

Quantification of resident inflammatory cells in the human nasal mucosa

Yasushi Igarashi, MD,^a Michael A. Kaliner, MD,^a Jeffrey N. Hausfeld, MD,^b Ann-Marie A. Irani, MD,^c Lawrence B. Schwartz, MD,^d and Martha V. White, MD^a Bethesda, Md., Washington, D.C., and Richmond, Va.

Background: To define the normal resident inflammatory cell population in the nasal mucosa, surgical specimens of human nasal turbinates were immunohistologically stained for various cell markers.

Methods: Freeze-dried paraffin-embedded sections were stained for lymphocyte cell-surface markers, and Carnoy's fixed sections were stained for mast cells and immunoglobulins. The numbers of stained cells were microscopically counted.

Results: T cells (CD3+ cells) were abundant in the lamina propria, and the number of CD4+ cells and CD8+ cells accounted for two thirds and one third of CD3+ cell number, respectively. Cells that stained for the α -chain of the interleukin-2 receptor (activated cells, CD25+) were limited and accounted for only 0.6% of CD3+ cell number. B cells (CD22+ cells) and monocytes and macrophages (CD14+ cells) were observed less frequently than T cells. Many immunoglobulin-producing cells were found in close proximity to the submucosal glands, and those cells were predominantly IgA+. Mast cells were widely distributed in the nasal mucosa, and about one third of these cells were stained for IgE molecules. Nonmast cells bearing IgE were rarely observed.

Conclusion: Thus the dominant cell in the nasal mucosa is a CD3+, CD4+, CD25- lymphocyte. (*J ALLERGY CLIN IMMUNOL* 1993;91:1082-93.)

Key words: Inflammatory cells, nasal mucosa, immunohistochemistry

The nasal mucosa provides the first line of defense against inhaled pathogens and inhaled foreign substances; the mucous layer forms a barrier between foreign substances and the mucosal surface, epithelial ciliary activity facilitates the elimination of trapped particulates, antimicrobial substances such as lactoferrin or lysozyme are secreted from the glands to kill susceptible pathogens, and phagocytic cells take up foreign parti-

Abbreviations used

IL:	Interleukin
CD:	Cluster designation
APAAP:	Alkaline phosphatase anti-alkaline phosphatase
BM:	Basement membrane
$\gamma\delta$ -T cell:	T cell bearing $\gamma\delta$ -chain T cell receptor
MC _T :	Tryptase-positive, chymase-negative mast cells
MC _{TC} :	Tryptase-positive, chymase-positive mast cells

From ^athe Allergic Diseases Section, Laboratory of Clinical Investigation, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, Md.; ^bDepartment of Facial, Plastic and Reconstructive Surgery, Washington Hospital Center, Washington, D.C.; Department of ^cPediatrics and ^dDepartment of Medicine, Medical College of Virginia, Virginia Commonwealth University, Richmond, Va.

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Reprint requests: Martha V. White, MD, NIH Building 10, Room 11C207, 9000 Rockville Pike, Bethesda, MD 20892.

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cles. In addition to these nonspecific factors, immunologic responses such as cell-mediated cytotoxicity and antibody production are involved in nasal defense mechanisms. Numerous inflammatory disorders involve the nasal mucosa, including allergic rhinitis, Sjögren's syndrome, Wegener's granulomatosis, and other diseases in which sinusitis is common (e.g., common variable immunodeficiency, acquired immunodeficiency syn-

TABLE I. Antibodies used

Antibody	Species	Cell specificity	Source
Anti-CD3	Mouse monoclonal	Pan-T cell	Becton-Dickinson*
Anti-CD4	Mouse monoclonal	Helper/inducer T cell	Becton-Dickinson*
Anti-CD8	Mouse monoclonal	Suppressor/cytotoxic T cell	Becton-Dickinson*
Anti-CD22	Mouse monoclonal	B cell	Becton-Dickinson*
Anti-CD14	Mouse monoclonal	Monocyte/macrophage	Becton-Dickinson*
Anti-CD25	Mouse monoclonal	Activated lymphocyte	Becton-Dickinson*
Anti-TCR δ -chain	Mouse monoclonal	$\delta\gamma$ -T cell	T Cell Sciences†
Anti-J-chain	Rabbit polyclonal	Immunoglobulin-producing cell	Nordic Immunological‡
Anti- α -chain	Rabbit polyclonal	IgA-bearing cell	Dako§
Anti- δ -chain	Rabbit polyclonal	IgD-bearing cell	Dako§
Anti- μ -chain	Rabbit polyclonal	IgM-bearing cell	Dako§
Anti- ϵ -chain	Rabbit polyclonal	IgE-bearing cell	Dako§
Anti-chymase	Mouse monoclonal	Mast cell	VCU
Anti-tryptase	Mouse monoclonal	Mast cell	VCU
Anti-major basic protein	Mouse monoclonal	Eosinophil	Sera-Lab

VCU, Virginia Commonwealth University.

*Mountain View, Calif.

†Cambridge, Mass.

‡Carpistrano Beach, Calif.

§Carpinteria, Calif.

||Sussex, England.

drome). As such, the nasal mucosa is an excellent model of mucosal immunity and is easily accessible for study.

Immune responses are regulated by various inflammatory mediators and cytokines produced by inflammatory cells. In allergic rhinitis, allergens bind to IgE antibody on the surface of mast cells in the nasal mucosa and induce the secretion of vasoactive substances, such as histamine and prostaglandin D₂,¹ which cause nasal allergy symptoms (rhinorrhea, nasal congestion, sneezing, and pruritus). Interleukin-3 (IL-3)² in conjunction with other factors may be responsible for mast cell proliferation. IgE production can be stimulated by IL-4³ and inhibited by interferon- γ .⁴ Acute allergic reactions induce an influx of eosinophils,⁵ which may participate in late allergic reactions or airway hyperresponsiveness. Therefore it is of interest to determine the identity of inflammatory cells in the nasal mucosa.

Few investigators have studied the inflammatory cells in the nasal mucosa with immunohistochemical staining techniques.⁵⁻⁸ T cells are the most commonly observed cells, although B cells and monocytes are also present. However, because of the tissue structure and immunoreactivity, quantification of these cells has not been easy.⁹ The technique used in the current study provides

improved tissue preservation because tissue is embedded in paraffin in order to cut thinner sections. To initiate the detailed examination of the nasal mucosal immune response in inflammatory states, it was necessary to define the normal resident population of inflammatory cells.

METHODS

Preparation of tissues

Human inferior nasal turbinate tissues were obtained from morbidly obese patients (age range, 15 to 67 years) who underwent turbinectomies to relieve sleep apnea. The turbinates were not inflamed at the time of surgery. Fresh unfixed nasal specimens were immediately dissected from the turbinate bone, and the 10 to 15 mm pieces were snap-frozen with liquid nitrogen or precooled (with dry ice) 2-methylbutane. Frozen specimens were lyophilized at -45°C , 5×10^{-2} torr for 24 hours.¹⁰ Freeze-dried tissues were then allowed to warm to room temperature and embedded in paraffin. The paraffin-embedded blocks were sliced in 4 μm sections and mounted on gelatin-coated slides. Sections were kept at 4°C until use. Some frozen pieces were sliced in 4 μm sections with a cryostat, mounted on gelatin-coated slides, and kept at -80°C until use. These sections were stained for cluster designation (CD) markers. For immunoglobulin and mast cell staining, tissues were fixed with Carnoy's fixative for 4 to 16 hours, then washed with absolute ethanol. Some pieces were fixed with 4% paraformaldehyde for 4 to 16 hours,

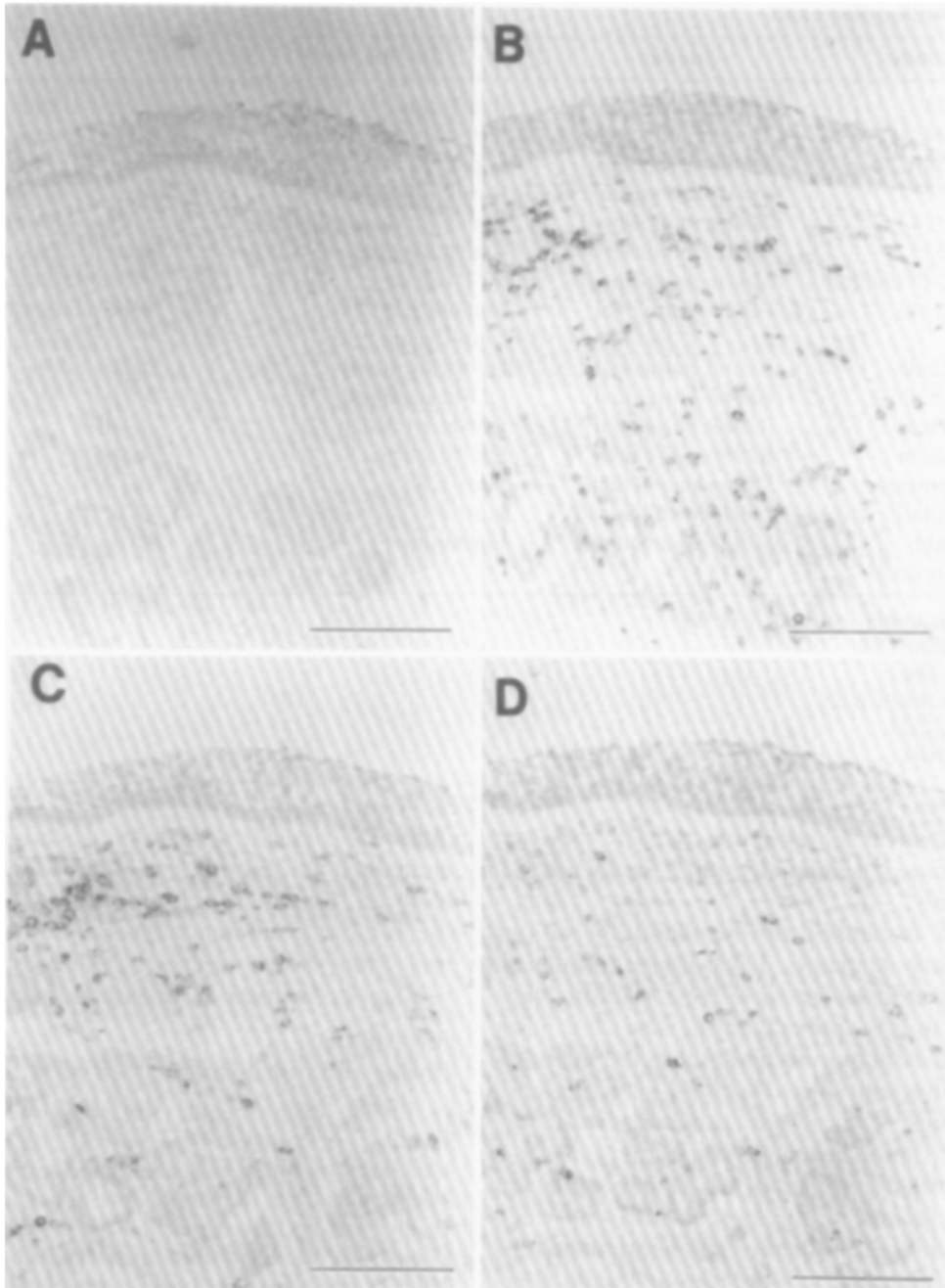


FIG. 1. Staining of nasal mucosa for T cells. Serial sections of freeze-dried paraffin-embedded human nasal mucosa were stained for **(B)** CD3, **(C)** CD4, and **(D)** CD8 with an APAAP technique. Control staining **(A)** was performed by substituting nonimmune serum for the first antibody. Scale bars indicate approximately 100 μ m.

then washed with phosphate-buffered saline. These fixed tissues were also embedded in paraffin, sliced in 4 μ m sections, and kept at 4° C.

Immunohistochemical staining

Staining for CD markers. Freeze-dried paraffin-embedded sections were stained for CD markers with an alkaline phosphatase anti-alkaline

phosphatase (APAAP) method (Dako, Carpinteria, Calif.) as described elsewhere.¹¹ Briefly, sections were deparaffinized in xylene for 15 minutes and then incubated with acetone for 10 minutes. After sequential incubation with mouse monoclonal antibodies raised against human leukocyte cell-surface antigens (the first antibody, Table I) at room temperature for 40 minutes and then against rabbit

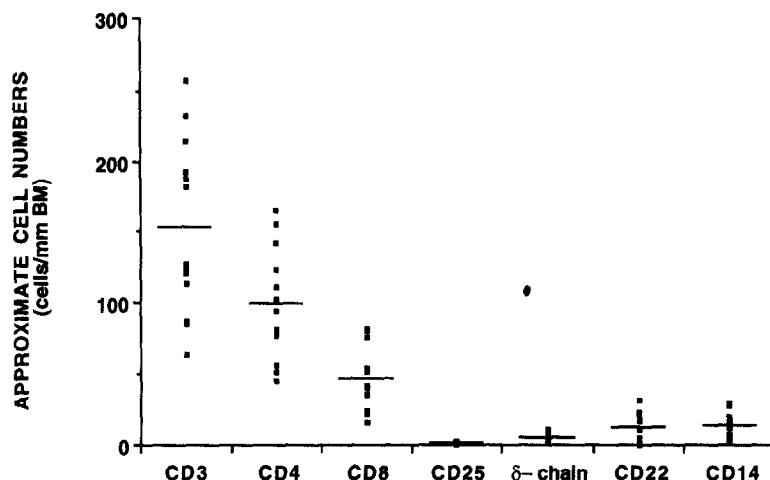


FIG. 2. The number of lymphocytes in the nasal mucosa. Positively stained cells were counted in the superficial 200 μ m below the basement membrane in the lamina propria and were divided by the length of the basement membrane (BM). Each dot represents the result from an individual patient's tissue ($n = 13$). Cross bars represent the mean.

anti-mouse immunoglobulin for 30 minutes, followed by APAAP (mouse) for 30 minutes. Sections were visualized by naphthol AS-MX/fast red (Dako). Frozen cryostat sections were allowed to dry over silica gel under vacuum for 1 hour, fixed with acetone at 4° C for 10 minutes, further dried over silica gel for 1 hour, and then stained for surface markers with the APAAP method described above. Freeze-dried sections and frozen cryostat sections stained equivalently for these CD markers. Staining for lymphocytes was also confirmed by use of freeze-dried tonsil tissues. Slides were counterstained with Mayer's hematoxylin stain and mounted in glycerin-gelatin solution.

Staining for immunoglobulins. Carnoy's fixed paraffin-embedded sections were stained for several kinds of immunoglobulins with a mouse APAAP method. Since antibodies for human immunoglobulins (the first antibodies, Table I) were rabbit antibodies, mouse anti-rabbit immunoglobulin was applied to serve as a link between the first antibody and rabbit anti-mouse immunoglobulin. In some experiments a rabbit peroxidase anti-peroxidase method¹² (Dako) was used instead of the APAAP method. In some parallel experiments paraformaldehyde-fixed tissues were stained for immunoglobulins. The staining was equivalent for Carnoy's fixed and paraformaldehyde-fixed tissues.

Staining for mast cells. Carnoy's fixed paraffin-embedded sections were sequentially double-stained for mast cell proteases, tryptase, and chymase, as described previously.¹³ Briefly, sections were incubated with biotinylated mouse anti-chymase at 4° C overnight, then with peroxidase conjugated streptavidin for 1 hour, and visualized with hydrogen peroxide/3-amino-9-ethylcarbazole. The sections were further incubated with alkaline-phosphatase-conjugated mouse anti-tryptase at 4° C

overnight and visualized with naphthol AS-MX/fast blue (Sigma Chemical Co., St. Louis, Mo.). Some tissues were double-stained for IgE and tryptase. In these experiments sections were first stained for IgE with a peroxidase anti-peroxidase method, then for tryptase with direct alkaline phosphatase immunostaining. Changing the staining order (first tryptase, then IgE) did not affect the results.

Cell counting

Counting of stained cells was done through a grid attached to the eyepiece of the microscope at a magnification of $\times 450$. The scale of the grid was determined by an objective micrometer. The number of positively stained cells was counted in the lamina propria (within 200 μ m of basement membrane [BM]). Because it was difficult to obtain accurate numbers of cells in cell clusters, the number of cells in a cluster was roughly counted, and the clusters were categorized into three groups according to size. The small clusters, which were estimated to contain no more than five cells, were considered uniformly to contain three cells. The medium clusters, which were larger than a small cluster and were estimated to contain no more than 10 cells, were considered uniformly to contain eight cells. The large clusters, which were larger than a medium cluster and were estimated to contain no more than 20 cells, were considered to contain 15 cells. Other clusters, which were larger than a large cluster, were considered to be the union of multiple clusters. The number of clusters in each category was counted, and approximate numbers of cells in the area were obtained by summing the number of solitary cells and the estimated number of cells in clusters. The cell numbers were divided by the length of the BM and expressed as the cell number per millimeter BM. Some tissue sections were counted several times, and the intraobserver

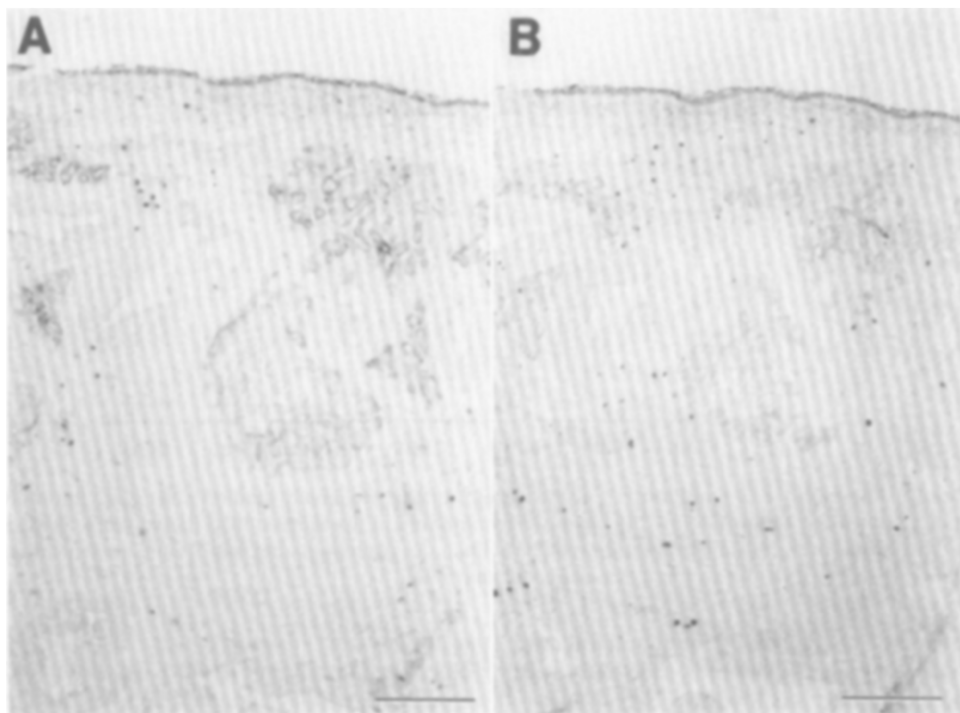


FIG. 3. Staining of nasal mucosa for B cells and monocytes. Serial sections of freeze-dried paraffin-embedded human nasal mucosa were stained for CD22 (**A**) and CD14 (**B**) with an APAAP technique. Scale bars indicate approximately 400 μ m.

variation was 2.5% (coefficient of variation, $n = 5$). The variation between different sections of the same tissue was 4.7% ($n = 5$). The variation between two observers was 3.5% ($n = 5$). In addition, the numbers of mast cells were counted in the entire epithelium and also expressed as the cell number per millimeter BM.

RESULTS

Staining for CD markers

Tissue morphology of tissues fixed in either Carnoy's fixative or paraformaldehyde was always superior to that of frozen sections. The interstitium of frozen cryostat sections was often enlarged and loose with large empty spaces (not shown). On the other hand, since freeze-dried sections that were used in this study were embedded in paraffin, the interstitium was much denser than in the cryostat sections and appeared similar to that of the fixed sections. Most of the anti-CD antibodies that were used do not stain fixed tissues. Therefore for the purpose of cell staining for CD markers, freeze-dried sections were used.

CD3+ cells (T cells) were numerous in the subepithelial region (Fig. 1, *B*), sometimes formed clusters, and were less abundant in the deep vascular bed (not shown). CD4+ cells (mainly helper T cells) and CD8+ cells (mainly cytotoxic

suppressor T cells) were distributed much in the same way as CD3+ cells, but CD4+ cells were more prominent than CD8+ cells (Fig. 1, *C*, *D*). Cell counting was performed in the superficial 200 μ m of the lamina propria. Approximately 150 CD3+ cells per millimeter BM were observed (Fig. 2). CD4+ cells accounted for 66% of the CD3+ cells, whereas CD8+ cells accounted for 32%. The sum of CD4+ cells and CD8+ cells approximated the number of CD3+ cells. CD25 (α -chain of IL-2 receptor) was examined as a marker of T cell activation.¹⁴ CD25+ cells were rarely detectable in these tissues (Fig. 2). T cells bearing the $\gamma\delta$ -chain of the T cell receptor¹⁵ were also sought. Staining of nasal mucosa for δ -chain revealed these cells to be located in the same area as CD3+ cells (not shown), but the numbers of $\gamma\delta$ -bearing T cells were minimal (Fig. 2).

CD22+ cells (B cells) and CD14+ cells (monocytes and macrophages) were less plentiful than CD3+ or CD4+ cells in the superficial lamina propria (Fig. 2). However, unlike T cells, B cells and macrophages were evenly distributed throughout the deeper vascular region and the lamina propria (Fig. 3). Although no specific staining for neutrophils was performed, polymorphonuclear cells were rarely found in hematoxylin

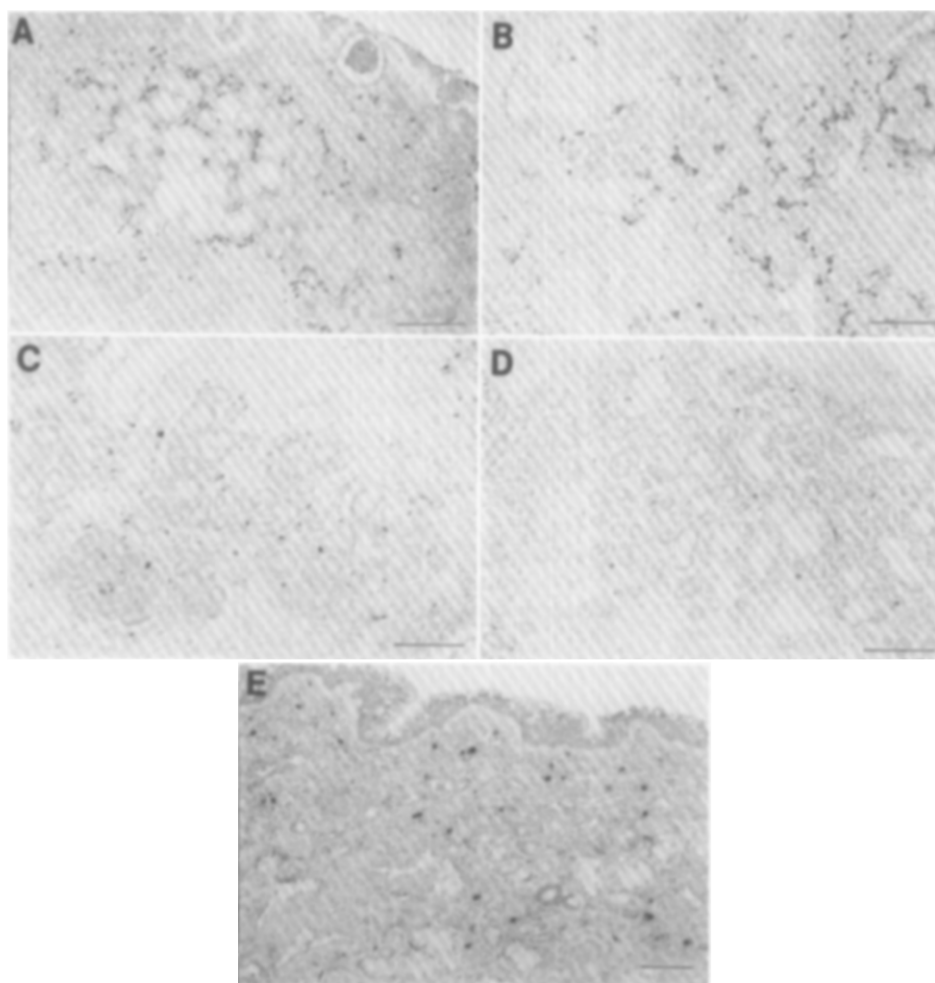


FIG. 4. Staining of nasal mucosa for immunoglobulin-bearing cells. Serial sections of Carnoy's fixed paraffin-embedded human nasal mucosa were stained for (A) J-chain, (B) IgA, (C) IgD, (D) IgM, and (E) IgE with an APAAP technique. Scale bars indicate approximately 100 μ m.

and eosin stained sections (not shown). The number of eosinophils stained with major basic protein was also small (approximately 1 cell per millimeter BM; not shown).

Staining for immunoglobulins

J-chain staining was used for the marker of Ig-producing cells.¹⁶ J-chain+ cells were seen in close proximity to submucosal glands in the nasal mucosa (Fig. 4, A). As expected, cells bearing IgA+, IgD+, and IgM+ were predominantly in the glandular region (Fig. 4, B, C, and D), with IgA+ cells the most plentiful. IgG stained the tissue diffusely, and IgG+ cells were rarely identified (not shown). However, when nasal tissue was incubated in phosphate-buffered saline for 3 days before fixation to wash out the interstitial IgG molecules, some IgG+ cells, which were also present in the glandular area, were identified (not shown). Because this treatment decreases tissue

preservation, it was not routinely performed. In contrast to other Ig+ cells, the distribution of IgE+ cells was not restricted to the glandular region (Fig. 4, E).

The numbers of Ig+ cells were also counted within 200 μ m of the lamina propria and expressed as the number of cells per millimeter BM (Fig. 5). In any individual the number of IgA+ cells was greater than that of IgM+ cells, whereas there was a large variation in IgD+ cell numbers.

There were variations in the richness of the glands among the specimens. Therefore the number of periglandular Ig-bearing cells in the lamina propria might not accurately reflect the magnitude of cellular infiltration into the tissues. For this reason, Ig+ cells were also counted in the submucosal glands and within 100 μ m of the glands and expressed as the number of cells per square millimeter (Fig. 6). The number of IgA+ cells was almost as great as that of J-chain+ cells,

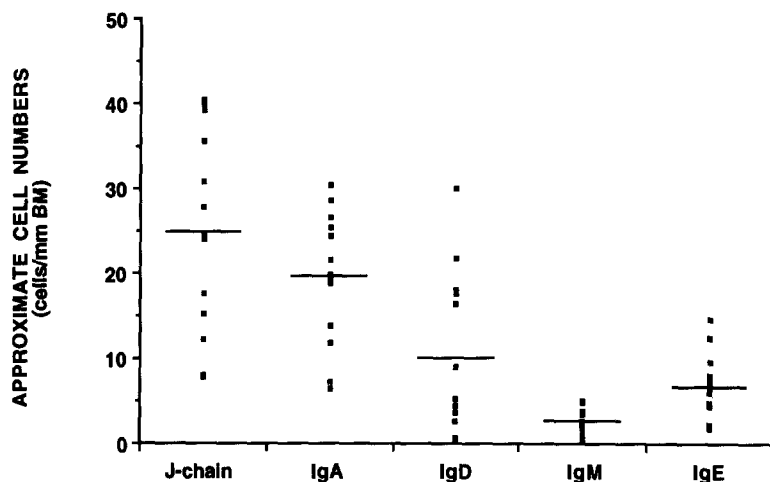


FIG. 5. The number of immunoglobulin-bearing cells in the nasal mucosa. Positively stained cells were counted in the superficial 200 μ m below the basement membrane in the lamina propria and divided by the length of the basement membrane (BM). Each dot represents the result from an individual patient's tissue ($n = 13$). Cross bars represent the mean.

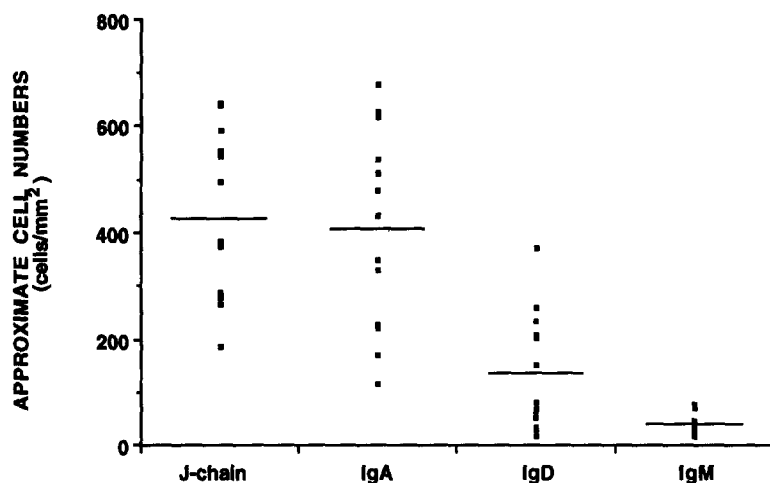


FIG. 6. The number of immunoglobulin-bearing cells in the nasal subepithelial gland area. Positively stained cells were counted within 100 μ m of the submucosal glands and divided by the total area in which cell numbers were counted. Each dot represents the result from an individual patient's tissue ($n = 13$). Cross bars represent the mean.

whereas IgD+ and IgM+ cells constituted a smaller number.

Staining for mast cells

When sections were double-stained for mast cell tryptase and chymase, the stained cells were widely distributed in the tissue, and most of them were chymase-positive, as well as tryptase-positive (Fig. 7, A and B). However, the subepithelial mast cells and the intraepithelial mast cells, although few, were predominantly chymase-negative (Fig. 7, C). The number of mast cells (chymase- or tryptase-positive cells) in the lamina propria was about 16 cells per millimeter BM, and more than 90% were chymase-positive cells (Fig. 8). There

were about 0.5 mast cells per millimeter BM in the epithelium, and more than 80% were chymase-negative cells (Fig. 8).

When sections were double-stained for IgE and tryptase (Fig. 7, D and E), more than 97% of IgE+ cells were also stained for tryptase, which indicated that these IgE+ cells were mostly mast cells. The number of IgE+ cells (5.5 cells per millimeter BM) represented about one third of the total number of mast cells.

DISCUSSION

Inflammatory cells in the local airways play important roles in airway defense mechanisms. To begin to understand the role of inflammatory cells

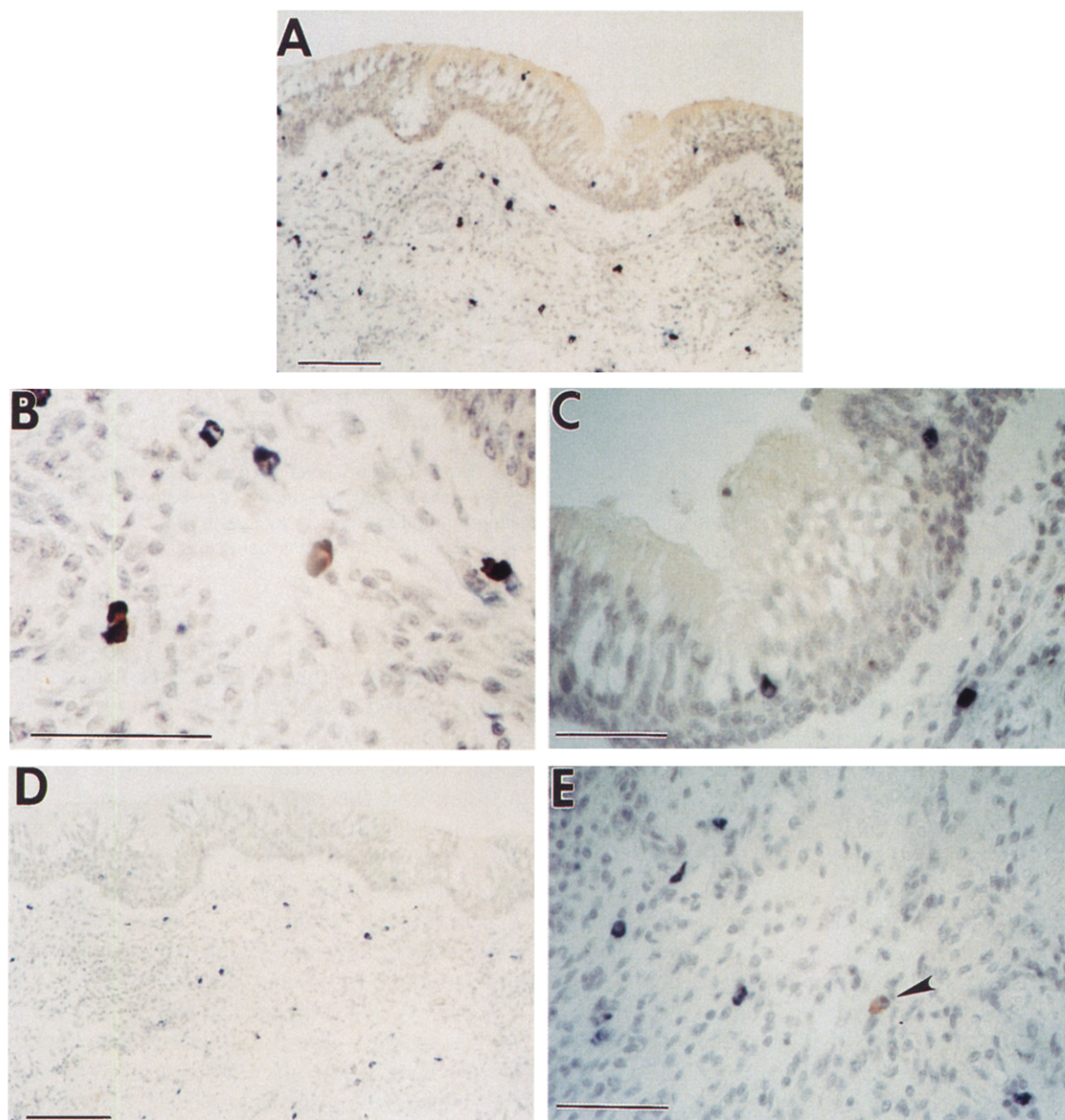


FIG. 7. Staining of nasal mucosa for mast cells. Carnoy's fixed paraffin-embedded sections from human nasal mucosa were sequentially stained for mast cell chymase and tryptase with peroxidase and alkaline phosphatase immunostaining, respectively. **Panel A** shows low magnification. Representative cells in the lamina propria (**B**) and in the epithelium (**C**) are shown at higher magnification. Mast cells in the lamina propria were mostly double-stained with blue and brown, indicating that these cells were both tryptase- and chymase-positive. Mast cells in the epithelium were mostly stained blue only, indicating that these cells were tryptase-positive and chymase-negative. Some sections were sequentially stained for IgE and mast cell tryptase with the peroxidase anti-peroxidase technique and alkaline phosphatase immunostaining, respectively. Most of the IgE-positive cells (brown) were costained for tryptase (blue) (**D**). At higher magnification the majority of IgE cells were stained blue (**E**). Only a few brown cells in the entire section failed to costain blue (arrow head). Scale bars indicate approximately 100 μ m (**A**, **D**) and 50 μ m (**B**, **C**, **E**).

in nasal inflammatory disorders, we evaluated the presence of those cells in the nasal mucosa. In order to quantify the inflammatory cell distribution, surgically removed inferior nasal turbinates from patients with sleep apnea were used. Inflammatory cells were abundant throughout the ante-

rior three quarters of the turbinate and somewhat less abundant in the most posterior portion (not shown). Inflammatory cells appeared most abundant at the site of maximal air flow. Although subjects from whom the turbinates were removed did not have inflammatory disease, these patients

T cells (CD3+, CD4+, and CD8+ cells) were located mainly in the superficial lamina propria of the nasal turbinate and less frequently in the deeper vascular bed. These findings agree with previous reports.⁶⁻⁸ Because T cells are often found in aggregates, we estimated the total number of cells in aggregates on the bases of size and number of cell clusters in order to minimize subjective error. The variation in T cell numbers among individuals in this study was about four-fold. This is low compared with a previous report, which showed about 100-fold variation in the numbers of CD4+ or CD8+ cells.⁸ Although some reports showed an equal or larger number of CD8+ cells as compared with CD4+ cells in normal subjects,^{7,8} we found almost twice as many CD4+ cells as CD8+ cells, which is consistent with the results of Winther's study.⁶ The possible reason for these differences is that the sensitivity for CD4 staining might be different in the tissue preparation used in the present study, since we observed similar numbers of CD8 staining cells compared with other reports, whereas CD4+ cells were more numerous. Further, the sum of the CD4+ and CD8+ cells approximated the CD3+ cells in our study but not in the study by Stoop et al.⁷ Stoop et al.⁷ explained the shortage of pan-T cells compared with the sum of CD4+ cells plus CD8+ cells in their study by referring to the finding of Ernst et al.,¹⁷ who demonstrated the lack of pan-T cell markers on mucosal lymphocytes. However, that study examined intestinal intraepithelial lymphocytes. Some monocytes express CD4,¹⁸ and some natural killer cells express CD8¹⁹; thus, there are CD3-, and CD4+ cells, as well as CD3-, CD8+ cells. On the other hand, there are some T cells that express neither CD4 nor CD8. It has been demonstrated that most T cells bearing the $\gamma\delta$ -chain T cell receptor ($\gamma\delta$ -T cell) lack CD4 and CD8 antigen expression²⁰ and are considered to recognize the nonclassical major histocompatibility complex molecules.²¹ $\gamma\delta$ -T cells were observed frequently in intestinal intraepithelial lymphocytes.²⁰ In the present study the number of $\gamma\delta$ -T cells in the nasal lamina propria was only about 3% of CD3+ cells. Further, $\gamma\delta$ -T-cells were rarely found in the epithelium (not shown). CD25, which recognizes the α -chain of the IL-2 receptor, appears on the surface of activated T cells¹⁵ and some other cells including B cells,²² natural killer cells,²³ and monocytes.²⁴ In a study of bronchial mucosa more CD25+ cells were seen in subjects with asthma than in subjects without asthma.²⁵ CD25+ cells

were rarely seen in our study, and the number of CD25+ cells accounted for only 0.6% of the CD3+ cells. Thus although there are many resident T cells in the nasal mucosa, most of them seem to be in the resting state.

B cells and monocytes and macrophages were less plentiful than T cells and were distributed equally throughout the mucosa. CD22 antigens, which are present on circulating B cells and on B cells in the lymph nodes,²⁶ are lost after stimulation with mitogens.²⁷ In order to detect more differentiated B cells, immunoglobulins on the cell surfaces were stained. J-chains are found in polymeric immunoglobulins (IgM and IgA) but can also be found on other Ig-producing cells.¹⁶ In contrast to CD22+ cells, J-chain+ and Ig+ cells were located around the submucosal glands. This observation is in agreement with data published by Meredith et al.²⁸ and suggests that B cells, which migrate into the tissues from subepithelial capillaries or from the deeper sinusoids, locate toward the glandular region as they differentiate into plasma cells. The Ig-bearing cells in the glandular area were predominantly IgA+. It is very likely that these IgA+ cells are supplying IgA molecules, which are taken up by secretory component expressed on serous cells of the submucosal glands and subsequently secreted after cholinergic stimulation.²⁹ Although IgG-bearing cells in the nasal mucosa were also found to be periglandular, unwashed nasal tissue stained diffusely positive for IgG. Since vascular permeability results in IgG secretion,²⁸ these findings suggest that IgG molecules in the nose are primarily of vascular origin. We also observed some IgD+ cells and IgM+ cells can be secreted from exocrine organs, but to our knowledge, IgD secretion has not been described. During B cell maturation, IgM and IgD initially appear on the cell surface but are lost after Ig-type switching.³⁰

In contrast to other Ig-bearing cells, IgE+ cells were located both in the superficial lamina propria and in the deeper vascular area and were not restricted to the glandular area. This observation suggested that the IgE+ cells might not belong to the B cell family. Various cells, such as mast cells and basophils,³¹ have IgE receptors on their surface. Double-staining for IgE and mast cell tryptase revealed almost all IgE+ cells to be mast cells. A few IgE+, but tryptase-negative, cells with round nuclei were identified; however, it could not be determined whether those rare cells were plasma cells or other mononuclear cell bearing low affinity IgE-receptors. Thus IgE-producing

ing plasma cells are rare, if present at all. Indeed, Ganzer and Bachert³² reported that IgE+ cells could not be costained with CD19 (B cell) or CD38 (plasma cell, activated T cell) in the nasal mucosa of patients with allergic rhinitis. However, IgE+ CD38+ cells were seen in the tonsils and lymph nodes, which suggests that IgE is produced in lymphoid tissues and then transported to the nasal mucosa.³² Indeed, others have demonstrated that the number of IgE+ cells in the nasal mucosa approximated the number of toluidine blue+ cells in normal subjects and those with bacterial or allergic rhinitis.³³ In our study IgE+ cells accounted for one third of the total number of mast cells. It is possible that some IgE molecules on mast cells were lost during the tissue preparation and/or that some mast cells possess too few IgE molecules to be detected by immunohistochemical staining.

The existence of mast cell heterogeneity is widely recognized. Mast cells may be categorized by their protease content. Tryptase-positive, chymase-negative mast cells (MC_T) are primarily mucosal type mast cells, and tryptase-positive, chymase-positive mast cells (MC_{TC}) are primarily connective tissue type mast cells.³⁴ It is reported that the distribution of MC_T and MC_{TC} is influenced by the local environment and by pathologic conditions.³⁵ In the present study intraepithelial mast cells were predominantly MC_T, which is consistent with previous reports that demonstrate mucosal type mast cells in nasal scrapings.³⁶ MC_{TC} were more predominant in the lamina propria than MC_T, which is in contrast to a previous report that demonstrated equal numbers of MC_T and MC_{TC}.³⁷ It is possible that unrecognized pathologic conditions in either of the studies might have influenced the results. Although the mediator contents and the biologic responses of the two types of mast cells have been extensively analyzed,³⁶ the difference between their *in vivo* roles is still unclear. The observation that many mast cells are present in the nasal mucosa, as well as in other tissues even under normal conditions, suggests a homeostatic role for mast cells. Unstimulated nasal lavage fluids contain considerable amounts of histamine (data not shown). Thus histamine may be instrumental in regulating vascular permeability and vascular tone in the nasal mucosa. Indeed, unlike mucosal type mast cells, connective tissue type mast cells have been demonstrated to respond to non-IgE-mediated stimuli, such as substance P and complement C5a,³⁸ both of which are produced endogenously

in human beings. Further investigation is required to determine the physiologic role of mast cells in the mucosa.

In this study we quantified resident inflammatory cells in human nasal mucosa by means of immunohistochemical techniques. Although functional studies were not performed, the analysis of T-cell receptor and Ig-bearing cells offers some insights into their functions. This information may now be used to examine changes in inflammatory cell populations in various inflammatory diseases that affect the nasal mucosa, such as allergic rhinitis.

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