Allergy Clin. Immunol. 70:361.

Frew AJ, Kay AB. 1990. Eosinophils and T-lymphocytes in late-phase allergic reactions. J. Allergy Clin. Immunol. 85(3):533-9.

Frigas E, Loegering DA, Gleich GJ. 1980. Cytotoxic effects of the guinea pig eosinophil major basic protein on tracheal epithelium. Lab. Invest. 42:35.

Frigas E, Loegering DA, Solley GO, et al. 1981. Elevated levels of the eosinophil granule major basic protein in the sputum of patients with bronchial asthma. Mayo Clin. Proc. 56:345.

Frigas E, Loegering DA, Solley GO, Farrow GM, Gleich GJ. 1981. Elevated levels of the eosinophil granule major basic protein in the sputum of patients with bronchial asthma. Mayo Clin. Proc. 56:345.

Frigas E, Gleich GJ. 1986. The eosinophil and the pathophysiology of asthma. J. Allergy Clin. Immunol. 77:527-37.

Frigas E, and Gleich GJ. 1986. The eosinophil and the pathophysiology of asthma. J. Allergy Clin. Immunol. 77:527.

Fukuda T, Dunnette SL, Reed CE, et al. 1985. Increased numbers of hypodense eosinophils in the blood of patients with bronchial asthma. Am. Rev. Resp. Dis. 132:981.

Glauert AM, Butterworth AE, Sturrock RF, and Houba V. 1978. The mechanism of antibody-dependent, eosinophil-mediated damage to schistosomula of *Schistosoma mansoni* in vitro: a study by phase-contrast and electron microscopy. J. Cell Sci. 34:173.

Gleich GJ, Flavahan NA, Fujisawa T, Vanhoutte PM. 1988. The eosinophil as a mediator of damage to respiratory epithelium: a model for bronchial hyperreactivity. J. Allergy Clin. Immunol. 81:776-81.

Gleich GJ. 1990. The eosinophil and bronchial asthma: current understanding. J. Allergy Clin. Immunol. 85(2):422-36.

Gleich GJ, Frigas E, Loegering DA, et al. 1979. Cytotoxic properties of the eosinophil major basic protein. J. Immunol. 123:2925.

Gleich GJ, Loegering DA, Bell MP, et al. 1986. Biochemical and functional similarities between eosinophil-derived neurotoxin and eosinophil cationic protein: homology with ribonuclease. Proc. Nat. Acad. Sci. (USA) 83:3146.

Gleich GJ, Ottesen EA, Leiferman Km, Ackerman SJ. 1989. Eosinophils and human disease. Int. Arch. Allergy Appl. Immunol. 88:59-62.

Gleich GJ, Frigas E, Loegering DA, Wassom DL, Steinmuller D. 1979. Cytotoxic properties of the eosinophil major basic protein. J. Immunol. 123:2925.

Gleich GJ. 1988. Current understanding of eosinophil function. Hosp. Prac. 23(3):137-47, 151-4, 159-60.

Gleich GJ, Olson GM, and Loegering DA. 1979. The effect of ablation of eosinophils on immediate-type hypersensitivity reactions. Immunology 38:343.

Gleich GJ, Adolphson CR. 1986. The eosinophilic leukocyte: structure and function. Adv. Immunol. 39:177.

Gleich GJ, Loegering DA, Mann KG, Maldonado JE. 1976. Comparative properties of the Charcot-Leyden crystal protein and the major basic protein from human eosinophils. J. Clin. Invest. 57:633.

Gomperts BD. 1990. GE: a GTP-binding protein mediating exocytosis. Ann Rev of Physiol. 52:591-606.

Gross R. 1962. The eosinophils. In Braunsteiner H, and Zucker-Franklin D, editors: The physiology and pathology of leukocytes, New York, Grune & Stratton, Inc.

Grover WH, Winkler HH, and Normansell DE. 1978. Phagocytic properties of isolated human eosinophils. J. Immunol. 121:718.

Hamann KJ, Barker RL, Loegering DA, Gleich GJ. 1987. Comparative toxicity of purified human eosinophil granule proteins for newborn larvae of *Trichinella spiralis.* J. Parasitol. 73:523.

Hamann KJ, Barker RL, Loegering DA, and Gleich GJ. 1987. Toxicity of purified human eosinophil granule proteins for newborn larvae of *Trichinella spiralis.* J. Parasitol. 7:523.

Hansel FK. 1929. Clinical and histopathologic studies of the nose and sinuses in allergy. J. Allergy 1:43.

Henderson WR, and Chi EY. 1985. Ultrastructural characterization and morphometric analysis of human eosinophil degranulation. J. Cell Sci. 73:33.

Henderson WR, Chi EY, and Klebanoff SJ. 1980. Eosinophil peroxidase-induced mast cell secretion. J Exp Med. 152:265.

Henderson WR, Jorg A, and Klebanoff SJ. 1982. Eosinophil peroxidase-mediated inactivation of leukotrienes B_4,$C_4$and D_4, J. Immunol. 128:2609.

Henderson WR, Chi EY, Jong EC, Klebanoff SJ. 1981. Mast cell-mediated tumor-cell cytotoxicity. J. Exp. Med. 153:520.

Henderson WR, Harley JB, and Fauci AS. 1984. Arachidonic acid metabolism in normal and hypereosinophilic syndrome human eosinophils: generation of leukotrienes B_4, C_4, D_4, and 15-lipoxygenase products. Immunology. 51:679.

Hertzman, PA, WL Blevins, J Mayer, B Greenfield, M Ting and GJ Gleich. 1990. Association of the eosinophilia-myalgia syndrome with the ingestion of tryptophan. New Eng. J. Med. 322:869-73.

Hirsch JG, Hirsch BI. 1980. Paul Ehrlich and the discovery of the eosinophil. In Mahmoud AF and Austen KR, editors: The eosinophil in health and disease, New York, Grune & Stratton, Inc., pp 3.

Hogg JC, Eggleston PA. 1984. Is asthma an epithelial disease? Am. Rev. Resp. Dis. 129:207-8.

Ihle JN, Keller J, Oroszlan S, et al. 1983. Biologic properties of homogeneous interleukin 3. J. Immunol. 131:282.

Ishizaka T., Saito H, Hatake K, Dvorak AM, Leiferman KM, Arai N, Ishizaka K. 1989. Preferential differentiation of inflammatory cells by recombinant human interleukins. Int. Arch. Allergy Appl. Immunol. 88:46-9.

Kajita T, Yui Y, Mita H, et al. 1985. Release of leukotriene C_4 from human eosinophils and its relation to the cell density. Int. Arch. Allergy Appl. Immunol. 78:406.

Kay AB, Frew AJ, Moqbel R, Walsh GM, Kurihara K, Cromwell O, Champion A, Hartnell A, Wardlaw AJ. 1989. The activated eosinophil in allergy and asthma. Prog. Clin. Biol. Research. 297:183-96; discussion 196-7.

Kay AB. 1987. Mediators and inflammatory cells in allergic disease. Ann. Allergy. 59:35-42.

Kroegel C. 1990. The role of eosinophils in asthma. Lung. 168(suppl):5-17.

Laitinen LA, Heino M, Laitinen A, Kava T, Haahtela T. 1985. Damage of the airway epithelium and bronchial reactivity in patients with asthma. Am. Rev. Respir. Dis. 131:599-606.

Lee T-C, Lenihan DJ, Malone B, et al. 1984. Increased biosynthesis of platelet-activating factor in activated human eosinophils. J. Biol. Chem. 259:5526.

Lewis DM, Lewis JC, Loegering DA, Gleich GJ. 1978. Localization of the guinea pig eosinophil major basic protein to the core of the granule. J. Cell Biol. 77:702.

Lewis DM, Lewis JC, Loegering DA, and Gleich GJ. 1978. Localization of the guinea pig eosinophil major basic protein to the core of the granule. J. Cell Biol. 77:702.

LJ Crofford, JI Rader MC Dalakas et al. 1990. L-tryptophan implicated in human eosinophilia-myalgia syndrome causes fasciitis and perimyositis in the Lewis Rat. J. Clin. Invest. 86:1757-63.

Lowell FC. 1967. Clinical aspects of eosinophilia in atopic disease. JAMA 202:875.

Maddox DE. 1988. Eosinophils. Assays and interpretation. Clinical Reviews In Allergy. 6(2):163-90.

Maddox DE, Butterfield JH, Ackerman SJ, et al. 1983. Elevated serum levels in human pregnancy of a molecule immunochemically similar to eosinophil granule major basic protein. J. Exp. Med. 158:1211.

Martin, RW, J Duffy, AG Engel, JT Lie, CA Bowles, TP Moyer and GJ Gleich. 1990. The clinical spectrum of the eosinophilia-myalgia syndrome associated with L-tryptophan ingestion. Ann. Int. Med. 113:124-34.

McLaren DJ, McKean JR, Olsson I, et al. 1981. Morphological studies on the killing of schistosomula of *Schistosoma mansoni* by human eosinophil and neutrophil cationic proteins in vitro. Parasite Immunol. 3:359.

Melski JW. 1990. Wells' syndrome, insect bites, and eosinophils. Derm. Clin. 8(2):287-93.

Metcalf D, Begley CG, Johnson GR, et al. 1986. Biologic properties in vitro of a recombinant human granulocyte-macrophage colony-stimulating factor, Blood. 67:37.

Miller F, DeHarven E, Palade GE. 1966. The structure of the eosinophil leukocyte granules in rodents and in man. J. Cell Biol. 31:349.

Mizel SB. 1989. The interleukins. FASEB J. 3(12):2379-88.

Morley J. 1986. Platelet activating factor and asthma. Agents & Actions. 19(1-2):100-8.

Nadel JA. 1983. Bronchial reactivity. Adv. Int. Med. 28:207-23.

Nagy L, Lee TH, Goetzl EJ, et al. 1982. Complement receptor enhancement and chemotaxis of human neutrophils and eosinophils by leukotrienes and other lipoxygenase products. Clin. Exp. Immunol. 47:541.

Nolan CR, Kelleher SP. 1988. Eosinophiluria. Clinics In Lab. Med. 8:555-65.

Olsen RL, Syse K, Little C, and Christensen TB. 1985. Further characterization of human eosinophil peroxidase. Biochem J. 229:779.

Owhashi M, and Ishii A. 1982. Purification and characterization of a high molecular weight eosinophil chemotactic factor from *Schistosoma japonicum* eggs. J. Immunol. 129:2226.

Page CP, Coyle AJ. 1989. The interaction between PAF, platelets and eosinophils in bronchial asthma. Eur. Resp. J. (Suppl.) 6:483s-487s.

Parrillo JE, and Fauci AS. 1978. Human eosinophils. Purification and cytotoxic capability of eosinophils from patients with the hypereosinophilic syndrome. Blood

51:457.

Parwaresch MR, Walle AJ, and Arndt D. 1976. The Peripheral kinetics of human radiolabelled eosinophils. Virchows Arch. (Cell Pathol) 21:57

Peters MS, Rodriguez M, Gleich GJ. 1986. Localization of human eosinophil granule major basic protein, eosinophil cationic protein, and eosinophil-derived neurotoxin by immunoelectron microscopy. Lab. Invest. 54:650.

Peters MS. 1987. The eosinophil Advances in Dermatology. 2:129-52.

Presentey B. 1970. Partial and severe peroxidase and phospholipid deficiency in eosinophils. Cytochemical and genetic considerations. Acta Haematol. 44:345.

Prin L, Capron M, Tonnel A-B, et al. 1983. Heterogeneity of human peripheral blood eosinophils: variability in cell density and cytotoxic ability in relation to the level and the origin of hypereosinophilia. Int. Arch. Allergy Appl. Immunol. 72:336.

Saito H, Yamada K, Breard J, et al. 1986. A monoclonal antibody reactive with human eosinophils. Blood. 67:50.

Sanderson CJ. 1990. The biological role of interleukin 5. Int. J. Cell Cloning. 8(suppl)1:147-53; discussion 153-4.

Sanderson CJ, O'Garra A, Warren DJ, and Klaus GGB. 1986. Eosinophil differentiation factor also has B-cell growth factor activity: proposed name interleukin 4. Proc. Nat. Acad. Sci. (USA) 83:437.

Sanderson CJ, Campbell HD, Young IG. 1988. Molecular and cellular biology of eosinophil differentiation factor (interleukin-5) and its effects on human and mouse B cells. Immunological Rev. 102:29-50.

Shaw RJ, Walsh GM, Cromwell O, et al. 1985. Activated human eosinophils generate SRS-A leukotrienes following IgG-dependent stimulation. Nature 316:150.

Shurin SB. 1988. Pathologic states associated with activation of eosinophils and with eosinophilia. Hematology-Oncology Clin. N. Amer. 2:171-9.

Silberstein DS, Austen KF, Owen WF,Jr. 1989. Hemopoietins for eosinophils. Glycoprotein hormones that regulate the development of inflammation in eosinophilia-associated disease. Hematology-Oncology Clin. N. Amer. 3:511-33.

Silberstein DS, and David JR. 1986. Tumor necrosis factor enhances eosinophil toxicity to *Schistosoma mansoni* larvae. Proc. Nat. Acad. Sci. (USA) 83:1055.

Silver, RM, MP Heyes, JC Maize, B Quearry, M Vionnet-Fuasset and EM Sternberg. 1990. Scleroderma, fasciitis and eosinophilia associated with the ingestion of tryptophan. New Eng. J. Med. 322:874-81.

Spry CJ. 1986. Eosinophils in eosinophilic endomyocardial disease. Postgrad. Med. J. 62:609-13.

Steinbach KH, Schick P, Trepel F., et al. 1979. Estimation of kinetic parameters of neutrophilic, eosinophilic, and basophilic granulocytes in human blood. Blut. 39:27

Sutton JM. 1986. Urinary eosinophils. Arch. Int. Med. 146:2243-4.

Telerman A, Amson RB, Delforge A, et al. 1982. A case of chronic aneosinocytosis. Am J Hematol. 12:187.

Ten RM, Torres VE, Milliner DS, Schwab TR, Holley KE, Gleich GJ. 1988. Acute interstitial nephritis: immunologic and clinical aspects. Mayo Clin. Proc. 63:921-30.

Thorne KJI, Richardson BA, Veith MC, Tai P-C, Spry CJF, Butterworth AE. 1985. Partial purification and biological properties of an eosinophil-activity factor. Eur. J. Immunol. 15:1083.

Venge P. 1990. What is the role of the eosinophil? Thorax. 45(3):161-3.

Venge P. 1990. The human eosinophil in inflammation. Agents Actions. 29(1-2):122-6.

Venge P, Dahl R., and Hallgren R. 1979. Enhancement of factor XII dependent reactions by eosinophil cationic protein. Throm. Res. 14:641.

Venge P, Dahl R. 1989. The eosinophil and intrinsic asthma. Agents Actions (Suppl) 28:67-74.

Vercelli D, Geha RS. 1989. The IgE system. Annals of Allergy. 63(1):4-11.

Walsh GM, and Kay AB. 1986. Binding of immunoglobulin classes and subclasses to human neutrophils and eosinophils. Clin Exp Immunol. 63:466.

Wardlaw AJ, Moqbel R, Cromwell O, and Kay AB. 1986. Platelet-activating factor. A potent chemotactic and chemokinetic factor for human eosinophils. J. Clin. Invest. 78:1701.

Warren DJ, Sanderson CJ. 1985. Production of a T cell hybrid producing a lymphokine-stimulating eosinophil differentiation. Immunology 54:615.

Warren DJ. 1988. The biology of murine eosinophil differentiation factor (an overview). Behring Institute Mitteilungen. (83):98-101.

Wasmoen TL, Coulam CB, Leiferman KM, and Gleich GJ. 1987. Increases of plasma eosinophil major basic protein levels late in pregnancy predict onset of labor. Proc. Nat. Acad. Sci. (USA) 84:3029.

Wasserman SI, Goetzl EJ, and Austen KF. 1975. Inactivation of slow reacting

substance of anaphylaxis by human eosinophil arylsulfatase. J. Immunol. 114:645.

Weller PF, Bach DS, and Austin KF. 1984. Biochemical characterization of human eosinophil Charcot-Leyden crystal protein (lysophospholipase). J. Biol. Chem. 259:15100.

Weller PF, Bach D, and Austin KF. 1982. Human eosinophil lysophospholipase: the sole protein component of Charcot-Leyden crystals. J. Immunol. 128:1346.

Weller PF, Goetzl EJ, Austen KF. 1980. Identification of human eosinophil lysophospholipase as the constituent of Charcot-Leyden crystals. Proc. Nat. Acad. Sci. (USA) 77:7440.

Weller PF, Goetzl EJ, and Austin KF. 1980. Identification of human eosinophil lysophospholipase as the constituent of Charcot-Leyden crystals. Proc. Nat. Acad. Sci. (USA) 77:7440.

Zheutlin LM, Ackerman SJ, Gleich GJ, and Thomas LL. 1984. Stimulation of basophil and rat mast cell histamine release by eosinophil granule-derived cationic proteins. J. Immunol. 133:2180.

Zucker-Franklin D. 1980. Eosinophil structure and maturation. In Mahmoud AAF, and Austin FK, editors: The eosinophil in health and disease, New York, Grune & Stratton, Inc. p. 43.

Zucker-Franklin D. 1985. Eosinophils. In Golde DW, and Takaku F., editors: Hematopoietic stem cells, Hematology 2:45.

APPENDIX 1

REPRESENTATIVE VALUES FOR CIRCULATING EOSINOPHILS*
IN A SAMPLING OF DISORDERS AND CONDITIONS
WITH WHICH EOSINOPHILIA IS COMMONLY ASSOCIATED†

10,000 to 50,000
- Eosinophilic leukemia
- Hypereosinophilic syndrome

5000 to 10,000
- Tropical eosinophilia
- Visceral larva migrans

3000 to 5000
- Allergic granulomatosus with angiitis
- Bronchopulmonary aspergillosis
- Chronic eosinophilic pneumonia‡

1500 to 3000
- Angioblastic lymphoid hyperplasia (Kimura's disease)
- Asthma of undefined etiology (intrinsic): hyperplastic sisusitis, nasal polyps, aspirin-intolerance syndromes
- Drug allergy, acute reactions (interstitial nephritis, cholestatic hepatitis, exfoliative dermatitis)
- Helminth infection (migratory stage)
- Immunoblastic lymphadenopathy
- Löffler's syndrome

± *1500*
- Asthma (allergic)
- Atopic dermatitis
- Chronic myelogenous leukemia
- Eosinophilic fasciitis
- Hypersensitivity vasculitis
- Immunodeficiency syndromes (Wiskott-Aldrich, hyper-IgE infection, IgA deficiency with atopy)
- Prolonged pulmonary eosinophilia, PIE syndromes
- Urticaria (acute allergic)

± *1000*
- Addison's disease
- Allergic rhinitis
- Carcinoma of ovary
- Coccidioidomycosis
- Cyclic neutropenia
- Drugs, biologic products
- Hodgkin's disease
- Immunologic skin diseases
- Polycythemia vera
- Serum sickness
- X-irradiation

*Total cell counts, eosinophilis per mm^3.

†Figures are intended to represent the most frequently observed ranges of reported counts and not the arbitrary diagnostic criteria.

‡In some instances circulating counts may be at normal values.

APPENDIX 2

Table 35-3. Clinical eosinophilia: disorders and conditions associated with local and/or circulating eosinophilia

Disorder*	Occurrence†					
	Tissue			Blood		
	Infrequent	Variable	Usual	Infrequent	Variable	Usual
Respiratory tract						
Allergic rhinitis			3+		1+	
Asthma, allergic			3+			2+
Asthma, cause undefined (intrinsic)			3+			2+
Bronchocentric granulomatosis		2+				2+
Bronchopulmonary aspergillosis			3+			3+
Eosinophilic pneumonia, chronic			2+		3+	
Hyperplastic sinusitis, nasal polyps			3+			2+
Hypersensitivity pneumonitis		1+				
Löffler's syndrome			3+			3+
Pleural inflammation, effusion secondary to hemorrhage, infarction, trauma, tumor, metastases	2+					
Pneumonia, convalescent stage	1+			1+		
Tropical eosinophilia			2+			4+
Vasomotor rhinitis	1+					
Gastrointestinal tract						
Crohn's disease	1+					
Eosinophilic esophagitis, gastritis, enteritis			2+			2+
Eosinophilic granuloma, polyp		1+				
Gastroenteropathy with pulmonary disease (Heiner's syndrome)		2+				1+
Peptic ulcer		1+				
Peritoneal inflammation (secondary to dialysis)		2+			1+	
Proctitis		1+				
Sclerosing cholangitis, cholecystitis		2+				
Ulcerative colitis		1+				
Skin						
Atopic dermatitis			3+			2+
Bullous pemphigoid		1+		1+		
Contact dermatitis	1+					
Dermatitis herpetiformis			2+			2+
Dermatitis vegetans		1+			1+	
Eosinophilic cellulitis, granulomatous dermatitis (Well's disease)			2+			2+
Eosinophilic cutaneous collagenosis			2+			2+
Erythema annulare centrifugum	1+					

*For a discussion and explanation of putative mechanisms of eosinophilia for each listed disorder see corresponding sections in the text.

†Relative degrees of eosinophilia are intended as a guide and should not be interpreted as absolute differential diagnostic points. Absence of entry indicates lack of eosinophilia.

Continued.

Disorder	Occurrence					
	Tissue			Blood		
	Infrequent	Variable	Usual	Infrequent	Variable	Usual
Skin—cont'd						
Erythema multiforme		1+		1+		
Erythema multiforme exudativum (Stevens-Johnson syndrome)			1+		1+	
Erythema neonatorum, erythema toxicum			1+			1+
Erythema nodosum		1+		1+		
Exfoliative dermatitis, pityriasis rubra			2+			2+
Herpes gestationis			1+		1+	
Ichthyosis			2+		1+	
Impetigo herpetiformis			1+			1+
Lichen planus	1+					
Lymphomatoid papulosis	1+					
Papular urticaria		1+				
Pemphigus			1+		1+	
Pityriasis rosea	1+					
Psoriasis	1+					
Scabies			2+		1+	
Toxic epidermal necrolysis (drug; Lyell's syndrome)			2+		1+	
Urticaria, angioedema (allergic recurrent or prolonged)			2+			2+
Urticaria pigmentosa, mastocytosis		1+		1+		
Urinary tract						
Cystitis, prostatitis, urethritis		1+				
Glomerulonephritis						
Acute poststreptococcal					1+	
Persistent hypocomplementemic					1+	
Granulomatous prostatitis			2+		2+	
Interstitial cystitis			2+		1+	
Interstitial nephritis (drug)			2+			2+
Endocrine glands						
Adrenal						
Addison's disease						1+
Adrenocortical hypofunction						1+
Following adrenalectomy						1+
Pituitary						
Pituitary hypofunction						1+
Sheehan's disease, Simmond's disease						1+
Ovary						
Ovarian tumors					2+	
Vasculature and heart						
Vasculitis						
Allergic granulomatosis with angiitis (Churg-Strauss syndrome)			3+			3+
Giant cell (temporal) arteritis		1+			1+	
Henoch-Schönlein purpura		1+			1+	
Hypersensitivity vasculitis			2+			2+
Polyarteritis nodosa	1+				1+	
Thromboangiitis obliterans		1+				
Wegener's granulomatosis		1+			1+	
Heart						
Löffler's fibroplastic endocarditis			3+			3+
Postcommissurotomy syndrome	1+				1+	
Postmyocardial infarction syndrome	1+				1+	
Serous myocarditis, endomyocardial syndrome			3+			3+

Disorder	Occurrence					
	Tissue			Blood		
	Infrequent	Variable	Usual	Infrequent	Variable	Usual
Central nervous system						
Cerebral cysticercosis		1+				
Eosinophilic meningitis						
Helminth infection (*Angiostrongylus cantonensis*)			3+			3+
Hodgkin's disease, lymphoma	1+				1+	
Following myelography	2+					
Microbial infection (coccidioidomycosis, syphilis, tuberculosis)	1+			1+		
Neoplasm, cerebral metastasis	1+			1+		
Sydenham's chorea				1+		
Liver						
Cholestatic hepatitis (drug hepatotoxicity)			3+			2+
Infectious hepatitis						
Acute, prodromal syndrome		1+				1+
Chronic active		1+			1+	
Sclerosing cholangitis		2+				
Spleen						
Following splenectomy						1+
Pancreas						
Acinar cell carcinoma					1+	
Chronic pancreatitis				1+		
Neonatal islet hypertrophy-hyperplasia			1+			
Eye						
Allergic conjunctivitis			2+		1+	
Vernal conjunctivitis			2+		1+	
Connective tissue diseases						
Arthrography and joint irradiation	2+					
Dermatomyositis	1+			1+		
Eosinophilic collagen disease			2+			2+
Eosinophilic fasciitis			3+		2+	
Progressive systemic sclerosis (scleroderma)	1+			1+		
Rheumatoid arthritis				1+		
Rheumatic fever				1+		
Sjögren's syndrome				1+		
Systemic lupus erythematosus				1+		
Immunodeficiency disorders						
Graft versus host disease			2+			2+
Hyperimmunoglobulinemia E with susceptibility to infection			1+			2+
IgA selective deficiency with atopy			2+			2+
Neonatal (transient, immature) immune response		1+			1+	
Thymic alymphoplasia					2+	
Thymic dysplasia with immunoglobulins (Nezelof syndrome)					2+	
Wiskott-Aldrich syndrome			2+			2+
Neoplasia						
Acinar cell carcinoma of pancreas					1+	
Adenocarcinoma of colon	1+					
Epidermoid carcinoma: lip, penis, tongue, uterine cervix, uterus, vulva		1+				

Continued.

Disorder	Occurrence					
	Tissue			Blood		
	Infrequent	Variable	Usual	Infrequent	Variable	Usual
Neoplasia—cont'd						
Carcinoma						
Lung	1+			1+		
Ovary		2+			2+	
Stomach	1+			1+		
Thyroid	1+			1+		
Tumors with necrosis, inflammation, metastases to bone marrow and serosal surfaces		2+			1+	
Undifferentiated carcinoma of nasopharynx		1+				
Villous carcinoma of urinary bladder	1+					
Hematologic disorders						
Chronic myelogenous leukemia					2+	
Cyclic neutropenia						2+
Eosinophilic leukemia			4+			4+
Hypereosinophilic syndrome			4+			4+
Idiopathic thrombocytopenic purpura				1+		
Leukemoid reactions						1+
Myelofibrosis						1+
Myeloid metaplasia						1+
Neutropenia					1+	
Polycythemia vera					2+	
Drugs						
Antigenic sensitizing agents (asymptomatic)		2+			2+	
Aspirin idiosyncrasy syndrome		2+				2+
Beta-adrenergic blocking agents		2+			2+	
Mast cell degranulating agents			1+		1+	
Infection						
Bacterial						
Brucellosis, pulmonary	1+			1+		
Leprosy	1+			1+		
Scarlet fever						2+
Tuberculosis (especially caseous and lymph node involvement)	2+				1+	
Chlamydia pneumonia					1+	
Mycotic						
Coccidioidomycosis		2+			2+	
Chronic mucocutaneous candidiasis					1+	
Parasitic						
Helminths, tissue invasive, visceral larva migrans			3+			3+
Pneumocystis carinii, pneumonia with immunodeficiency			2+		2+	
Protozoa: amebiasis, giardiasis, coccidiosis	1+					
Viral (presumptive)						
Acute infectious lymphocytosis					1+	
Erythema infectiosum					1+	

Lymphoproliferative and lymphoreticular disorders					
Acute lymphatic leukemia				2+	
Angioblastic lymphoid hyperplasia, eosinophilic lymphofolliculosis (Kimura's disease)			3+		2+
Eosinophilic granuloma (stage of histiocytosis X)			3+		1+
Familial reticuloendotheliosis		2+			2+
Granuloma faciale		1+			
Histiocytic lymphoma				1+	
Hodgkin's disease			2+		1+
Immunoblastic lymphadenopathy		1+			2+
Lymphomatoid papulosis	1+				
Multicentric giant cell reticulohistiocytosis		1+			
Mycosis fungoides			1+		1+
Plasma cell dyscrasias					
Heavy chain disease				1+	
Primary macroglobulinemia				1+	
Pseudolymphoma, cutaneous and lymph node		2+		1+	
Sarcoid					1+
Sézary's syndrome			1+		1+
X-irradiation					
Radiation exposure					1+
Radiation of infant thymus					1+
Radiation of lesions		1+			1+

APPENDIX 3

Table 38-2. Disorders characterized by pulmonary infiltrates with eosinophilia (PIE syndromes)—cont'd

Clinical associations	Blood eosinophilia*	IgE level
Usually asymptomatic, or mild cough, low-grade fever, constitutional symptoms; wheezing and dyspnea infrequent; self-limited, 3-4 wk duration of causative factor removed (drug, migratory helminth)	Moderate	Moderate
Course protracted beyond 4 wk; moderate to marked cough, dyspnea, fever; associated systemic symptoms dependent on cause; variants of pulmonary infection (tuberculosis, coccidioidomycosis, schistosomiasis), drug hypersensitivity reactions	Variable, dependent on cause; usually moderate	Variable, dependent on cause; drug allergy—moderate
Predilection for women ages 20-50; marked cough, dyspnea, fever, and constitutional symptoms; asthma mild if coincident; rapid response to corticosteroids; prolonged course	Moderate to high	Not established
Major presentation of chronic asthma; fever, expectoration of mucoid plugs containing eosinophils and fungal hyphae; hypersensitivity to *Aspergillus;* partial control with corticosteroids	High	High
Marked cough, chest pain, fever, and constitutional symptoms; one subset with presentation of asthma and hypersensitivity to *Aspergillus;* responsive to corticosteroids	Subsets: 1. Nonasthmatic—low 2. Asthmatic—moderate to high	 Not established Not established
Continuous or remittent-recurrent pattern of marked bouts of cough, wheezing, and dyspnea, especially nocturnal; fever and constitutional symptoms; represents subset of filarial infection; responsive to specific therapy, diethylcarbamazine	High	High
Systemic vasculitis involving heart, lungs, skin, and central nervous system; pulmonary involvement consistent; associated with atopic disease, usually asthma; respiratory illness (bronchitis, pneumonia, asthma) precedes systemic dissemination of vasculitis	High	Moderate to high (in presence of allergic state)
Systemic vasculitis with variable involvement of lungs, skin, heart, spleen, joints, kidney, and liver; skin most commonly affected with lesions secondary to leukocytoclastic vasculitis of postcapillary veins; variable pericarditis, myocarditis, peripheral neuropathy, and major vascular occlusion; asthma and pulmonary symptoms only infrequently present	Low to moderate	Low to moderate
Cough, dyspnea, variable cardiovascular and constitutional symptoms as part of systemic disorder; multiorgan involvement; hepatosplenomegaly, frequent cutaneous and neurologic lesions	High (highest)	Subsets: 1. Low 2. Moderate to high

Table 38-2. Disorders characterized by pulmonary infiltrates with eosinophilia (PIE syndromes)

PIE disorder	Pathologic features	Radiographic characteristics
Löffler's syndrome	Alveolar spaces and septa infiltrated with eosinophils *and* histiocytes	Patchy homogeneous densities; peripheral and upper lobe distribution, unilateral or bilateral, transient, migratory
Eosinophilic pneumonia		
Prolonged pulmonary eosinophilia syndromes	Dependent on cause of localized or protracted pneumonitis; alveolar and interstitial cellular infiltrates of varied mixed types, eosinophils, multinucleate giant cells; fibrin deposits	Variable, dependent on cause; single or multiple infiltrates, nodular or confluent densities; cavitation
Chronic eosinophilic pneumonia	Alveoli filled with eosinophils and vacuolated mononuclear cells; interstitial tissue and walls of small bronchi and bronchioles infiltrated by eosinophils and smaller numbers of plasma cells, lymphocytes and macrophages; Charcot-Leyden crystals in some macrophages	Dense opacities with ill-defined margins; peripheral distribution apposed to pleura, usually axillary or apical; rapidly progressive
Bronchopulmonary aspergillosis	Bronchi dilated and filled with inspissated mucus and exudate containing fungal hyphae; eosinophil, lymphocyte, and plasma cell infiltration of bronchial walls; eosinophilic pneumonia of surrounding pulmonary parenchyma	Recurrent changing shadows; usually bilateral and in upper zones, homogeneous but may be mottled, nodular, ring, or unusually shaped; proximal segmental bronchiectesis; lobar shrinkage
Bronchocentric granulomatosis	Necrotic granuloma surrounded by palisaded epithelial cells involving small bronchi and adjacent to pulmonary arteries; granulomas containing remnants of bronchial elastic tissue, admixture of cellular debris with some identifiable eosinophils, neutrophils, desquamated lining cells, and erythrocytes; interstitial infiltrate of eosinophils, lymphocytes, and plasma cells	Shadows initially unilateral; predominating upper lobe involvement; infiltrates, lobar consolidation, or atelectasis; nodular and mixed nodular-linear patterns
Tropical eosinophilia	Diffuse parenchymal infiltration; early histiocyte interstitial and alveolar infiltrates followed by eosinophilic abscesses; later organized nodules of eosinophils, histiocytes, and lymphocytes evolving to histiocytic granulomas and fibrosis; rare degenerating microfilariae.	Normal to bilateral miliary lesions and mottled opacities; uniform distribution, middle and lower lobes; occasional confluence of mottled shadows; increased bronchovascular markings
Pulmonary vasculitis		
Allergic granulomatosis with angiitis (Churg-Strauss syndrome)	Necrosis, fibrinoid deposition and leukocyte infiltration of small and medium sized arteries with extravascular granulmatous nodules; granulomas containing central core of necrotic cells and altered collagen fibers surrounded by eosinophils, macrophages, and giant cells, proximal to small veins; healing with fibrous tissue proliferation	Vary from normal to parenchymal infiltrates, usually diffuse but may be focal; multiple areas of consolidation, occasional cavitation; changing pattern with resolution of some lesions and appearance of new areas of involvement
Hypersensitivity vasculitis	Fibrinoid necrosis and cellular inflammation of small branches of arteries, veins, and capillaries; progressive fibrinoid necrosis from intimal and subendothelial region to entire vessel wall; pleomorphic cellular infiltrates with eosinophils predominating; small foci of necrotizing pneumonia	Variable, migratory infiltrates and nodular pattern; cavitation of nodules; pleural effusion
Hypereosinophilic syndrome	Pulmonary parenchyma diffusely infiltrated by eosinophils; infiltrates involve pulmonary interstitium, pulmonary vasculature, periadventitial regions, and myocardium; endomyocardial fibroelastosis and mural thrombi	Variable pattern of pulmonary infiltrates; cardiomegaly

APPENDIX 4

Table 38-4. Eosinophilia and parasitic infections

Infecting organism	Principal sites of involvement	Eosinophilic response: Tissue	Eosinophilic response: Blood	Comments
Roundworms (nematodes)				
Ascaris lumbricoides (giant roundworm)	Intestine	±	±	Adult stages, habitat within intestinal lumen
	Lung	+	+	Migratory stages, transient eosinophilic infiltrate as lung invasion occurs
Nector americanus	Intestine	+	+	Adult stages, mucosal penetration
Ancylostoma duodenale (human hookworms)	Lung	+	+	Migratory stages, transient eosinophilic infiltrate as lung invasion occurs
Enterobius vermicularis (pinworm)	Intestine	−	−	Superficial mucosal attachment
Trichuris trichiura (whipworm)	Intestine	−	−	Superficial mucosal attachment
Strongyloides stercoralis (threadworm)	Intestine	+	+	Adult stages, mucosal penetration
	Lung	+	+	Migratory stages, transient eosinophilic infiltrate as lung invasion occurs
Trichostrongylus species	Intestine	±	±	Mucosal penetration
Trichinella spiralis	Intestine	±	±	Adult stages
	Muscle	+	−	Migratory stage
	Muscle	−	±	Encapsulated larvae
Toxocara canis and *cati* (dog, cat roundworm)	Migratory: liver, lung, central nervous system, eye	+	+	Visceral larva migrans, transient eosinophilic infiltrate if lung invasion occurs, human not normal host
Ancylostoma braziliense (dog hookworm)	Skin	+	−	Cutaneous larva migrans, transient eosinophilic infiltrate if lung invasion occurs, human not normal host
Gnathostoma spinigerum	Skin, subcutaneous tissue			Cutaneous larva migrans, human not normal host
Angiostrongylus cantonensis (rodent lungworm)	Central nervous system	+	−	Eosinophilic meningitis, human not normal host
Wuchereria bancrofti	Lymphatics	+	−	Filariasis
Brugia malayi	Lung	+	−	Tropical pulmonary eosinophilia
Onchocerca volvulus	Subcutaneous tissue, eye	+	−	"River blindness"
Loa loa (eye worm)	Subcutaneous tissue, eye	+	−	"Fugative swellings"
Dirofilaria immitis (dog heartworm)	Lung	+	−	Pulmonary nodule, human not normal host
Flatworms (trematodes)				
Schistosoma mansoni	Intestine, liver, lung	+	−	Adults intravascular, pathology results from
Schistosoma japonicum	Intestine, liver, lung	+	−	eggs in tissues, transient eosinophilic infil-
Schistosoma haematobium	Genitourinary tract, lung	+	−	trate as lung invasion occurs
Trichobilharzia species	Skin	+	−	"Swimmer's itch," human not normal host
Clonorchis sinensis (Chinese liver fluke)	Liver, biliary tree	+	−	Eggs deposited in tissues, adult stages
Fasciola hepatica (sheep liver fluke)	Liver, biliary tree	+	−	Egg deposited in tissues, adult stages
Paragonimus westermani (oriental lung fluke)	Lung	+	−	Adult stages
	Liver, central nervous system, etc.	+	−	Migratory stages
Flatworms (cestodes)				
Taenia saginata (beef tapeworm)	Intestine	±	±	Adult stages, habitat within intestinal lumen
Taenia solium (pork tapeworm)	Intestine	±	±	Adult stages, habitat within intestinal lumen
	Subcutaneous tissue, muscle, central nervous system	+	−	Cysticercosis, migratory stages, human not normal host
Hymenolepis nana (dwarf tapeworm)	Intestine	±	±	Superficial mucosal attachment

Table 38-4. Eosinophilia and parasitic infections—cont'd

Infecting organism	Principal sites of involvement	Eosinophilic response		Comments
		Tissue	Blood	
Flatworms (cestodes)—cont'd				
Diphyllobothrium latum (fish tapeworm)	Intestine	±	±	Adult stages, superficial mucosal attachment
Echinococcus granulosus and *E. multilocularis* (hydatid worms)	Liver, lung, central nervous system	+	+	Migratory stages, human not normal host
Protozoa				
Plasmodium vivax, P. malariae, and *P. falciparum*	Blood, erythrocytes	−	−	
Entamoeba histolytica	Intestinal tract	±	−	Mucosal penetration
	Liver	±	−	
Giardia lamblia	Gastrointestinal tract	−	−	Superficial mucosal attachment
Trichomonas vaginalis	Genitourinary tract	−	−	
Pneumocystis carinii	Lung	±	±	Eosinophilia in presence of associated immunodeficiency
Toxoplasma gondii	Lymphatics, central nervous system, eye	−	−	
Leishmania donovani	Spleen, liver, bone marrow, lymph nodes	−	−	Visceral leishmaniasis
Leishmania tropica and *L. braziliensis*	Skin	−	−	Cutaneous leishmaniasis
Trypanosoma gambiense and *T. rhodesiense*	Lymphatics, central nervous system	−	−	Sleeping sickness
Trypanosoma cruzi	Muscle, heart, central nervous system	−	−	Chagas disease

APPENDIX 5

Table 35-5. Drugs associated with eosinophil responses and eosinophil-related drug reactions*

Drug	Asymptomatic eosinophil response†	Eosinophil associated reaction type
Acetylsalicylic acid		5, 7, 9, 10
Allopurinol		2, 5
Amitriptyline (tricyclics)		3, 9
Arsenicals	+	3, 5, 6, 9
Azathioprine		1, 2
Busulfan		5
Cephalosporin		2, 4, 9
Chloramphenicol		6, 9
Chlordiazepoxide		3
Chlorpromazine (phenothiazines)	+	3, 5, 9
Chlorothiazide (benzothiadiazines)		2, 3, 4, 5, 9
Codeine (morphine)		8
Dapsone (sulfones)		6, 9
Dextran		8
Erythromycin estolate		3
Gold	+	1, 2, 6, 9
Halothane		3, 10
Indomethacin		7
Iodides	+	1, 4, 5, 9
Isoniazid		3, 6
Mercurials		5
Nitrofurantoin	+	1, 3
Para-aminosalicylic acid	+	1, 3, 4, 9
Penicillin		2, 4, 5, 9, 10
Penicillin analogues		
Ampicillin	+	2, 4, 6
Methicillin		2, 4
Oxacillin		2, 3
Phenacemide		3, 9
Phenolphthalein		6
Phenylbutazone (pyrazalon: aminopyrine/phenacetin)		2, 3, 4, 5, 6, 9
Phenytoin	+	2, 4, 5, 6, 9, 10
Polymyxin		8
Protamine		8
Quinidine		5, 9
Stilbamidine		8
Streptomycin	+	4, 6
Sulfonamides	+	1, 2, 3, 4, 6, 9, 10
Sulfonylureas		
Chlorpropamide		1, 3, 6, 9
Tolbutamide		3, 6, 9
Tetracycline		4, 5
Thiouracil, propylthiouracil (thoiurea)		3, 4, 5, 9
Tranylcypromine		3
Trimethadione, paramethadione		3, 6, 9
d-Tubocurarine		8

1. Pulmonary infiltrates (PIE syndrome)
2. Interstitial nephritis
3. Cholestatic hepatitis (hepatocanicular or combined hepatocanicular-hepatocellular)
4. Serum sickness syndrome (urticaria, arthralgias, fever, adenopathy)
5. Hypersensitivity vasculitis
6. Severe dermatologic syndromes‡, (erythroderma, exfoliative dermatitis, erythema multiforme/Stevens-Johnson syndrome, toxic epidermal necrolysis/Lyell's syndrome)
7. Asthma, hyperplastic sinusitis, nasal polyps syndrome, urticaria
8. Mast cell degranulation phenomena (flushing, urticaria, nausea)
9. Hematologic dyscrasia syndromes (neutropenia, agranulocytosis, thrombocytopenia)
10. Immunoblastic lymphadenopathy

*Exclusive of parenterally administered biologics of animal derivation (foreign proteins) that frequently produce eosinophilia by IgE-immediate hypersensitivity mechanisms (allergic reactions) or as antigens in immune complex phenomena (serum sickness reactions) (e.g., horse serum antitoxins, insulin, liver extract, virus vaccines derived from chicken egg cultures, ACTH, pancreatic extract, and pituitary snuff).

†Drugs most commonly associated with asymptomatic eosinophilia and with the highest frequency of eosinophil responses.

‡Skin eruptions in a variety of types can appear as manifestations of reactions to most of the listed drugs; only those associated with the indicated severe dermatologic syndromes are mentioned.