

Nasal polyps: Effects of seasonal allergen exposure

Paul K. Keith, MD,^a Mary Conway, RN,^b Susan Evans, BA,^b
Dennis A. Wong, MD,^a Gloria Jordana, MD,^a David Pengelly, PhD,^c and
Jerry Dolovich, MD^b Hamilton, Ontario, Canada

Nasal polyps are characterized by chronic eosinophilic inflammation and often coexist with rhinitis and asthma. Many patients with polyps have no detectable allergy, and it is considered that allergy, at least in many cases, is not relevant to polyp pathogenesis. To explore the association of nasal polyps with allergy, 16 patients with polyps and ragweed allergy (PRW+) and 16 patients with polyps who were not allergic to ragweed (PRW-) were compared with patients without polyps, 16 who were allergic to ragweed (NPRW+) and 16 who were not allergic to ragweed (NPRW-), before and during the ragweed season. The level of ragweed allergy was comparable in the PRW+ and NPRW+ populations as determined by ragweed skin test wheal diameter, ragweed IgE RAST percent binding, and total serum IgE. Symptom scores before the ragweed season recorded on visual analog scales for the symptoms of blockage, sneezing, decreased smell, itch, postnasal drip, and runny nose were high in patients in the PRW+ and PRW- groups and did not change during ragweed season. Mean symptom scores were low in the NPRW+ group before ragweed season and increased during the season to levels similar to those of patients in the PRW+ and PRW- groups. Preseason nasal lavage albumin concentration was higher in subjects with polyps than those without polyps (58.5, 98) versus (13.6, 15 µg/ml) ($p = 0.02$) and did not change significantly in any group with seasonal exposure. Data are presented as mean, 1 SD; comparisons are made with unpaired t tests. The preseason percent eosinophils in the nasal irrigation fluid was higher in the subjects with polyps than in subjects without polyps (18, 25) versus (0.9, 2%), ($p = 0.0002$) and changed little for patients in the PRW+ group during ragweed season. In the NPRW+ group the percent eosinophils increased greatly from low preseason counts, (comparison made with paired t test) (2.2%) to 28, 33% ($p = 0.02$), the latter comparable to those of subjects with polyps. The nasal lavage eosinophil cationic protein concentration out of season was higher in subjects with polyps than in those without polyps (9.7, 10) versus (1.4, 1.0) ($p = 0.0001$) and did not change in subjects with polyps during seasonal exposure. Nasal resistance was measured by head-out body plethysmography. It was slightly higher in subjects with polyps than in those without polyps out of season. In conclusion, highly ragweed-allergic patients with polyps had symptoms and elevated markers of nasal mucosal inflammation (percent eosinophils and eosinophil cationic protein and albumin concentrations) out of season, and these did not increase in relation to natural seasonal allergen exposure. Ragweed allergic subjects with no polyps had large seasonal increases in symptoms and nasal irrigation fluid percent eosinophils. We conclude that ongoing conditions, perhaps nasal airway mucosal inflammation, in nasal polyposis may lead to a loss of susceptibility to potential additional effects of inhaled allergen. (J ALLERGY CLIN IMMUNOL 1994;93:567-74.)

Key words: Nasal polyps, ragweed, eosinophil, eosinophil cationic protein, albumin

From the Departments of ^aMedicine, ^bPaediatrics, and ^cEngineering, McMaster University, Hamilton, Ontario, Canada. Supported by an operating grant of the Medical Research Council of Canada and a grant from Astra Pharma Inc., Mississauga, Ontario.

Received for publication Jan. 25, 1993; accepted Aug. 20, 1993.

Reprint requests: Jerry Dolovich, MD, 3V41, McMaster University, 1200 Main St. West, Hamilton, Ontario, L8N 3Z5, Canada.

Copyright © 1994 by Mosby-Year Book, Inc.

0091-6749/94 \$3.00 + 0 1/1/50957

Polyps are smooth, gelatinous, grape-like and often multiple structures of unknown cause,¹ which extend down into and can lead to obstruction of the nasal cavity. They commonly arise from the mucosal lining of the ethmoid sinuses, which prolapses into the nasal cavity. Less frequently, they can arise from the maxillary or sphenoid sinuses or nasal turbinates. Nasal polyposis often coexists with asthma, rhinitis, and sinusitis. Clinically, it is characterized mainly by

Abbreviations used

ECP:	Eosinophil cationic protein
EO%:	Percent eosinophils
NPRW+:	No polyps, ragweed allergy
NPRW-:	No polyps, no ragweed allergy
PRW+:	Polyps, ragweed allergy
PRW-:	Polyps, no ragweed allergy

nasal congestion and anosmia, but there can also be sneezing and rhinorrhea. Like asthma, nasal polyps have been ascribed to infection and allergen exposure.

The relationship between nasal polyps and allergy remains unclear.² In a series³ of 249 patients who had nasal polypectomy in the Hamilton area, 66% had at least one positive allergy skin test result. In a retrospective study⁴ of 6037 patients referred for allergy assessment, 211 had nasal polyps; 55% of them had positive allergy test results, whereas 70% of the referrals without polyps were atopic. From this data, one could calculate that of the 6037 patients referred to one allergy practice, 2.8% of the atopic patients had polyps and 5.2% of the nonatopic patients had polyps. It has been reported⁵ that multiple positive skin test results are no more common than expected in patients with nasal polyps (25%), making the presence of allergy seem coincidental. Fifty-four percent of the patients with polyps studied had at least one positive allergy skin test result, and tree and ragweed pollen were not tested. It was concluded that the prevalence of multiple positive test results in the patients with polyps was the same as that in the general population. The presence of allergy did not correlate with the number of repeat polypectomies.⁵ Caplin et al.⁶ examined 3000 consecutive atopic patients and found that only 0.5% had polyps; he concluded that allergy was likely unimportant in the pathogenesis. Previous reviews have pointed to allergy as a possible exacerbating factor of symptoms, but there seem to be no direct studies of the relationship. In a study of patients undergoing nasal polypectomy,⁷ 19 of 24 had demonstrable IgE antibody in polyp fluid, suggesting that allergy is involved in polyp pathogenesis; only nine of these 19 had corresponding positive skin test results; this suggests that relying on skin tests may cause us to overlook allergy as an underlying cause. Blood eosinophil counts were studied by logistic regression methods in 249 patients who

required nasal polypectomy.³ There was no relationship between the blood eosinophil counts and atopy, as indicated by skin test reactivity or serum IgE concentration.

Polyp histology typically shows chronic, eosinophilic inflammation. The pseudostratified ciliated epithelium is commonly damaged⁸ and sometimes shows squamous metaplasia. The stroma is typically edematous. The inflammatory cell infiltrate generally includes eosinophils, lymphocytes, plasma cells, and mast cells. These inflammatory changes are similar to those observed in the inflamed bronchial mucosa in asthma⁹⁻¹¹ and are characteristic of polyp tissue whether or not allergy is present.

The nasal turbinate mucosa in patients with nasal polyposis may not be markedly abnormal.¹² However, we have observed an increased number of mast cells in the epithelium of turbinate mucosa of patients with polyposis, as well as in the epithelium of the polyp itself.¹³ The accumulation of mast cells and eosinophils in the tissue and in the nasal cavity in nasal polyposis produces conditions that might be expected to enhance a superimposed allergic reaction.

The purpose of this study was to assess the effects of seasonal ragweed allergen exposure on allergic and nonallergic patients with and without nasal polyps.

METHODS

The study was approved by the ethics committee of the McMaster Medical Centre. Informed consent was obtained from all subjects. Adult subjects were evaluated during two study periods, winter 1991 (February 12 to April 26, 1991) and the ragweed season 1991 (August 19 to September 12, 1991). Sixteen normal, nonallergic subjects (NPRW- group), 16 patients with nasal polyps who were allergic to ragweed pollen (ragweed skin prick test wheal > 3 mm) (PRW+ group), 16 patients with nasal polyps who were not allergic to ragweed (ragweed skin test negative) (PRW- group), and 16 patients with a history of seasonal allergic rhinitis during ragweed pollen season, a positive ragweed allergy skin test result (wheal > 3 mm), and no polyps (NPRW+ group). None of the patients who were not allergic to ragweed had a positive skin test result to the 14 inhalant antigens tested. The clinical characteristics of the subjects are summarized in Table I.

Subjects were excluded if they were under 18 years of age or had concomitant disease such as cystic fibrosis, immunodeficiency, or another major illness. The following medications, which could potentially interfere with evaluation, could not be used for 2 weeks before

TABLE I. Patient characteristics

	Group			
	NPRW +	NPRW -	PRW +	PRW -
Sex				
Male	11	14	8	7
Female	5	2	8	9
Age (yr)				
Mean	46	47	37	38
Min-max	23-68	23-66	23-63	21-61
No. of positive skin tests				
Median	3	0	3.5	0
Min-max	1-13		1-10	
Asthma	7	2	4	0
Conjunctivitis	9	3	12	1
Eczema	2	1	1	1
History of ASA intolerance	3	2	3	3
No. of polypectomies				
Mean	2.6	4.4		
Min-max	0-19	0-20		
Polyp present				
Right	16	15		
Left	15	16		
Polyp size (cm) (median, min-max)				
Right	0.5, 0.25-1.5	1.0, 0-1.5		
Left	0.75, 0-1.5	1.25, 0.25-1.5		
Polyp obstruction (0-3) (median, min-max)				
Right	1, 0-3	2, 0-3		
Left	1, 0-3	2, 0-3		

Min-max, Minimum to maximum; *ASA*, acetylsalicylic acid.

each of the two periods of evaluation: antihistamine (astemizole, 4 weeks), corticosteroid (intranasal or systemic), decongestant (all modes of administration).

Subjects were studied on 2 consecutive days at approximately the same time each day during the winter before the pollen season and on 2 consecutive days during the following ragweed pollen season. At the initial visit a case history was taken. Allergy skin prick tests were performed (if not done during the preceding year) with extracts of 14 common inhalant allergens, which were considered most prevalent in the area, obtained from commercial sources.

Symptoms experienced during the preceding 24-hour period were evaluated with a visual analogue scale 16.5 cm long with no divisions. Subjects were asked to place an X where they believed their symptoms lay between two extremes; for example, for nasal obstruction they were asked to estimate the blockage in relation to the extreme possibilities of the nose totally obstructed or

totally clear. The distance from the well, nonaffected end of the line was measured in centimeters and recorded as the score. Physical examination of the nasal cavities included an estimate of polyp length (0.5, 1.0, 1.5 cm) and obstruction caused by polyps (none, mild, moderate, severe).

Nasal secretions were collected in a manner outlined by Powell et al.¹⁴ Five milliliters of phosphate-buffered saline without calcium or magnesium (Gibco, Grand Island, N.Y.) warmed to 37° C was instilled into each nostril while the subject's neck was held extended approximately 30 degrees from horizontal and the head held back. The saline solution was held in the nasal cavity for 10 seconds and then drained into a plastic specimen cup. This was done twice each day, separated by 15 minutes, and the two samples were pooled. The fluid collected each day was centrifuged at 1700 rpm for 20 minutes at 4° C. The supernatant samples were sterilized by filtering with an Acrodisc (Gelman Sci-

TABLE II. Reproducibility (intraclass correlation coefficient)

	Preseason	In season
Symptoms: Visual analog scale		
Nasal blockage	0.83	0.82
Nasal discharge	0.79	0.85
Nasal itch	0.66	0.72
Sneezing	0.69	0.69
Postnasal drip	0.78	0.87
Decreased sense of smell	0.96	0.91
Lavage		
Albumin	0.77	0.86
Total protein	0.96	0.82
ECP	0.76	0.62
EO %	0.92	0.73

The intraclass correlation coefficients are summarized for both the preseason and in season measurements. There is a high degree of agreement between repeated measures for both nasal lavage and visual analogue scale before and in season.

ences, Inc., Ann Arbor, Mich.) 0.2 μ m low protein binding filter. Phosphoramidon (Peninsula Laboratories, Inc., Belmont, Calif.) a protease inhibitor, was added to the samples collected to a final concentration of 10^{-6} mol/L. Aliquots of 0.5 ml were made and stored at -70° C and assayed together at a later date. Albumin in the nasal secretions was measured by nephelometry (Kallestad QM 300; Sanofi Diagnostics Pasteur, Inc., Redmond, Wash.). Eosinophil cationic protein (ECP) was measured by double antibody radioimmunoassay (ECP RIACT, Pharmacia, Uppsala, Sweden).

The cell pellet was resuspended to 300 μ l. Cytospin preparations were made and stained with chromotrope 2R (Sigma Chemical Co., St. Louis, Mo.) diluted in phenol and distilled water. Eosinophils were enumerated as a percentage of the total nucleated cell count.

Blood was taken during one visit before and one visit during the ragweed season. Blood eosinophils were counted manually. Total serum IgE was measured by the Phadebas method (Pharmacia). IgE antibody to ragweed was measured by RAST (Pharmacia).

Data were transcribed from the records and stored in an IBM-compatible computer with dBase III software (Ashton-Tate, Torrance, Calif.). Analysis was carried out initially with repeated measures analysis of variance (BMDP 2V; BMDP Statistical Software, Inc., Los Angeles, Calif.). Student's *t* tests were then performed, with paired and nonpaired tests as appropriate. A *p* value of less than 0.05 was considered significant. The mean value for measurements obtained on consecutive days was used to compare the groups. Reproducibility of consecutive day measurements was assessed by intraclass correlation coefficient,¹⁵ which combines a measure of correlation with

a test of the difference of means of repeated measures for measures repeated on days 1 and 2 before ragweed season and on days 3 and 4 during ragweed season.

RESULTS

Allergy

The level of ragweed allergy was comparable in the PRW+ and NPRW+ groups, respectively, before ragweed season as determined by ragweed skin test wheal diameter (5.9, 3.0 mm) versus (6.8, 2.5 mm), ragweed IgE RAST percent binding (12.4%, 15.2%) versus (16.6%, 15.0%) and total serum IgE (82.5, 86 μ g/ml) versus (76.9, 58 μ g/ml). Data are presented as mean, 1 SD; comparisons are made with unpaired *t* tests.

Symptoms

Intraclass correlation coefficients for the total population for symptoms assessed by repeated use of the visual analog scale before and during ragweed season, respectively, were high (0.66 or greater) and are summarized in Table II.

Season-related symptom score changes in the four groups are shown for nasal blockage and nasal itch in Figs. 1 and 2. In the PRW+ and PRW- groups scores recorded on visual analog scales for the symptoms of blockage, sneezing, decreased smell, itch, and runny nose were high in the winter and did not change in the ragweed pollen season. These scores were low in the NPRW+ group before ragweed season, and this was the only group to have a significant increase in symptom scores during the season. This occurred with all symptoms. During the ragweed season subjects in the NPRW+ group had higher scores than subjects in the PRW+ group for nasal itch (9, 4 vs 6, 5, *p* = 0.10) and sneezing (7, 4 and 6, 5, not significant).

Nasal lavage

Intraclass correlation coefficients for repeated measurements of nasal lavage albumin, total protein, ECP, and percent eosinophils (EO%) were 0.62 or greater and are summarized in Table II. Differences between subjects with and without polyps are recorded in Table III, and season-related differences are shown in Fig. 3.

The preseason EO% in the nasal irrigation fluid was higher in the subjects with polyps than in those without polyps and changed little for patients with polyps during ragweed season. Lavage EO% was the only lavage variable to show

NASAL BLOCKAGE: VISUAL ANALOGUE SCALE (0-16)

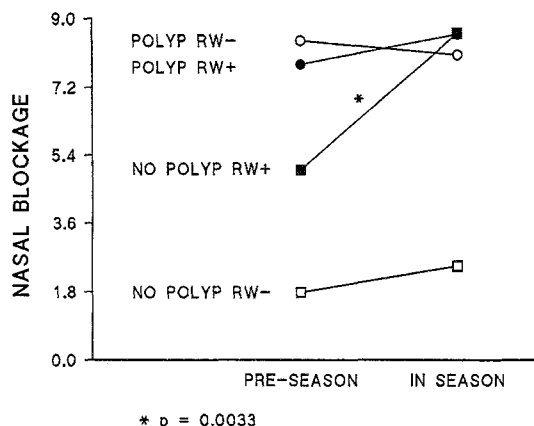


FIG. 1. This graph plots the means for each of the four groups before (*pre-season*) and in season for the symptom of nasal blockage on visual analogue scale. Only the NPRW+ group had a significant change in symptoms ($p = 0.0033$), increasing to the level of the polyp groups.

NASAL ITCH: VISUAL ANALOGUE SCALE (0-16)

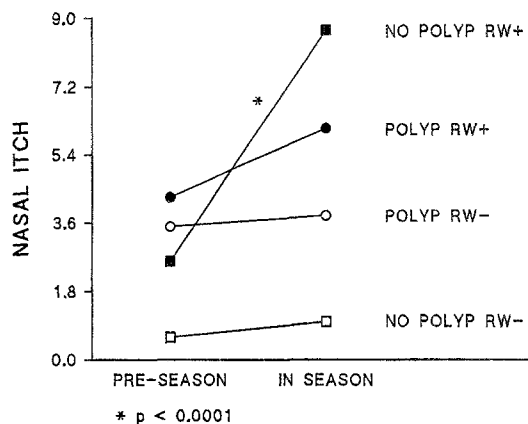


FIG. 2. This graph plots the means for each of the four groups before (*pre-season*) and in season for the symptom of nasal itch on visual analogue scale. Only the NPRW+ group had a significant change in symptoms ($p < 0.0001$).

a significant interaction on analysis of variance. In the NPRW+ group the EO% increased greatly from low preseason to high in-season counts and were comparable to in-season EO% counts in subjects in the PRW+ and PRW- groups. In the NPRW- group there was no change in EO% from before season to in-season. There was a difference between NPRW+ and NPRW- groups in season (28, 33 vs 1.7, 3.1, $p = 0.02$).

Nasal lavage ECP concentration was higher in subjects with polyps than in those without and did not change in subjects with polyps and seasonal exposure. There was a seasonal increase in ECP only in subjects in the NPRW+ group (from 1.6, 1.3 to 6.3, 13, not significant).

Preseason nasal lavage albumin concentration was higher in subjects with polyps than in those without and did not change significantly in any group with seasonal exposure. There was no difference in albumin concentration between the two polyp groups before or during the ragweed season, but there was a difference between the NPRW+ and NPRW- groups both before ($p = 0.01$) and during the season ($p = 0.04$).

The mean lavage albumin:total protein ratio was greater in patients with polyps than in subjects without polyps out of season ($p = 0.0001$) (Table III), suggesting a component of plasma exudation.

EOSINOPHIL % IN NASAL LAVAGE

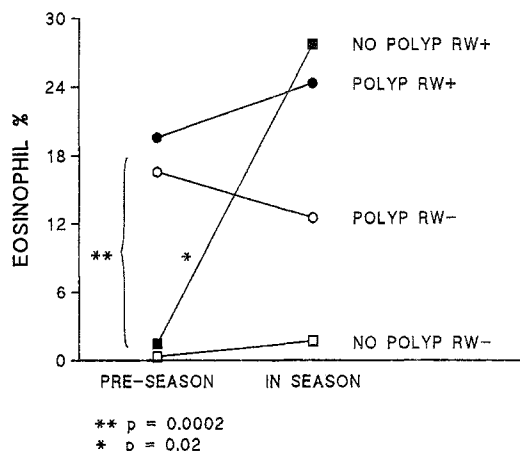


FIG. 3. This graph plots the means for each of the four groups for EO% in nasal lavage. Only the NPRW+ group had a significant change. There was also a significant difference between the groups with polyps and the groups without polyps before ragweed season.

Nasal resistance

Nasal resistance was measured by head-out body plethysmography.¹⁶ The total nasal resistance was slightly, significantly higher when subjects with polyps were compared with subjects without polyps out of season¹⁷ ($p = 0.03$) but not in season. When the ragweed-allergic groups

TABLE III. Comparison of subjects with and without polyps before ragweed season

	Polyp	No polyp	p Value*
EO %	18, 25	0.9, 2	0.0002
Albumin ($\mu\text{g/ml}$)	58.5, 98	13.6, 15	0.02
Albumin: Total protein	15, 8	8, 5	0.0001
ECP ($\mu\text{g/ml}$)	9.7, 10	1.4, 1.0	0.0001

Values are expressed as mean, 1 SD.

*Determined by unpaired *t* test.

(PRW+ and NPRW+) were compared, there was no difference before or during ragweed season.

DISCUSSION

Subjects with nasal polyps had symptoms and elevated markers (EO% and ECP and albumin concentrations) of mucosal inflammation in nasal irrigation fluid both before and during the ragweed season. In ragweed-allergic subjects with polyps, we did not find an increase in the symptoms or markers of inflammation from natural seasonal ragweed exposure. Ragweed-allergic subjects with hayfever who did not have polyps had large seasonal increases in symptoms and EO%. The difference between ragweed-allergic subjects with and without polyps cannot be attributed to the level of allergy to ragweed, which was comparable in the two groups.

The lack of seasonal increase in albumin and ECP concentrations in ragweed-allergic subjects with hayfever with no polyps was unexpected because previous studies have demonstrated increased albumin¹⁸ and ECP concentrations after nasal allergen challenge in allergic patients. The conditions were probably comparable to those in a previous study¹⁹ in which nasal lavage was performed daily throughout the birch-pollen season; albumin and ECP concentrations did not rise significantly until 14 days after the onset of symptoms and presence of birch pollen in the air. Most of our ragweed-allergic patients underwent lavage before they had experienced symptoms for 2 weeks. In a clinical trial of therapy in 20 allergic patients²⁰ no increase in ECP concentration was found during seasonal exposure to birch pollen, but there was a decrease with intranasal steroid. With no therapy, measurements were 950 and 1170 $\mu\text{g/L}$ before the season and 1160 $\mu\text{g/L}$ during the season. With therapy the level was 530 $\mu\text{g/L}$. These values are higher than reported here, probably because of our use of a larger volume of saline solution in the irrigation. In this study the

patients experienced seasonal ragweed pollen exposure sufficient to produce symptoms, but the exposure was probably not sufficient to elicit a maximal mucosal reaction.

Subjects with nasal polyps had ongoing symptoms and evidence of mucosal inflammation throughout the year. The higher nasal irrigation fluid albumin concentration and albumin:total protein ratio in patients with polyps than in patients without polyps suggests plasma exudation from the polyps or from the nasal mucosa.²¹ The increased albumin concentrations in patients with polyps may be due to greater surface area of the mucosa in patients with nasal polyps.

The higher concentration of ECP in nasal irrigation fluid from patients with polyposis before and during ragweed season suggests involvement of activated eosinophils in the disease. ECP is one of the eosinophil products considered to contribute to tissue changes. ECP effects include stimulation of airway mucus secretion²² and respiratory epithelial cell exfoliation.²³ In a histochemical and immunohistochemical study of the cellular content of nasal polyps and control turbinate tissue, the major difference that we found was an 8- to 10-fold higher tissue density of eosinophils, 70% of which were activated (positive with EG2 anti-serum) in the polyps.²⁴

The lack of effect of seasonal exposure to ragweed in the PRW+ group was unexpected. Before the study, it was reasoned that because there is an accumulation of eosinophils²⁴ in polyps and increased numbers of epithelial mast cells²⁵ in nasal polyps and adjacent turbinate epithelium, this might have a priming effect, producing an exaggerated response to antigen. The lack of increase in symptoms and inflammatory markers in this group may indicate that the ongoing cellular accumulation and activation is already maximal. Another possibility is that chronic inflammation elicits mechanisms of natural inhibition such as the formation of inhibitory cytokines or soluble receptor molecules,²⁶ which maintain a plateau in the level

of symptoms and inflammatory response markers. Transforming growth factor- β is a potent inhibitory cytokine, and cellular transforming growth factor- β protein and gene product have been found to be increased in nasal polyp tissue compared with nasal turbinate tissue.²⁷ Inhibitory mechanisms could affect cellular activation. In isolated mast cells from polyp epithelium, an unresponsiveness to anti-IgE has been demonstrated.²⁸ It is also possible that the nasal mucosa in subjects with polyps did not encounter the same amount of antigen stimulation as that in patients without polyps because of ongoing nasal obstruction and consequent mouth breathing, which leads to less pollen exposure. However, the nasal resistance measurement was not appreciably higher in patients in the PRW+ group than in patients in the NPRW+ group before the season, and although the level was higher for the PRW+ group during the season, differences between the groups were not significant. The reduced capacity of the nasal mucosa to respond could be further examined by nasal allergen challenge experiments.

The present study suggests that seasonal allergen exposure in patients with nasal polyps does not enhance the level of expression of the disease. The relevance of allergen exposure might further be explored by studies of careful avoidance of allergen exposure. The present findings are consistent with the view that chronic eosinophilic mucosal inflammatory disease in nasal polyposis involves self-sustaining mechanisms, largely independent of allergen stimulation of polyp and nasal turbinate mucosa; however, these findings do not exclude the possibility that the processes are maximally activated, and any added effect of an allergic provocation is not measurable.

We thank Roxanne Kolendowicz and Adele Guergis-Gabardo for technical assistance.

REFERENCES

- Slavin RG. Nasal polyps and sinusitis. In: Middleton E Jr, Reed CE, Ellis EF, Adkinson NF Jr, Yunginger JW, eds. *Allergy: principles and practice*. 3rd ed. St. Louis: CV Mosby, 1988:1291-1304.
- Kern RA, Schenck HP. Allergy: a constant factor in the etiology of so-called mucous nasal polyps. *J Allergy* 1933; 4:485-97.
- Wong D, Dolovich J. Blood eosinophilia and nasal polyps. *J Rhinol* 1992;6:195-8.
- Settipane G, Chafee F. Nasal polyps in asthma and rhinitis. *J ALLERGY CLIN IMMUNOL* 1977;58:17-21.
- Drake-Lee AS. Nasal polyps. In: Mackay IS, ed. *Rhinitis: mechanisms and management*. London: Royal Society of Medicine, 1989:141-52.
- Caplin I, Haynes TJ, Spahn J. Are nasal polyps an allergic phenomenon? *Ann Allergy* 1971;29:63.
- Small P, Barrett D, Frenkiel S, Rodhon L, Cohen C, Black M. Local specific IgE production in nasal polyps associated with negative skin tests and serum RAST. *Ann Allergy* 1985;55:736-9.
- Wladislavsky-Waserman P, Kern EB, Holley KE, Eisenbrey AB, Gleich GJ. Epithelial damage in nasal polyps. *Clin Allergy* 1984;14:241-7.
- Dolovich J, Ohtoshi T, Jordana M, Gauldie J, Denburg J. Nasal polyps: local inductive microenvironment in the pathogenesis of the inflammation. In: Mygind N, Pipkorn U, Dahl R, eds. *Rhinitis and asthma—similarities and differences*. Copenhagen: Munksgaard, 1990:233-41.
- Djukanovic R, Wilson JW, Britten KM, et al. Quantitation of mast cells and eosinophils in the bronchial mucosa of symptomatic atopic asthmatics and healthy control subjects using immunohistochemistry. *Am Rev Respir Dis* 1990;142:863-71.
- Azzawi M, Bradley B, Jeffery PK, et al. Identification of activated T lymphocytes and eosinophils in bronchial biopsies in stable atopic asthma. *Am Rev Respir Dis* 1990; 142:1407-13.
- Stoop AE, Hameleers DMH, van Run PEM, Biewenga J, van der Baan S. Lymphocytes and non-lymphoid cells in the nasal mucosa of patients with nasal polyps and of healthy persons. *J ALLERGY CLIN IMMUNOL* 1989;84:734-41.
- Ruhno J, Howie K, Anderson M, et al. The increased number of epithelial mast cells in nasal polyps and adjacent turbinates is not allergy-dependent. *Allergy* 1990;45: 370-4.
- Powell KR, Shorr R, Cherry JD, Hendley JO. Improved method for collection of nasal mucus. *J Infect Dis* 1977; 136:109-11.
- Kramer MS, Feinstein AR. Clinical biostatistics: LIV. The biostatistics of concordance. *Clin Pharmacol Ther* 1981; 29:111-23.
- Niinimaa V, Cole P, Mintz S, Shephard RJ. A head-out exercise body plethysmograph. *J Appl Physiol* 1979;47: 1336-9.
- Keith PK, Wong DA, Anderson M, Conway M, Dolovich J. Reproducibility of nasal peak inspiratory flow (NPIF) and nasal airway resistance (NAR) measures [Abstract]. *Ann Allergy* 1992;68:90.
- Brofeldt S, Mygind N, Sorensen CH, Readman AS, Marriott C. Biochemical analysis of nasal secretions induced by methacholine, histamine, and allergen provocations. *Am Rev Respir Dis* 1986;133:1138-42.
- Svensson C, Andersson M, Persson CGA, Venge P, Alkner U, Pipkorn U. Albumin, bradykinins, and eosinophil cationic protein on the nasal mucosal surface in patients with hay fever during natural allergen exposure. *J ALLERGY CLIN IMMUNOL* 1990;85:828-33.
- Linder A, Venge P, Deusbl H. Eosinophil cationic protein and myeloperoxidase in nasal secretion as markers of inflammation in allergic rhinitis. *Allergy* 1987;42:583-90.
- Raphael GD, Meredith SD, Baraniuk JN, Druce HM, Banks SM, Kaliner MA. The pathophysiology of rhinitis. II. Assessment of the sources of protein in histamine-induced nasal secretions. *Am Rev Respir Dis* 1989;139: 791-800.
- Lundgren JD, Davey RY, Lundgren B, et al. Eosinophil cationic protein stimulates and major basic protein inhibits airway mucus secretion. *J ALLERGY CLIN IMMUNOL* 1991;87: 689-98.

23. Motojima S, Frigas E, Loegering DA, Gleich GJ. Toxicity of eosinophil cationic proteins for guinea pig tracheal epithelium in vitro. *Am Rev Respir Dis* 1989;139:801-5.
24. Kanai N, Ohno I, Dolovich J, Jordana M. Immunohistochemical localization of GM-CSF in human inflamed upper airways. Effect of topical nasal steroid. *Am J Respir Cell Mol Biol* (in press).
25. Ruhno J, Howie K, Anderson M, et al. The increased number of epithelial mast cells in nasal polyps and adjacent turbinates is not allergy-dependent. *Allergy* 1990;45:370-4.
26. Fanslow WC, Sims JE, Sassenfeldt H, et al. Regulation of alloreactivity in vivo by a soluble form of the interleukin-1 receptor. *Science* 1990;248:739-42.
27. Ohno I, Lea RG, Flanders KC, et al. Eosinophils in chronically inflamed human upper airway tissues express transforming growth factor gene (TGF β_1). *J Clin Invest* 1992;89:1662-8.
28. Finotto S, Dolovich J, Denburg J, Jordana M, Marshall J. Functional heterogeneity of mast cells isolated from different microenvironments within nasal polyp tissue. *Clin Exp Immunol* (in press).

ANNOUNCEMENT

AMERICAN BOARD OF ALLERGY AND IMMUNOLOGY

The American Board of Internal Medicine,
and The American Board of Pediatrics
are pleased to announce

CERTIFICATION IN CLINICAL AND LABORATORY IMMUNOLOGY

Examination Date: Monday, October 10, 1994

St. Louis, Missouri

Registration: through April 30, 1994

Completed applications must be postmarked by April 30, 1994. A nonrefundable late fee will apply to those applications received after the close of registration and before the cancellation deadline of August 1, 1994.

Please address all requests to: Herbert C. Mansmann, Jr., MD, Executive Secretary, American Board of Allergy and Immunology, 3624 Market St., Philadelphia, PA 19104-2675; phone: 215-349-9466, fax: 215-222-8669.