

Deposition of ragweed pollen and extract on nasal mucosa of patients with allergic rhinitis: Effect on nasal airflow resistance and nasal mucus velocity

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This study was undertaken to ascertain whether nasal mucus velocity (NMV) could be altered by short-term exposure to antigen. Asymptomatic patients with a history of allergic rhinitis who had a positive cutaneous reaction to ragweed extract were investigated. The plan was to achieve approximately a fourfold elevation of nasal airflow resistance (NAR) with antigen challenge and then obtain serial measurements of NAR and NMV. NMV was not significantly altered when the antigen was introduced by nasal inhalation of (1) ragweed pollen grains, (2) nebulized ragweed extract for 10 breaths, and (3) nebulized ragweed extract for 30 min on each of 3 successive days. When ragweed extract was introduced by direct instillation of the solution into the nose, NMV fell below baseline values at either 0.5 or 1.5 hr, or at both times after administration. Persistence of impairment of mucociliary transport at a time when nasal airway constrictor response had dissipated suggested that a chemical mediator might have been responsible for the alteration of clearance. The failure to demonstrate depression of mucus transport with the inhalation studies might have been due to insensitivity of the radiopaque Teflon disk method or to a qualitatively different allergic reaction to direct instillation of antigen solution.

Patients with symptomatic allergic rhinitis complain of nasal itching, sneezing, and stuffy or runny nose. Itching and sneezing are manifestations of mucosal irritation as reflected by a boggy purplish mucosa. Together with retained secretions, the swollen mucosa produces an increase of nasal airflow resistance (NAR) and accounts for the nasal stuffiness. Runny nose is related to excessive secretions in the presence of intact ciliary function, impaired mucus transport, or a combination of the two.^{1, 2} Inhalation of ragweed pollen grains by patients who have cutaneous sensitivity to ragweed extract produces a rise in NAR.³ The effects of ragweed pollen on the muco-

ciliary system in allergic rhinitis including histology of the ciliated epithelium, ciliary beating, and mucus transport have not been fully investigated. In sensitized rabbits and guinea pigs, deposition of pollen grains produces destruction of nasal ciliated epithelium 10 to 30 min later,⁴ but these alterations have not been observed in humans with light and scanning electron microscopy.⁵ In patients with allergic rhinitis who have chronic symptoms, the number of ciliated cells appears to be diminished.⁶ In sensitized rabbits and guinea pigs, nasal ciliary beating ceases 1 to 10 min after deposition of pollen grains,⁷ but in humans, mucus transport measured by the saccharin test is not altered by such a challenge.⁵ However, the saccharin test is semiquantitative and is of questionable usefulness for assessing transient changes in mucus transport; further, there is a poor correlation with NMV measured from the motion of radiopaque Teflon disks.⁸ Because of this uncertainty as to whether nasal mucus transport is intact in allergic rhinitis after short-term challenge with antigen, we designed the present study. Nasal inhalation of ragweed pollen

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TABLE I. Characteristics of patients

Patient No.	Age (yr)	Sex	Experiments*	Skin reaction to ragweed	Cutaneous wheals, induration (mm ²)				Number of pollen grains to produce 2- to 4-fold rise in NAR
					Mt. cedar	Cedar elm	Sagebrush	Russian thistle	
1	23	F	1	4					79
2	48	F	1	4					3,000
3	58	F	1	4					110
4	33	F	1	4					109
5	28	M	1	4					50
6	31	F	1,2,3,6,7,8	4	63†	55†	320	86†	388
7	22	F	3,4,6,7,8	4	59†	102†	176†	130†	
8	26	M	4	4					
9	22	M	4	4					
10	34	F	2,4,8	4†	38	50	61	33	
11	30	M	2,3,4	4					
12	30	F	2,4	4					
13	36	M	4,5,6,7,8	4	71	79	95	112	
14	22	M	4	4					
15	26	M	3,4,5,6,7,8	4	39	47	231	85	
16	34	M	4	2					
17	34	F	6,7	4†					
18	37	M	2	4					
19	41	F	2,3	2†					
20	25	F	5	3					
21	32	M	5	3					
22	27	F	5,6,7,8	4	78†	47	164	123†	
23	21	F	3,5,6,7,8	2	38	94†	55	42	
24	28	F	5,6,7,8	4	69†	100†	118†	36	
25	21	F	6,7,8	4	59†	33	66	44	
26	33	M	3,7,8	4	75†	104†	148	123	
27	26	M	3						

*1, Ragweed pollen inhalation (Table II); 2, air sham (Table II); 3, nebulized saline (Table III); 4, nebulized ragweed antigen extract to increase NAR fourfold (Table III); 5, nebulized ragweed antigen extract over 3 consecutive days (Table IV); 6, intranasal instillation of phosphate-buffered saline (Table V); 7, intranasal instillation of ragweed antigen extract solution (Table V); 8, intranasal instillation of other antigen extract solution.

†Dilution of antigen at 1:1,000 v/v; assume 1:10,000 v/v dilution otherwise.

grains and nebulized ragweed extract as well as direct instillation of a solution of ragweed extract were studied. Serial measurements of NAR by posterior rhinometry and nasal mucus velocity (NMV) by the radiopaque Teflon disk method were obtained for a period up to 2 hr after challenge.^{8, 9}

MATERIALS AND METHODS

Subjects

Volunteers who had experienced seasonal rhinitis were recruited and enrolled in the study if they displayed a positive cutaneous reaction which was defined as a wheal of at least 6 mm, 15 min after 0.02 ml of 1:10,000 or a wheal of at least 10 mm, 15 min after 0.02 ml of 1:1,000 dilution of ragweed extract (Greer Labs, Inc., Lenoir, N. C.) was injected intradermally. The area of each injected wheal was divided into the area of induration, and the resultant quo-

tient used to quantify cutaneous reactivity on a grading scale of 1 to 4. Each subject was asked specifically about symptoms of allergic rhinitis and none was symptomatic at the times of the various investigations. The age and sex distribution of the subjects, their cutaneous sensitivity, and the amount of antigen utilized to provoke a response are listed in Table I. All subjects gave informed consent and were paid. The study was approved by the Human Rights Committee of Mount Sinai Medical Center.

Nasal mucus velocity

The subject sat in a dental chair so that the hard palate lay within the horizontal plane. NMV was estimated by a method which has been described previously.^{8, 9} Briefly, Teflon disks rendered radiopaque by mixture with 50% bismuth trioxide by volume were gently deposited above the inferior turbinate by dropping them through a 3-mm diame-

**Dilution of ragweed antigen
extract to effect 4-fold rise in NAR**

Aerosol	Instillation
	1:5,000
1:10,000	1:500
1:1,000	
1:10,000	
1:5,000	
1:50,000	
1:10,000	
1:500,000	1:500
1:1,000	
1:50,000	1:5,000
1:10,000	
1:5,000	1:1,000
1:500	
1:5,000	
1:100,000	1:5,000
1:100,000	1:50,000
1:1,000	1:500
	1:500
	1:500

ter polyethylene catheter connected to a 10-ml plastic syringe. The disks were 1 mm in diameter, 0.8 mm in thickness, and 1.8 mg in weight. The nasal passage was visualized in its lateral projection by means of a portable fluoroscopic image intensifier unit connected to a video monitor. The output of a digital clock was also displayed on the monitor. Disk motion was recorded on videotape which was replayed to identify the distance traveled by each disk; a 1-cm length of radiopaque Teflon tape placed on the cheek served as a calibrating image. The axial motion of 5 to 10 Teflon disks not greater than 0.5 cm superior to the middle portion of the hard palate was measured. The NMV was computed from the distance traveled by each disk divided by a standard unit of time, generally 1 min.

Nasal airflow resistance

NAR was obtained by posterior rhinometry.^{2, 9} An air-tight rubber scuba diving mask covered the nose and the eyes. A pneumotachograph (Fleisch No. 2, Godart, Holland) was mounted on the plastic shield of the mask and the pressure difference across the pneumotachograph was measured with a variable reluctance differential gauge (model P7D $\pm$ 0 to 0.2 PSID, Whittaker Corp., Cal.), calibrated to

flow by a rotameter. A polyethylene tube of 6-mm diameter was held firmly between the subject's lips; the distal tip projected along the upper surface of the tongue 7 to 9 cm inside the mouth cavity. This tube was connected to a second variable reluctance differential gauge (model P7D $\pm$ 0 to 0.2 PSID, Whittaker Corp.) along with another tube connected to the other side of the transducer from the mask. The electrical outputs of the pneumotachograph and posterior pharyngeal pressure transducers were displayed on a cathode ray oscilloscope (Tektronix Model 5103N, Beaverton, Ore.). The subject was instructed to breathe solely through his nose and the expiratory NAR was measured over a flow rate of 0.3 L/sec. Five sequential measurements were obtained and the screen image photographed on a single Polaroid print so that the mean value could be computed.

Forced expiratory volume in 1 second

FEV₁ was measured with a rolling-seal dry spirometer (MedScience Electronics, Inc., Model No. 460, St. Louis, Mo.). The value was obtained prior to and serially after each challenge to detect possible lower airway responses. It was planned to terminate a patient study if the FEV₁ fell 20% below baseline prior to reaching the desired increase in NAR.

Inhalation of ragweed pollen grains

After obtaining baseline measurements of NMV, NAR, and FEV₁, short ragweed pollen was administered by the technique of Connell.³ Six patients participated in this study (Table I). The apparatus employed to deliver the pollen was obtained from Dr. Connell and consisted of a rotating 55-L glass sphere charged with pollen grains and approximately a hundred 0.25-diameter paper disks placed in the sphere. These disks, which were in constant motion, served to agitate the air and also, by brushing the surface, aided in resuspending the pollen adhering to the glass surface. The amount of pollen delivered to the subject depended upon the resistance of bored corks which allow either greater or lesser amounts of room air to dilute the pollen inhaled from the rotating glass sphere. The subject inhaled from a nozzle connected to the pollinator via one nostril utilizing a slow inspiratory vital capacity maneuver. Simultaneously, a glass microscopic slide coated with petroleum jelly which had been placed in the air stream was employed to collect an aliquot of the pollen grains in order to provide an estimate of the quantity administered.³ Two measurements were obtained from the slide according to the method of Connell³ and the reproducibility fell within 25% of the mean. NAR was measured 5 and 10 min after inhalation. The aim was to increase overall NAR from two to four times the baseline value. The inhalations were continued by doubling the number of breaths with each challenge until this point was reached. Measurements of FEV₁ were also obtained after each challenge. When the end point for NAR was attained, both NAR and NMV were measured and repeated 1 and 2 hr later. To account for variability in NMV and NAR due to repeated measurements over time the testing procedure was

TABLE II. Effects of nasal inhalation of ragweed pollen grains and room air sham

	Baseline	After exposure		
		Immediate	1 hr	2 hr
<i>Ragweed pollen inhalation*</i>				
NAR (cm H ₂ O/L/sec)	2.1 (0.2)	5.9 (0.9)†	3.0 (0.2)	4.4 (0.3)†
NMV (mm/min)	5.0 (0.8)	5.6 (1.3)	4.5 (1.5)	3.7 (0.9)
FEV ₁ (L/TPS)	2.9 (0.3)	3.0 (0.3)	2.9 (0.3)	2.9 (0.3)
<i>Air sham‡</i>				
NAR (cm H ₂ O/L/sec)	3.0 (0.5)	—	3.8 (1.0)	3.3 (0.7)
NMV (mm/min)	9.0 (2.3)	—	8.2 (2.0)	6.6 (2.1)

*Subjects were 1 man, 5 women, mean age 36 yr, range 23-52 yr; mean values (SE) for parameters.

†NAR immediately after pollen inhalation elevated compared with baseline ($p < 0.01$) and 2 hr later elevated compared with air sham ($p < 0.05$).

‡Subjects were 2 men, 4 women, mean age 34 yr, range 30-41 yr; mean values (SE) for parameters; FEV₁ not performed.

repeated in seven subjects without their inhaling the pollen (Table I).

Inhalation of nebulized ragweed extract

Baseline measurements of NAR, NMV, and FEV₁ were obtained in 10 subjects. A dosimeter (Model D-2, Johns Hopkins University, Baltimore, Md.) was connected to a mask (Bird Corp., Model No. 4243) so that the subject could inhale the nebulized solution (mass median aerodynamic diameter of 3.86 μ m, geometric SD 2.35 μ m) through both nostrils. The provocation was begun by having the subject inhale a phosphate-buffered diluent over 10 consecutive breaths from the functional residual capacity (FRC) level to total lung capacity (TLC). NAR and FEV₁ were measured 5 and 10 min after the inhalations. The patient then inhaled aerosol from a 1:500,000 dilution (1:10 w/v, 144,000 protein nitrogen units [PNU]/ml stock solution) of short ragweed pollen extract. Further dilutions, e.g., 1:100,000, 1:50,000, 1:10,000, and 1:5000, were repeated until NAR reached approximately four times the baseline. NMV in the same nostril as the baseline was again measured and NAR, NMV, and FEV₁ were repeated 1 and 2 hr later.

In another group of eight patients with allergic rhinitis, NAR, NMV and FEV₁ were measured immediately and 1 and 2 hr after the subject inhaled 10 breaths of phosphate-buffered saline aerosol from the dosimeter (Table I).

Repeated ragweed inhalation of nebulized ragweed extract over 3 days

Baseline measurements of NAR, NMV, and FEV₁ were obtained. The subject donned the mask used for NAR which was modified with one-way valves such that ragweed antigen extract aerosol could be continuously administered over a 30-min period (Table I). NAR was monitored at 7-min intervals after exposure began. The dilution of ragweed extract chosen was that which had previously evoked a fourfold increase in NAR. NAR, NMV, and FEV₁ were measured immediately and 1 hr after termination of the expo-

sure. The study was performed in the same subject on each of 3 consecutive days.

Intranasal instillations of solutions of ragweed extract, phosphate-buffered saline, and other antigen extracts

Measurements of NAR and NMV were obtained as baselines in 10 patients (Table I). Three drops of a solution of ragweed antigen extract, the concentration of which had previously evoked a fourfold increase in NAR by aerosolization, or a 1:500,000 dilution if previous aerosol data were not available, were instilled intranasally in both nostrils while the patient lay supine with the neck superextended. NAR was measured 10 min later and the concentrations were increased until values of NAR had increased approximately fourfold over baseline, at which time the instillation was terminated. Both NMV and NAR were measured 0.5 and 1.5 hr after the final intranasal instillation of ragweed antigen extract.

To serve as a control, measurements of NAR and NMV were obtained in nine patients immediately and 0.5 and 1.5 hr after instillation of three drops of phosphate-buffered saline in each nostril (Table I). As an additional control to exclude a nonspecific effect of foreign protein on nasal function, measurements of NAR and NMV in eight patients were obtained immediately and 0.5 and 1.5 hr after instillation of other plant pollen extract solutions into each nostril (Table I). Pollen extracts were chosen from plants grown in the Western part of the United States and included mountain cedar, Russian thistle, cedar elm, and cinnamon sagebrush (Table I). Intradermal injections of 0.02 ml of 1:10,000 (100 PNU/ml) or 1:1,000 (1,000 PNU/ml) were placed on the volar surface of the forearm and the diameter of the wheal measured immediately and 15 min after injection. The allergen that produced the smallest cutaneous reaction was selected for instillation into the nose as in the ragweed extract instillation. The dilution of antigen was the same as the dilution of ragweed extract which had produced a fourfold elevation of NAR.

TABLE III. Effects of nasal inhalation of nebulized phosphate-buffered saline (10 breaths) and ragweed antigen extract for fourfold rise of baseline NAR

	Baseline	After exposure		
		Immediate	1 hr	2 hr
<i>Nebulized phosphate-buffered saline*</i>				
NAR (cm H ₂ O/L/sec)	5.0 (1.4)	4.2 (0.7)	4.8 (1.0)	5.1 (1.4)
NMV (mm/min)	4.7 (0.2)	4.8 (0.2)	4.6 (0.3)	4.7 (0.2)
FEV ₁ (LBTPS)	3.9 (0.4)	3.8 (0.4)	3.8 (0.3)	3.8 (0.3)
<i>Nebulized ragweed antigen extract† to 4-fold NAR</i>				
NAR (cm H ₂ O/L/sec)	3.6 (0.5)	14.2 (1.7)‡	6.6 (1.3)	5.5 (1.0)
NMV (mm/min)	7.6 (1.3)	7.0 (0.7)	10.7 (1.8)	8.6 (1.4)
FEV ₁ (LBTPS)	3.6 (0.3)	3.3 (0.3)	3.5 (0.2)	3.5 (0.2)

*Subjects were 4 men, 4 women, mean age 29 yr, range 21-41 yr, mean values (SE) for parameters.

†Subjects were 7 men, 3 women, mean age 28 yr, range 22-34 yr, mean values (SE) for parameters.

‡Significant elevation compared with baseline and saline values ($p < 0.05$).

Statistical analysis

Data were treated with a paired *t* test and analysis of variance (ANOVA) with Newman-Keuls test where appropriate.¹⁰

RESULTS

Nasal inhalation of ragweed pollen grains

The quantity of pollen grains required to produce a two- to fourfold elevation of NAR varied from 50 to 3,000 grains and was not dependent upon the cutaneous reaction, which was uniformly grade 4 (Table I). None of the subjects completing the study developed a fall in FEV₁ greater than 20%, wheezing, or cough. The rise in NAR reached a maximum value of 281% over baseline and was still elevated at the 2-hr observation point to 210% above baseline ($p < 0.05$) (Table II). There was a slight fall in NMV ($p < 0.05$) compared with baseline 2 hr after pollen grain administration but this was not statistically different from the air sham over the same time period (Table II).

Nasal inhalation of nebulized ragweed extract

The dilutions of ragweed extract which produced an approximately fourfold rise of baseline NAR varied from 1:500 to 1:500,000 (Table I). This fourfold increase in NAR was reached in seven patients; in three patients (Nos. 10, 11, and 16; Table I), a 20% fall in FEV₁ occurred before the desired increase in NAR. These three patients had a history of bronchial asthma but none had asthmatic symptoms on the day of and the day preceding the nasal provocation study. The mean coefficient of variation of five baseline estimations of NAR for the seven subjects was 8%, range 1% to 18%. The mean baseline value of NAR

for the provocation studies with nebulized ragweed extract was 3.6 cm H₂O/L/sec, SE 0.5, and immediately after administration of phosphate-buffered diluent it rose to 4.6 cm H₂O/L/sec, SE 0.8 ($p = \text{NS}$). It reached a peak value of 14.2 cm H₂O/L/sec, SE 1.7, after serial dilutions of the nebulized ragweed extract were administered. The mean coefficient of variation of five estimations of NAR at the peak response was 9%, range 5% to 19%. When serial values of NAR obtained after exposure to nebulized ragweed extract were compared with nebulized phosphate-buffered saline, the only statistical difference was this 394% peak rise in NAR (Table III). There was no significant alteration of NMV produced either by saline or ragweed extract over the 2-hr observation period.

Repeated nasal inhalation of nebulized ragweed antigen extract on three consecutive days

The dilution of ragweed antigen extract chosen for the 30-min exposure was equal to that which had previously been found to elevate NAR fourfold after short-term inhalation. At the end of the first day of exposure, NAR rose to 224% over baseline ($p < 0.05$) and returned toward the baseline value 1 hr later (Table IV). On the subsequent two days, there was no significant increase of NAR with exposure to ragweed antigen since less than half the subjects responded with a significant increase of NAR (Table IV). Furthermore, there were no statistically significant differences among the baseline values of NAR over the three-day period. No significant alterations of baseline values of NMV occurred on the successive days or following the short-term daily challenge days.

TABLE IV. Effects of 30 min nasal inhalation of nebulized ragweed antigen extract over 3 consecutive days*

	Baseline	After exposure	
		Immediate	1 hr
<i>Day 1</i>			
NAR (cm H ₂ O/L/sec)	2.3 (0.6)	6.8 (1.3)†	3.8 (0.6)
Number of patients with significant increase in NAR		5 of 7	4 of 7
NMV (mm/min)	8.0 (1.2)	4.7 (1.5)	6.4 (1.2)
FEV ₁ (LBTPS)	3.5 (0.3)	3.3 (0.3)	3.4 (0.3)
<i>Day 2</i>			
NAR (cm H ₂ O/L/sec)	3.3 (1.1)	6.6 (4.0)	4.2 (0.9)
Number of patients with significant increase in NAR		3 of 7	4 of 7
NMV (mm/min)	7.4 (1.7)	6.7 (1.1)	4.7 (1.4)
FEV ₁ (LBTPS)	3.4 (0.3)	3.3 (0.3)	3.5 (0.3)
<i>Day 3</i>			
NAR (cm H ₂ O/L/sec)	3.3 (0.7)	7.8 (3.4)	8.4 (4.9)
Number of patients with significant increase in NAR		3 of 7	3 of 7
NMV (mm/min)	7.3 (1.7)	6.8 (1.9)	5.2 (1.8)
FEV ₁ (LBTPS)	3.6 (0.3)	3.4 (0.3)	3.4 (0.2)

*Subjects were 3 men, 4 women, mean age 28 yr, range 21-36 yr, mean values (SE) for parameters.

†Significant elevation compared with baseline ($p < 0.05$).**TABLE V.** Effects of intranasal instillation of phosphate-buffered saline, ragweed antigen extract, and various antigen extracts

	Baseline	After exposure	
		0.5 hr	1 hr
<i>Phosphate-buffered saline*</i>			
NAR (cm H ₂ O/L/sec)	5.0 (1.1)	4.8 (1.0)	6.2 (1.6)
NMV (mm/min)	5.8 (1.1)	7.7 (1.4)	6.8 (1.6)
<i>Ragweed antigen extract†</i>			
NAR (cm H ₂ O/L/sec)	4.7 (0.4)	26.1 (4.5)‡	8.7 (1.3)§
NMV (mm/min)	7.7 (1.2)	4.0 (1.1)	4.4 (1.5)¶
<i>Various antigen extracts#</i>			
NAR (cm H ₂ O/L/sec)	6.4 (1.1)	7.6 (1.5)	6.1 (1.0)
NMV (mm/min)	6.2 (0.5)	5.9 (1.1)	7.6 (0.8)

*Subjects were 2 men, 7 women, mean age 27 yr, range 21-36 yr; mean values (SE) for parameters.

†Subjects were 3 men, 7 women, mean age 28 yr, range 21-36 yr, mean values (SE) for parameters.

‡ $p < 0.001$ (compared with saline and various antigen extracts).§ $p < 0.01$ (compared with various antigen extracts).¶ $p < 0.01$ (compared with saline and various antigen extracts).|| $p < 0.05$ (compared with saline).

#Subjects were 3 men, 5 women, mean age 28 yr, range 21-36 yr; mean values (SE) for parameters.

Intranasal instillation

There were no significant changes in NAR and NMV after intranasal instillation of phosphate-buffered saline and antigen extracts to which there was minimal cutaneous reaction. Intranasal instillation of ragweed extract to the point where NAR increased fourfold (556% of baseline; Table V) was accompanied by a 48% fall in NMV the fall in NMV was

statistically significant ($p < 0.05$) when compared with intranasal instillation of saline but not the various extracts. The 1.5-hr follow-up revealed a 43% fall in NMV which occurred when NAR was 114% above baseline. This decrease in NMV was significant ($p < 0.01$) when compared with the saline and various antigen extracts. This level of statistical significance was also observed when the mean of the

NMV values 0.5 and 1.5 hr after intranasal instillation of ragweed antigen extract was compared with saline and various antigen extracts. Individual values of NMV after intranasal instillation of ragweed antigen extract solution are depicted in Fig. 1.

DISCUSSION

The present study is in agreement with other investigations which indicate that a rise in NAR can be consistently demonstrated in patients with allergic rhinitis after nasal inhalation of pollen grains or specific nebulized antigen extracts.^{2, 3} Chevance^{4, 7} reported that deposition of pollen grains induces ciliary paralysis within 1 to 10 min after application to sensitized mucous membranes of rabbits and guinea pigs *in vitro* and destruction of ciliated epithelium 10 to 30 min later. Contrary to these observations, Wihl and Mygind,⁵ in a short-term challenge of patients with allergic rhinitis with pollen grains, failed to find depression of nasal mucociliary transport or histologic destruction of ciliated epithelium. However, this study had deficiencies in experimental design. The pollen grains were deposited by micropipette on the lower edge of the inferior turbinate 1 cm posterior to the front edge and this deposition pattern did not mimic the clinical state in which more even deposition on the nasal mucosa might be expected. Wihl and Mygind⁵ assessed nasal mucociliary transport by the saccharin-dye test. The saccharin-dye particle was placed on a different area than the pollen grains, the upper part of the inferior turbinate 4 cm from the tip of the nose, thereby testing remote rather than local effects of pollen on nasal mucus transport. The saccharin-dye test consisted of asking the patient to swallow once a minute after placing a saccharin-dye particle on the nasal mucosa. The end point of this test was the time when the patient sensed a sweet taste and was verified by observing the dye color in the back of the throat. The mean saccharin time was 11 min both before and after challenge 30 min later. This value would correspond to a relatively slow baseline NMV and therefore it might not be possible to detect further slowing because of the insensitivity of the method. Yergin et al.⁸ pointed out that secondary end points occur with repeated saccharin tests such that repetition is subject to artifacts if carried out more than once every 1 to 2 hr.

To promote more even deposition of ragweed pollen grains than Wihl and Mygind,⁵ who deposited pollen locally by means of a pipette, the inhalation technique of Connell³ was used in our initial investigation. The number of inhaled pollen grains to produce a two- to fourfold elevation of NAR was in the

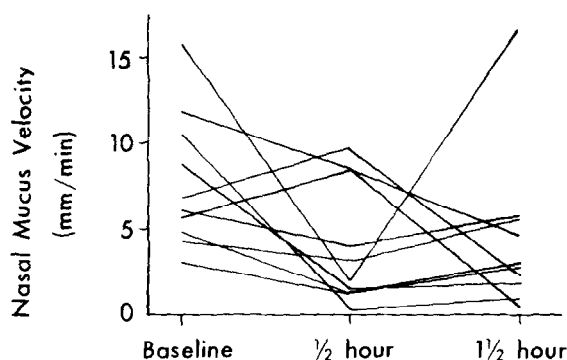


FIG. 1. Nasal mucus velocity in patients with allergic rhinitis after intranasal instillation of ragweed extract. In several instances the velocity of the Teflon disks almost ceased after administration of antigen.

same range as Connell¹¹ had used to provoke a symptomatic response. In agreement with his work, a sham maneuver with air did not produce a rise in NAR in our patients with allergic rhinitis. Furthermore, NMV was not altered by the sham maneuver nor by inhalation of pollen. These data suggested that elevation of NAR might be a more sensitive indicator of antigen-antibody reaction than mucus transport. But the possibility of local slowing of mucus transport accompanied by generalized increase of NAR still remained unanswered by this experimental design in which ragweed pollen grains were not distributed evenly over the nasal mucosa.

To ensure a more even deposition of antigen onto the nasal mucosa and a more intense nasoconstrictive response, the patients inhaled nebulized ragweed extract solution to achieve an approximately fourfold increase in NAR over baseline. There was no correlation between the degree of cutaneous hypersensitivity and dilution of ragweed antigen to produce this rise in NAR confirming the observations of Taylor and Shivalkar.² The phosphate-buffered saline diluent was nebulized as a control since McLean et al.¹² had reported previously that NAR increased an average of 22.5% both in atopic and nonatopic persons after administration. Most of the peak rise reported by these authors took place within the first 5 to 10 min after nebulization. We also found a mean rise of 26% in NAR immediately after nebulization of phosphate-buffered diluent but this rise was not statistically significant possibly because of a smaller number of subjects employed in our study. Further, there was no significant alteration of NMV in these subjects.

We were concerned that alteration of nasal mucus transport might have been missed because of uneven deposition of nebulized antigen over the short period

of nasal inhalation. The same dilution of antigen known to produce a fourfold elevation of NAR in 10 breaths was nebulized over 30 min. The challenges were performed on 3 successful days to promote a priming effect should this factor be necessary to effect a change in NMV. Connell¹³ defined the "priming effect" as an increase in reactivity as measured by nasal patency occurring only after repeated challenges had been carried out in some patients with allergic rhinitis. He subsequently demonstrated that the "priming effect" was nonspecific, i.e., after priming with a specific antigen, nonspecific antigen in small dosages produced symptoms of severe rhinitis. This effect could not be demonstrated in the present experiments possibly because the "priming effect" might take much longer to develop than the 3 days we utilized.

As a final method to provoke elevation of NAR and elicit the most even deposition of antigen, a solution of ragweed antigen extract was instilled directly into each nostril to attain the quadrupling of NAR. McLean et al.¹² reported rises in NAR in atopic subjects ranging from 22% to 104% over baseline in a 15-min period following pipetting 0.1 ml of phosphate-buffered saline into each nostril, an effect blocked by prior nasal inhalation of aerosolized atropine sulfate. This early parasympathetic stimulation by saline (or other instilled solutions) presumably had dissipated in the present experiments since NAR and NMV were similar to baseline values 30 min after intranasal instillation of the buffered saline solution. NMV fell 0.5 hr after provocation with antigen when there was intense nasal airway constriction and this fall continued to the next observation point 1.5 hr later when NAR had returned toward baseline values. This time course was similar to the tracheal mucus velocity alterations observed in patients with bronchial asthma after inhalation of aerosolized ragweed antigen extract.¹⁴

The present experiments demonstrate that patients with allergic rhinitis develop nasal airway constriction in the laboratory after introduction of specific antigen in several ways, i.e., pollen grains, nebulized antigen extract, and intranasal instillation of antigen extract solution. In the laboratory, the nasal airway constrictor response to antigen is a more sensitive indicator of the allergic reaction than alteration of the mucus transport system since inhalation of pollen grains and nebulized antigen produce an elevation of NAR without change in NMV. But when antigen extract is instilled directly into the nose to effect a comparable degree of nasal airway constriction, there is a significant fall in NMV both when nasal airway constrict-

tion is intense as well as during its subsidence. This effect appears to be a specific one as evidenced by the lack of effect of instillation of antigen in which there is little cutaneous reaction to intradermal injection.

One can only speculate at the explanation of the apparently discordant results between nasal inhalation of pollen grains and nebulized antigen on the one hand and intranasal instillation of ragweed antigen extract solution on the other. Nasal airway constriction might be primarily mediated through a neural reflex mechanism such that a small number of pollen grains or nebulized antigen depositing on the nasal mucosa might provoke widespread nasal airway constriction. The depression of mucus transport observed with direct instillation of antigen extract solution might be related to a heavier dosing of the nasal mucosa. Alternatively, local depression of mucus transport due to release of mediator substances might be missed because the disks were not deposited over these regions. Theoretically, if large numbers of disks (e.g., 50 to 100) were employed as markers of mucus transport, potential local effects from pollen grains and nebulized antigen deposition would be measured but this method is impractical to use in such a manner.

Persistence of depression of nasal mucus velocity after intranasal instillation of ragweed extract 1.5 hr after administration when the nasal constrictor response has dissipated is analogous to depression of tracheal mucus velocity observed in bronchial asthma after challenge with nebulized antigen.¹⁴ Investigations in dogs suggest that slow-reacting substance of anaphylaxis (SRS-A) might be the responsible mediator for this phenomenon.¹⁵ In this regard, it is noteworthy that SRS-A has been extracted from nasal polyps of patients with allergic rhinitis after challenge with ragweed antigen.¹⁶

REFERENCES

1. Connell JT: Allergic rhinitis: Human experimental model. *NY State J Med* 70:1751, 1970.
2. Taylor G, Shivalkar PR: Changes in nasal airways resistance on antigenic challenge in allergic rhinitis. *Clin Allergy* 1:63, 1971.
3. Connell JT: Quantitative intranasal pollen challenges. I. Apparatus design and technique. *J Allergy* 39:358, 1967.
4. Chevance LG: Experimental pollinosis: An electron microscopic study. *Acta Otolaryngol* 72:121, 1971.
5. Wihl JA, Mygind N: Studies on the antigen-challenged nasal mucosa. *Acta Otolaryngol* 84:281, 1977.
6. Tos M, Mogensen C: Nasal glands in nasal allergy. *Acta Otolaryngol* 83:498, 1977.
7. Chevance LG: The first stages of nasal anaphylactic shock. *Acta Otolaryngol* 47:480, 1957.
8. Yergin BM, Sakethkoo K, Michaelson ED, et al: A roentgenographic method for measuring nasal mucous velocity. *J Appl Physiol* 44:964, 1978.

9. Saketkhoo K, Januszkiewicz A, Sackner MA: Effects of drinking hot water, cold water and chicken soup on nasal mucus velocity and nasal airflow resistance. *Chest* **74**:408, 1978.
10. Mendenhall W, Mc Clave JT, Ramey M: *Statistics for psychology*, ed. 2. North Scituate, Mass., 1977, Duxbury Press, p. 246, 306-322.
11. Connell JT: Quantitative intranasal pollen challenge. II. Effect of daily pollen challenge, environmental pollen exposure, and placebo challenge on the nasal mucosa. *J ALLERGY* **41**:123, 1968.
12. McLean JA, Mathews KP, Ciarkowski AA, et al: The effects of topical saline and isoproterenol on nasal airway resistance. *J ALLERGY CLIN IMMUNOL* **58**:563, 1976.
13. Connell JT: Quantitative intranasal pollen challenges. III. The priming effect in allergic rhinitis. *J ALLERGY* **43**:33, 1969.
14. Mezey RJ, Cohn MA, Fernandez RJ, et al: Mucociliary transport in allergic patients with antigen-induced bronchospasm. *Am Rev Respir Dis* **118**:677, 1978.
15. Wanner A, Zarzecki S, Hirsch J, et al: Tracheal mucous transport in experimental canine asthma. *J Appl Physiol* **39**:950, 1975.
16. Kaliner M, Wasserman SI, Austen KF: Immunologic release of chemical mediators from human nasal polyps. *N Engl J Med* **289**:277, 1973.

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