

Unilateral nasal allergic reactions increase bilateral sinus eosinophil infiltration

Fuad M. Baroody, Marcella deTineo and Robert M. Naclerio

J Appl Physiol 115:1262-1267, 2013. First published 22 August 2013;

doi:10.1152/jappphysiol.00547.2013

You might find this additional info useful...

This article cites 30 articles, 8 of which can be accessed free at:

</content/115/9/1262.full.html#ref-list-1>

Updated information and services including high resolution figures, can be found at:

</content/115/9/1262.full.html>

Additional material and information about *Journal of Applied Physiology* can be found at:

<http://www.the-aps.org/publications/jappl>

This information is current as of November 9, 2014.

Unilateral nasal allergic reactions increase bilateral sinus eosinophil infiltration

Fuad M. Baroody, Marcella deTineo, and Robert M. Naclerio

Section of Otolaryngology-Head and Neck Surgery, The University of Chicago Medical Center and The Pritzker School of Medicine, The University of Chicago, Illinois

Submitted 3 May 2013; accepted in final form 20 August 2013

Baroody FM, deTineo M, Naclerio RM. Unilateral nasal allergic reactions increase bilateral sinus eosinophil infiltration. *J Appl Physiol* 115: 1262–1267, 2013. First published August 22, 2013; doi:10.1152/jappphysiol.00547.2013.—We have previously shown that unilateral nasal challenge with antigen causes an increase in the number of eosinophils in the ipsilateral maxillary sinus. Here we aimed to determine whether there was an eosinophil response in the contralateral maxillary sinus after unilateral nasal challenge with antigen. Twenty subjects with a history of seasonal allergic rhinitis and a positive nasal challenge to ragweed or grass allergens were studied outside of their allergy season. Catheters were placed in both maxillary sinuses and the subjects were challenged with antigen via the left nostril. The subjects recorded nasal symptoms before and after each allergen challenge and hourly for 8 h afterward. We performed nasal lavages of the nose and sinuses at the same time as symptoms were recorded. The lavages were analyzed for the number of eosinophils and levels of albumin. Subjects showed a symptomatic response to challenge accompanied by an influx of eosinophils into the nose and increased vascular permeability. The number of eosinophils increased in both maxillary sinuses. The total change from diluent in eosinophils during the late phase response was higher in the ipsilateral maxillary sinus (median = 8,505; range = 0–100,360) compared with the contralateral sinus (median = 1,596; range = –13,527–93,373; $P = 0.03$). We conclude that eosinophils increase in both maxillary sinuses after unilateral nasal challenge. We speculate that a central neurologic reflex initiated in the nose by the nasal challenge contributes to the bilateral eosinophil response in the maxillary sinuses. We further speculate that, since there are more eosinophils in the ipsilateral compared with the contralateral maxillary sinus, there is also an axonal reflex into the ipsilateral maxillary sinus that contributed to the eosinophil response.

maxillary sinus; nasal allergen challenge; eosinophils

THE NOSE IS RICHLY INNERVATED by sensory fibers from the second division of the trigeminal nerve and sympathetic and parasympathetic fibers that pass through the sphenopalatine ganglion. Whereas the sympathetic fibers merely pass through the ganglion, the parasympathetic fibers synapse sending efferent fibers to the nasal cavity, the paranasal sinuses, and the eye (2).

Numerous neural reflexes originating inside and outside the nose affect nasal pathophysiology. The nasonasal reflex results from stimulation of one nasal cavity with a stimulus such as allergen or histamine. It produces a bilateral secretory response with the contralateral response being blocked by topical atropine administration (8, 3). Stimulation of the nose with allergen also induces bilateral ocular symptoms, the nasal ocular reflex (4). The nasal cycle, the process by which airway resistance of the two nasal cavities fluctuates in a cyclic manner, also

involves neural mechanisms (15). Gustatory rhinitis, which involves an increase in rhinorrhea after food ingestion, is an example of a neural reflex that affects the nasal cavity and originates in the mouth (27). Data support neural interactions between the nose and the lung and vice versa (9–11).

In previous studies, we demonstrated that nasal challenge with allergen led to an increase in the number of eosinophils infiltrating the ipsilateral maxillary sinus cavity (6, 7). The numbers of eosinophils exceeded the number obtained after control challenge of the nasal cavity with lactated Ringer (LR) solution (6). Supporting a neural reflex, antigen could not be detected in the sinus after nasal challenge. Furthermore, the percentage of eosinophils in the sinus lavage was greater than that in the peripheral blood, suggesting that bleeding into the sinus cavity during sampling did not explain the eosinophil influx. To further explore the interaction between the nose and sinuses when the nose is stimulated by allergen, we compared the eosinophil response in the ipsilateral and contralateral maxillary sinuses after unilateral antigen challenge.

METHODS

Study design. We performed a single center study in 20 healthy subjects who had a history of grass and/or ragweed allergy symptoms. Subjects came to the Nasal Physiology Laboratory for screening, where they completed an allergy questionnaire and underwent skin prick testing for confirmation of a positive grass or ragweed allergy. Depending on skin test results, eligible subjects then underwent a screening nasal challenge with either grass or ragweed allergen outside their allergy season. The study was approved by the Institutional Review Board of The University of Chicago, and all subjects gave written informed consent prior to entry. The study was performed before the requirement for registration with ClinicalTrials.gov and an investigational new drug for the antigen challenge was required.

We selected volunteers whose ages ranged between 18 and 55 years and who were in good health, other than having a history of allergic rhinitis. Patients with a history of sinusitis were excluded, as well as those with medical illnesses or bleeding diathesis. All subjects were asymptomatic and off all medications for a minimum of 48 h prior to challenge. Steroids were not permitted for 2 weeks prior to the start of the study, whereas immunotherapy was not permitted in the preceding year.

Subjects who passed the screening challenge (i.e., they exhibited a positive nasal response to nasal allergen challenge, as determined by increased sneezing) were allowed to wash out for 2 wk, and 20 responders were asked to return to the Nasal Physiology Laboratory. Subjects had bilateral maxillary sinus catheters placed and then underwent a unilateral nasal challenge with allergen. One subject was replaced after the initial sinus puncture showed purulent secretions prior to nasal allergen challenge.

Study day. We inspected the nose and then decongested (oxymetazoline in spray form, 2 puffs) and anesthetized (4% lidocaine with cotton-soaked pledgets in the inferior meatus) on the right and left

Address for reprint requests and other correspondence: F. Baroody, Section of Otolaryngology-Head and Neck Surgery, 5841 S. Maryland Ave., MC1035, Chicago, IL 60637 (e-mail: fbaroody@surgey.bsd.uchicago.edu).

sides in preparation for the introduction of the sinus catheter, SinoJect (Atos Medical, distributed by Bivona Medical Technologies, Gary, IN). We placed SinoJect on the right side as previously described (5). The sinus catheter was connected to a syringe that was used to lavage the maxillary sinus antrum with four 10-ml aliquots of 37°C LR solution, saving the first lavage for mediators and cell counts. We then inserted the SinoJect on the left side and lavaged the sinus with four 10-ml aliquots of LR solution, saving the first lavage for mediators and cell counts. We also drew blood for complete blood count (CBC) and differential. We waited at least 4 h to allow for clotting around the catheters and then recorded nasal symptoms. The left side of the nose was lavaged with four 10-ml aliquots of LR solution, saving the first and fourth washes as nasal baseline to be sampled for cell counts. We next lavaged both sinuses with four 10-ml aliquots of LR solution, saving the fourth from each side as the left and right sinus baseline samples.

A nasal challenge with diluent or antigen was performed on the left side. First, the diluent for antigen extract (phenol-buffered saline) was delivered from a metered dose inhaler (2 puffs ~0.17ml). Ten minutes later, subjects recorded symptom scores and we lavaged the challenged side of the nose (left) with 5 ml of LR solution and both sinuses with 5 ml each. Two minutes later we challenged the nose (as above) with 0.1 ml of 300 bioequivalent allergy units (BAU) of grass antigen or 0.1 ml of 1:2,000 wt/vol of ragweed antigen. Ten minutes later subjects recorded symptom scores and we lavaged the nose and both sinuses with 5 ml LR solution. We repeated the above challenge with 1,000 and 3,000 BAU of grass extract or 1:200 wt/vol and 1:20 wt/vol of ragweed antigen, respectively. Ten minutes following each antigen dose, 5-ml lavages of nose and both sinuses were performed. One hour after the last antigen challenge, subjects recorded symptom scores and we lavaged the left nostril and both the left and right sinuses with 5 ml LR solution. These steps were repeated every hour for the next 7 h to monitor the late phase response. After the collection at Hour 3, we sprayed each nostril with two puffs of oxymetazoline to decrease nasal congestion and enable washes. After the eighth hour, we drew another tube of blood for CBC and differential and removed the SinoJect catheters from the sinuses.

Symptoms. Nasal symptoms (congestion, rhinorrhea, and itchy nose,) were rated on a scale of 0 (none), 1 (mild), 2 (moderate), and 3 (severe) and recorded by the volunteers. Total nasal symptoms were calculated by adding the individual symptoms. The number of sneezes and symptoms were recorded after each challenge and hourly thereafter during the late phase response.

Nasal and sinus lavages. Subjects had their nasal cavity lavaged (5 ml warm LR solution on each side) as previously described, and the lavages were transferred to plastic tubes and placed on ice until processing (6). Lavages collected from subjects were initially shaken to break up mucus and centrifuged at 5,000 rpm for 15 min, and the supernatant was decanted. The cell pellets were then suspended in 9 ml of distilled water to lyse the red blood cells, and then 1 ml of 10× PBS was added to bring the samples back to an isotonic solution. Total cells were then counted using a hemocytometer. The sample was again centrifuged and the supernatant discarded. The cell pellet was suspended in LR solution and cytospun on slides. Pilot experiments showed that the percentage of recovered cells was not affected by this processing technique. These slides were evaluated for eosinophil numbers by an observer blinded to treatment protocol. Two hundred cells were evaluated on slides (when possible), and we enumerated eosinophils, neutrophils, and mononuclear cells. We thus obtained a percentage of eosinophils in the lavage samples. Because the total number of cells was known, the total number of eosinophils was calculated. Low numbers of cells in the cytospun slides occurred because of low total cell counts or technical difficulties during the process of preparation of the smears. From our previous experience using this technique, a large sampling error can occur when the total number of cells evaluated in the smear is <50. Because of the paucity of cells in some of the specimens, we made some assumptions relating

to eosinophil number. The lowest total number of eosinophils obtained from slides with adequate cells counted and an evaluable total cell count was 55. This number was assigned to specimens in the following situations: 1) when the number of cells evaluated in the smear was <50 and 2) when the number of eosinophils and thus percentage of eosinophils obtained from examining the differential count was zero. Exceptions to this assumption were patients who had a large number or total cells (>60,000) and where the resultant smears had fewer than 50 cells to evaluate. These samples were considered technically inadequate and are not included in data analysis.

Albumin. Human serum albumin was measured with an ELISA sensitive to 1 ng/ml albumin. Any sample that had concentrations lower than 1 ng/ml was assigned a value of 0.5 ng/ml.

Statistics. Because the data were not distributed normally except for the eosinophil percentage in peripheral blood, nonparametric statistics (Friedman ANOVA and Wilcoxon signed-rank test) were used for evaluating the responses. The data are presented as individual points connected by lines to indicate the paired nature of the data, and the group data are depicted as medians. When numerical data are reported, the median (range) is shown. To compare the two sides, we calculated the change from the diluent response induced by antigen by subtracting the response after the diluent from that obtained after antigen challenge. Paired *t*-test was used to compare the percentage of eosinophils in the systemic circulation to that in the nasal and sinus cavities. $P < 0.05$ was considered significant. All statistical tests were performed using a Macintosh computer (Apple Computer, Cupertino, CA) and Statview II statistical software (Abacus concepts).

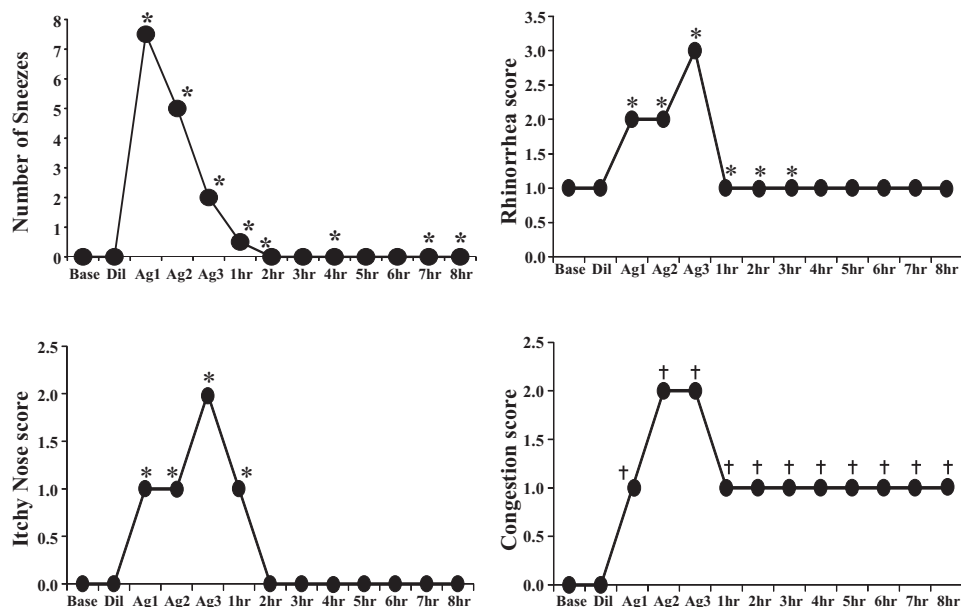
RESULTS

Subjects. Twenty subjects completed the protocol. The median age of the subjects was 25.5 years with a range from 19 to 41 years. There were 15 males and 5 females, and 14 Caucasians, 4 African Americans, and 2 Hispanics. Twelve subjects underwent challenge with grass and eight with ragweed allergen. All challenges were performed outside of the allergy season. There were no adverse events reported by the subjects either during or after the challenges and sinus punctures.

Sneezes and nasal symptoms. The response to challenge showed a typical increase in sneezes (ANOVA: $P < 0.001$) and individual nasal symptoms of rhinorrhea, congestion, and itchy nose (ANOVA: $P < 0.001$ for all) after allergen challenge. Sneezing and nasal itching responses were most prominent during the early phase response, whereas rhinorrhea and congestion were present during both early and late phase responses (Fig. 1). When total nasal symptoms were evaluated, there was also a typical increase after allergen challenge (ANOVA: $P < 0.001$) during the early and late phase response after allergen challenge (Fig. 2).

Eosinophil influx. Eosinophils in nasal lavage were few in number before challenge and increased after antigen stimulation, reaching a peak 8 h later (Fig. 3). There was an overall significant increase in nasal eosinophils (ANOVA: $P < 0.001$), with a significant increase in the number compared with the diluent challenge at the following time points: third allergen challenge, and Hours 2–8 after challenge ($P \leq 0.04$). The lavages of the sinuses showed a significant increase in eosinophil numbers in both the sinus ipsilateral (ANOVA: $P < 0.01$) and that contralateral (ANOVA: $P < 0.01$) to the challenged nostril, with counts being about 10-fold less than those obtained from the nasal cavity (Fig. 4). When comparing the counts to those obtained after the diluent challenge, the ipsilateral sinus had significantly higher numbers of eosinophils after the third allergen challenge and Hours 2 and 3 after

Fig. 1. Sneezes and symptoms of rhinorrhea, congestion, and itchy nose after nasal allergen challenge. The x-axis depicts the challenge protocol: Base, baseline; Dil, diluent for the allergen extract; Ag, ragweed or grass allergen extracts at increasing concentrations; hr, hour after the last allergen challenge. Median responses for 20 subjects are depicted. There was a significant increase in sneezes and symptom scores for rhinorrhea, congestion, and itchy nose after allergen compared with diluent during the early and late phase responses. * $P \leq 0.05$ and $^{\dagger}P < 0.01$ vs. diluent challenge.



challenge ($P \leq 0.05$). For the contralateral sinus, the eosinophils were higher in number than those after diluent after the third allergen challenge and Hours 3, 4, and 5 after challenge ($P \leq 0.05$; Fig. 4). To compare the eosinophil influx between the two sinus cavities, we calculated the total change from diluent during the late phase response (Hours 1–8 postchallenge) by subtracting the diluent response from each of the hourly responses and summing the resultant values. The total change from diluent in eosinophils during the late phase response was higher in the ipsilateral maxillary sinus (median = 8,505; range = 0–100,360) compared with the contralateral sinus (median = 1,596; range = –13,527–93,373), $P < 0.05$.

We examined the percentage of eosinophils in the peripheral blood before and after challenge and compared it to the percentages in the nose and both sinus cavities. There was no significant change in the percentage of eosinophils in the peripheral circulation after allergen challenge. We used the postchallenge circulating eosinophil percentage to compare to

the nasal and sinus lavages. Since the blood was drawn at the 8-h time point after challenge, we compared the percentage of eosinophils in the blood at that time with the percentages obtained in the corresponding 8-h time point lavages of the nose and both sinuses. The percentage of eosinophils in the blood after challenge was 2.4 ± 0.4 , in the nose was 31.9 ± 7.4 , in the ipsilateral sinus was 5.9 ± 2.1 , and in the contralateral sinus was 3.8 ± 1.7 . Although all cavities had a higher eosinophil count than the circulating levels, only the nasal eosinophils were significantly higher than the circulating eosinophils at the 8-h time point ($P = 0.001$). We also compared the postchallenge blood eosinophil percentage to the highest percentage of eosinophils obtained in the nose and sinuses during the late phase response. The means $\pm$ SE values were as follows: blood = 2.4 ± 0.4 , nose = 48.9 ± 8.4 , ipsilateral sinus = 33.8 ± 8.1 , and contralateral sinus = 20.0 ± 6.2 . The values were significantly higher than the blood value for all three cavities ($P < 0.05$).

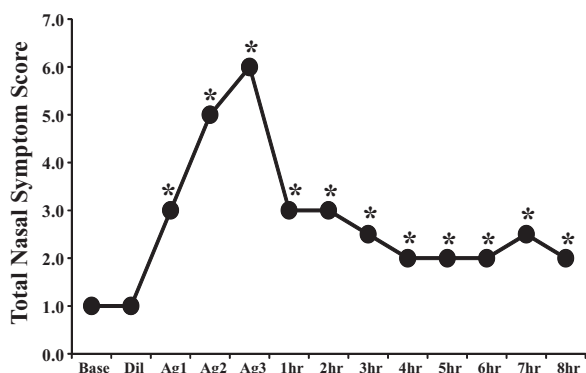


Fig. 2. Total nasal symptoms (rhinorrhea, congestion, and itchy nose) after nasal allergen challenge. The x-axis depicts the challenge protocol: Base, baseline; Dil, diluent for the allergen extract; Ag, ragweed or grass allergen extracts at increasing concentrations; hr, hour after the last allergen challenge. Median responses for 20 subjects are depicted. There was a significant increase in total nasal symptoms after allergen compared with diluent during both the early and late phase response. * $P \leq 0.05$ vs. diluent challenge.

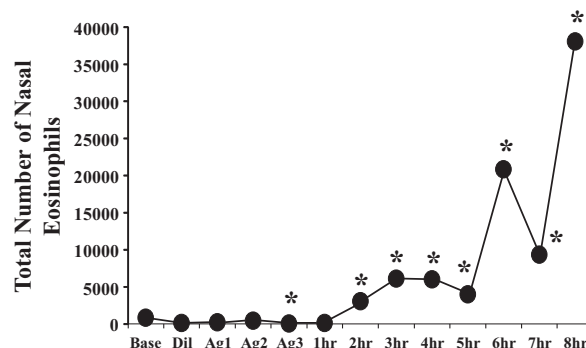


Fig. 3. Eosinophils in nasal secretions after nasal allergen challenge. The x-axis depicts the challenge protocol: Base, baseline; Dil, diluent for the allergen extract; Ag, ragweed or grass allergen extracts at increasing concentrations; hr, hour after the last allergen challenge. The y-axis depicts the total number of eosinophils recovered in each nasal lavage. Median responses for 20 subjects are depicted. There was a significant increase in the number of eosinophils after allergen compared with diluent after the third allergen challenge and Hours 2–8 of the late phase response. * $P \leq 0.05$ vs. diluent challenge.

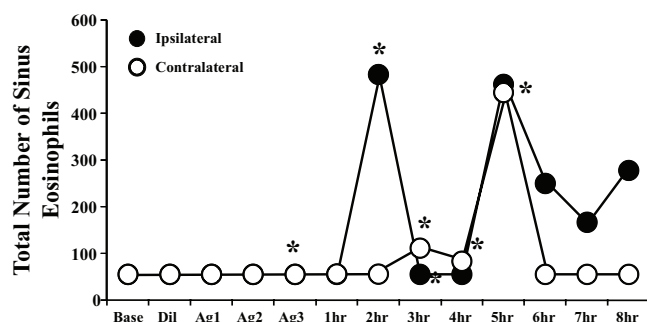


Fig. 4. Eosinophils in ipsilateral and contralateral maxillary sinus secretions after nasal allergen challenge. The x-axis depicts the challenge protocol: Base, baseline; Dil, diluent for the allergen extract; Ag, ragweed or grass allergen extracts at increasing concentrations; hr, hour after the last allergen challenge. The y-axis depicts the total number of eosinophils recovered in each nasal lavage. Median responses for 20 subjects are depicted. There was a significant increase in the number of eosinophils compared with diluent at different time points after nasal allergen challenge in both the ipsilateral and contralateral sinus cavities. * $P \leq 0.05$ vs. diluent challenge.

Albumin levels. There was a significant increase in nasal lavage albumin levels during both the early and late phase responses (ANOVA: $P < 0.05$; Fig. 5). Similarly, there was a significant increase in the level of albumin in both the ipsilateral and contralateral sinuses (ANOVA: $P < 0.001$ for both), which mostly occurred during the late phase response (Fig. 6). Of note is that the magnitude of the sinus response was around threefold smaller than that of the nasal response. There was no difference in the total change from the diluent response for either the early or the late phase responses between ipsilateral and contralateral sinus albumin levels ($P = 0.2$ and $P = 0.5$, respectively).

DISCUSSION

The number of eosinophils recovered from the ipsilateral maxillary sinus was significantly greater than the contralateral maxillary sinus, supporting the hypothesis that a central reflex initiated in the nose contributes to the bilateral eosinophil infiltration and that an axonal or ganglion reflex adds to the ipsilateral response.

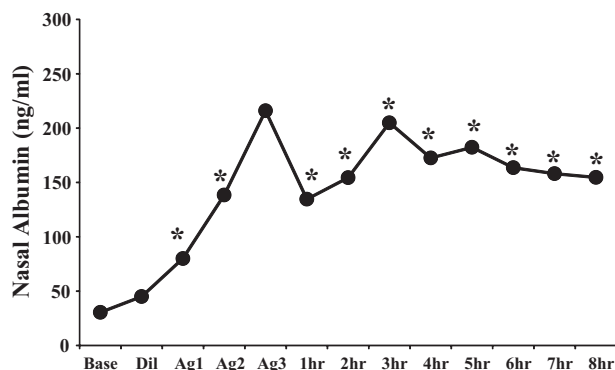


Fig. 5. Levels of albumin in nasal secretions after nasal allergen challenge. The x-axis depicts the challenge protocol: Base, baseline; Dil, diluent for the allergen extract; Ag, ragweed or grass allergen extracts at increasing concentrations; hr, hour after the last allergen challenge. Median responses for 20 subjects are depicted. There was a significant increase in albumin after allergen compared with diluent during both the early and late phase responses. * $P \leq 0.05$ vs. diluent challenge.

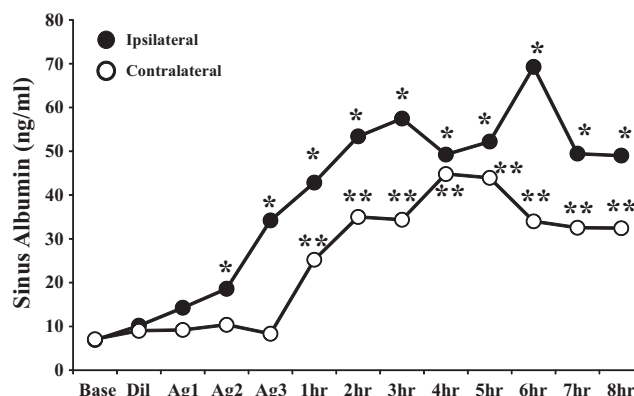


Fig. 6. Levels of albumin in ipsilateral and contralateral maxillary sinus secretions after nasal allergen challenge. The x-axis depicts the challenge protocol: Base, baseline; Dil, diluent for the allergen extract; Ag, ragweed or grass allergen extracts at increasing concentrations; hr, hour after the last allergen challenge. Median responses for 20 subjects are depicted. There was a significant increase in levels of albumin compared with diluent at different time points after nasal allergen challenge in both the ipsilateral and contralateral sinus cavities. * $P \leq 0.05$ and ** $P \leq 0.01$ vs. diluent challenge.

This study clearly shows an increased percentage of eosinophils in both maxillary sinuses compared with the eosinophil percentage in blood. The number of eosinophils in both maxillary sinuses exceeded that after a prior sham challenge (6). The eosinophil percentages in the ipsilateral maxillary sinus are similar to our previously published results, and the baseline levels of eosinophils are similar to previously published control levels (6). This suggests that the previously published observations are reproducible and justified our decision not to include a nonallergic control group to undergo bilateral antral sinus punctures as an additional control.

Pelikan and Pelikan-Felipek (25) challenged the nose with antigen and showed changes in the ipsilateral maxillary sinus on sinus radiographs and on ultrasound evaluations. Others have challenged the nose with antigen and found eosinophils in the lower airway (11). These reports support our findings.

Previously we showed that antigen does not enter the ipsilateral maxillary sinus after nasal challenge with dust mite antigen extract (6). Because of the additional anatomic separation (nasal septum) between the contralateral maxillary sinus and the site of antigen challenge, we do not believe that antigen entering the sinus explains our contralateral or ipsilateral findings.

Neurogenic inflammation involves vasodilatation, glandular activation, hyperreactivity, and cellular infiltration. It mediates these processes through the release of neuropeptides and, like the response to nasal challenge with antigen, there is an early or immediate response followed hours later by cellular inflammation. We believe that the localized allergic reaction in the nose releases cytokines systemically that upregulate peripheral blood eosinophils, which subsequently are recruited to the sinuses. We propose that a second stimulus originating from a neural reflex recruits eosinophils to the sinus in two phases: an immediate response mediated by substance P on the ipsilateral side, and a bilateral late response mediated by acetylcholine. There is literature to support this hypothesis.

Fischer and colleagues (18) showed the presence of neuropeptides in nasal biopsies of patients with persistent perennial allergic rhinitis. Most of these peptides have also been shown

to increase in nasal lavage after nasal provocation with antigen (23). Numao and Agrawal (24) showed that purified substance P and calcitonin gene-related peptide potentiated eosinophil chemotaxis induced by platelet-activating factor. Foster and Cunningham (19) showed substance P induced eosinophil migration in vitro. In guinea pigs, Sagara and colleagues (28) showed that capsaicin infusion led to an early increase in eosinophils in the lower airway, which was attenuated by a substance P antagonist. Tiberio and colleagues (30) also working with guinea pigs showed intracardiac capsaicin led to an eosinophil influx in the alveolar wall that was attenuated by pretreatment with substance P and neurokinin A antagonists. Bailey and Cunningham (1) showed that equine eosinophils adhere to endothelial cells in response to substance P. In vitro, eosinophils harvested from healthy volunteers were primed by low doses of eotaxin to release eosinophil-derived neurotoxin by substance P (16). Substance P also induced eosinophil cationic protein release from human eosinophils in vitro (20). Substance P, applied to the nasal mucosa of asymptomatic dust mite allergic rhinitis patients, led to an increase in eosinophils in five of seven subjects (12). In a nasal challenge model of subjects during a pollen season, substance P nasal challenge 24 h after an antigen challenge markedly increased the number of eosinophils immediately postchallenge (17). Secretoneurin, a neuropeptide released from sensory fibers by capsaicin in vitro, is a chemoattractant for human eosinophils, which is mediated by a phosphodiesterase pathway (13).

Acetylcholine, which is released from parasympathetic nerves on both sides of the nose during the nasosinal reflex, can stimulate bronchial epithelial cells to release eosinophil chemotactic activity (22). Acetylcholine can also stimulate alveolar macrophages to release an eosinophil chemotactic activity, which is blocked by lipoxygenase activity (29). Acetylcholine can also cause vasodilatation.

We speculate that the bilateral sinus response results from systemic activation of eosinophils by the nasal challenge followed by recruitment to both sinuses. This would not explain the ipsilateral response being greater than the contralateral response, and thus we believe that an axonal or ganglion response adds to the recruitment in the ipsilateral sinus. The latter is suggested by Fig. 4 where there is an early increase in eosinophils in the ipsilateral sinus that is not seen on the contralateral side.

In a seasonal study, we have shown that eosinophils increase in the maxillary sinus compared with their numbers out of the season (7). In seasonal exposure, unlike in our challenge model, there would be additional mechanisms involved in the process. Namely, the eosinophils recruited to the site of challenge can associate with nerves to increase their reactivity to a subsequent exposure to pollen (21, 14). Nerve growth factor may contribute to this phenomenon as well as enhancing eosinophil recruitment (26). This probably reduces the amount of antigen needed to initiate the response compared with that used in our challenge model involving a single antigen exposure in asymptomatic individuals outside their season.

In conclusion, our studies support an interaction between the nose and paranasal sinuses in response to nasal allergen challenge and suggest that both central and local reflexes are contributory. This would help explain the close clinical relationship observed between allergic rhinitis and sinusitis.

GRANTS

This work was supported in part by the National Institutes of Health Grant AI-45583 and by the McHugh Otolaryngology Research Fund.

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

AUTHOR CONTRIBUTIONS

Author contributions: F.B. and R.N. conception and design of research; F.B. and M.d. performed experiments; F.B. and R.N. analyzed data; F.B. prepared figures; F.B. and R.N. edited and revised manuscript; R.N. drafted manuscript; R.N. approved final version of manuscript.

REFERENCES

1. Bailey SR, Cunningham FM. Inflammatory mediators induce endothelium-dependent adherence of equine eosinophils to cultured endothelial cells. *J Vet Pharmacol Ther* 24: 209–214, 2001.
2. Baroody FM. The nasal airway. In: *Sleep Disordered Breathing in Children, Respiratory Medicine*, edited by Kheirandish-Gozal L and Gozal D. New York: Springer Science+Business Media, 2012, chapt. 4, p. 37–55. DOI 10.1007/978-1-60761-725-9_4.
3. Baroody FM, Ford S, Lichtenstein LM, Kagey-Sobotka A, Naclerio RM. Physiologic responses and histamine release after nasal antigen challenge: effect of atropine. *Am J Respir Crit Care Med* 149: 1457–1465, 1994.
4. Baroody FM, Foster K, Markaryan A, DeTineo M, Naclerio RM. Nasal ocular reflexes and eye symptoms in patients with allergic rhinitis. *Ann Allergy Asthma Immunol* 100: 194–199, 2008.
5. Baroody FM, Gungor A, deTineo M, Haney L, Blair C, Naclerio RM. A comparison of the response to histamine challenge of the nose and the maxillary sinus: effect of loratadine. *J Appl Physiol* 87: 1038–1047, 1999.
6. Baroody FM, Mucha SM, deTineo M, Naclerio RM. Nasal challenge with allergen leads to maxillary sinus inflammation. *J Allergy Clin Immunol* 121: 1126–1132, 2008.
7. Baroody FM, Mucha SM, deTineo M, Naclerio RM. Evidence of maxillary sinus inflammation in seasonal allergic rhinitis. *Otolaryngol Head Neck Surg* 146: 880–886, 2012.
8. Baroody FM, Wagenmann M, Naclerio RM. A comparison of the secretory response of the nasal mucosa to histamine and methacholine. *J Appl Physiol* 74: 2661–2671, 1993.
9. Braunstahl GJ, Kleinjan A, Overbeek SE, Prins JB, Hoogsteden HC, Fokkens WJ. Segmental bronchial provocation induces nasal inflammation in allergic rhinitis patients. *Am J Respir Crit Care Med* 161: 2051–2057, 2000.
10. Braunstahl GJ, Overbeek SE, Fokkens WJ, Kleinjan A, McEuen AR, Walls AF, Hoogsteden HC, Prins JB. Segmental bronchoprovocation in allergic rhinitis patients affects mast cell and basophil numbers in nasal and bronchial mucosa. *Am J Respir Crit Care Med* 164: 858–865, 2001.
11. Braunstahl GJ, Overbeek SE, Kleinjan A, Prins JB, Hoogsteden HC, Fokkens WJ. Nasal allergen provocation induces adhesion molecule expression and tissue eosinophilia in upper and lower airways. *J Allergy Clin Immunol* 107: 469–476, 2001.
12. Braunstein G, Fajac I, Lacronique J, Frossard N. Clinical and inflammatory responses to exogenous tachykinins in allergic rhinitis. *Am Rev Respir Dis* 144: 630–635, 1991.
13. Dunzendorfer S, Schratzberger P, Reinisch N, Kähler CM, Wiedermann CJ. Secretoneurin, a novel neuropeptide, is a potent chemoattractant for human eosinophils. *Blood* 91: 1527–1532, 1998.
14. Durcan N, Costello RW, McLean WG, Blusztajn J, Madziar B, Fenech AG, Hall IP, Gleich GJ, McGarvey L, Walsh MT. Eosinophil-mediated cholinergic nerve remodeling. *Am J Respir Cell Mol Biol* 34: 775–786, 2006.
15. Eccles R. A role for the nasal cycle in respiratory defence. *Eur Respir J* 9: 371–376, 1996.
16. El-Shazly A, Ishikawa T. Novel co-operation between eotaxin and substance-P in inducing eosinophil-derived neurotoxin release. *Mediators Inflamm* 8: 177–179, 1999.
17. Fajac I, Braunstein G, Ickovic MR, Lacronique J, Frossard N. Selective recruitment of eosinophils by substance P after repeated allergen exposure in allergic rhinitis. *Allergy* 50: 970–975, 1995.
18. Fischer A, Wussow A, Cryer A, Schmeck B, Noga O, Zweng M, Peiser C, Dinh QT, Heppert W, Groneberg DA. Neuronal plasticity in persistent perennial allergic rhinitis. *J Occup Environ Med* 47: 20–25, 2005.
19. Foster AP, Cunningham FM. Substance P induces activation, adherence and migration of equine eosinophils. *J Vet Pharmacol Ther* 26: 131–138, 2003.

20. Iwamoto I, Nakagawa N, Yamazaki H, Kimura A, Tomioka H, Yoshida S. Mechanism for substance P-induced activation of human neutrophils and eosinophils. *Regul Pept* 46: 228–230, 1993.
21. Kingham PJ, Costello RW, McLean WG. Eosinophil and airway nerve interactions. *Pul Pharmacol Ther* 16: 9–13, 2003.
22. Koyama S, Sato E, Nomura H, Kubo K, Nagai S, Izumi T. Acetylcholine and substance P stimulate bronchial epithelial cells to release eosinophil chemotactic activity. *J Appl Physiol* 84: 1528–1534, 1998.
23. Mosimann BL, White MV, Hohman RJ, Goldrich MS, Kaulbach HC, Kaliner MA. Substance P, calcitonin gene-related peptide, and vasoactive intestinal peptide increase in nasal secretions after allergen challenge in atopic patients. *J Allergy Clin Immunol* 92: 95–104, 1993.
24. Numao T, Agrawal DK. Neuropeptides modulate human eosinophil chemotaxis. *J Immunol* 149: 3309–3315, 1992.
25. Pelikan Z, Pelikan-Filipek M. Role of nasal allergy in chronic maxillary sinusitis—diagnostic value of nasal challenge with allergen. *J Allergy Clin Immunol* 86: 484–491, 1990.
26. Quarcoo D, Schulte-Herbrüggen O, Lommatzsch M, Schierhorn K, Hoyle GW, Renz H, Braun A. Nerve growth factor induces increased airway inflammation via a neuropeptide-dependent mechanism in a transgenic animal model of allergic airway inflammation. *Clin Exp Allergy* 34: 1146–1151, 2004.
27. Raphael G, Raphael MH, Kaliner M. Gustatory rhinitis: a syndrome of food-induced rhinorrhea. *J Allergy Clin Immunol* 83: 110–115, 1989.
28. Sagara H, Yukawa T, Arima M, Makino S. Effect of capsaicin on the migration of eosinophils into the bronchi of guinea pigs. *Alerugi* 42: 236–242, 1993.
29. Sato E, Koyama S, Okubo Y, Kubo K, Sekiguchi M. Acetylcholine stimulates alveolar macrophages to release inflammatory cell chemotactic activity. *Am J Physiol Lung Cell Mol Physiol* 274: L970–L979, 1998.
30. Tiberio IF, Leick-Maldonado EA, Miyahara L, Kasahara DI, Spilborghs GM, Martins MA, Saldiva PH. Effects of neuropeptides on airway and alveolar eosinophil recruitment. *Exp Lung Res* 29: 165–177, 2003.

