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Association of Ambient Ozone Exposure with Airway Inflammation and Allergy in Adults with Asthma

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Abstract

Rationale—Previous studies have demonstrated associations of high ozone levels with increased epidemiologic as well as lung function measures of asthma activity.

Objectives—In an observational study during the summer months, we hypothesized that higher ambient ozone levels are associated with more frequent symptoms, higher airway and systemic inflammation, as well as worse lung function in asthmatics as compared with non-asthmatic individuals.

Methods—Thirty-eight asthmatics and thirteen healthy control subjects residing in metropolitan Atlanta were enrolled during peak ozone season. Medical histories, quality-of-life questionnaires, spirometry, serum immunoglobulin (IgE), peripheral eosinophil counts, and exhaled nitric oxide (NO) were obtained during study visits. Personal ozone exposures over the 2 days before presentation were estimated based on location and activity surveys.

Main results—Upper airway symptoms were more frequent in asthmatics. Higher levels of ozone were associated with worse airflow obstruction, lower quality of life scores, greater eosinophilia, and higher exhaled NO levels in asthmatics. Finally, both asthmatics and non-asthmatics with allergies showed associations between air quality and airway inflammation.

Conclusions—In adults with asthma but not controls studied during peak ozone season, increasing ozone exposure predicted lower lung function and increased biomarkers of respiratory and systemic inflammation. These associations were enhanced in atopic participants, both with and without asthma. Importantly, the study findings were noted while atmospheric ozone levels were predominantly within the current and revised national air quality standards.

Keywords

ozone; asthma; lung function; allergies

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Declaration of Interest

The authors report no conflict of interest. The authors alone are responsible for the content and writing of the paper.

Introduction

Atmospheric ozone is an important component of photochemical air pollution that is formed from interaction of primary pollutants such as volatile hydrocarbons, halogenated organics, and oxides of nitrogen in the presence of sunlight (1,2). Epidemiologic studies have demonstrated increased asthma-related morbidity in association with higher levels of atmospheric ozone. Periods following higher ozone levels are associated with increases in asthma medication use, number of asthma exacerbations and hospitalizations, and risk of death (3–9). Large cohort studies have demonstrated worsening of lung function, i.e., decrement in morning peak flow rates or small airways function, in the days after elevated atmospheric ozone levels (8,10). Although asthmatics are particularly susceptible to the harmful effects of ozone air pollution, controlled exposure chamber studies demonstrated acutely diminished lung function in asthmatics as well as healthy individuals after being exposed to high concentrations of ozone (11–13). The capacity to induce pulmonary function alterations depends on ozone levels, duration and pattern of exposure, and respiratory minute volume (14). Some investigators revealed ozone air pollution's association with the incidence of asthma in adult men (15). Also, the incidence of asthma was increased among children playing three or more outdoor sports in high ozone areas (16).

Although epidemiologic and chamber protocol studies have explored the mechanisms underlying these associations, some investigators believe that ozone-associated changes in pulmonary function are greater in natural than in controlled exposure settings (11,17). Studies of children and adults in natural ozone environments have demonstrated a dose-response relationship between ambient ozone levels, asthma symptoms, and worse lung function; however, the mechanisms responsible remain poorly understood (18–22). The body of literature regarding ozone and lung health underscores the need to further define the mechanistic relevance of outdoor summer ozone levels with asthma morbidity, even if pollution levels may be within Environmental Protection Agency (EPA) standards. The purpose of this study was to use the well-defined high ozone seasons in Atlanta to examine the associations of summertime ozone levels on airway inflammation in individuals with asthma and healthy control subjects. We hypothesized that higher ambient ozone levels during the summer months are associated with increased symptoms, higher airway and systemic inflammation, as well as worse lung function in asthmatics as compared with non-asthmatic individuals.

Methods

To test the hypothesis, we conducted a cross-sectional study of asthmatics and healthy control subjects residing in the metropolitan Atlanta area during May through September 2003, 2005, and 2006. Before enrollment, participants were asked to review and sign an informed consent that was approved by the Emory University Institutional Review Board. Asthmatics included were at least 18 years of age, nonsmokers (less than 5 pack-year smoking history more than 5 years ago), and lived in metropolitan Atlanta. Individuals who were acutely ill or had received systemic corticosteroids within the last 2 weeks were excluded from participation. The definition and severity of asthma was based on clinical history and presence of airflow obstruction and/or documentation of a significant bronchodilator response (23,24). Healthy control subjects were non-smokers with no history of pulmonary disease or allergic rhinitis and normal spirometry. A brief medical and medication history was obtained, including questions about subjective exposures to pollution and allergies. *“(Have you been exposed to pollution in the last 24 or 48 hours? If yes, was the level of exposure low, medium, or high?)”*. Similar questions were asked about exposures to common allergens such as pets, pollen, mold, and dust. Participants were asked to provide hourly information about their location by zip codes and level of physical activity for the previous 2 days, subjectively

qualifying their levels of activity as low (no exertion), medium, and high exertion. Spirometry was performed according to American Thoracic Society (ATS) standards (24). A peripheral blood sample was sent to a certified clinical laboratory for leukocyte, eosinophil, and immunoglobulin E (IgE) levels. The Juniper asthma quality of life questionnaire (QOLQ) was orally administered to all asthmatic participants (25).

Testing for Atopy

Atopic status was obtained by skin testing or by history. Skin testing was not performed during the first year of the study. Historical information or documented testing was used. Subsequently, presence of allergies was determined by skin prick testing to 12 common aeroallergens (e.g., dust mite, grass, *Aspergillus*, etc.) A wheal of 2 mm and flare of 3 mm were considered positive. Individuals with prior testing within the past 2 years were not retested.

Exhaled Nitric Oxide Measurement

Exhaled nitric oxide levels (eNO) were collected in participants according to the ATS standards for offline measurements and analyzed by chemiluminescence analyzer (Sievers 280i, Boulder, CO) (26–28). Ambient NO samples were also obtained simultaneously. The Mylar collection balloons were flushed with argon gas between uses and sealed to reduce contamination by ambient NO.

Quantification of Personal Ambient Ozone Exposure

Peak 1-hour ozone levels, peak 8-hour averaged ozone levels, peak Air Quality Index (AQI) measurements, as well as temperature and relative humidity of the previous 48-hour and 24-hour time period, were abstracted from regional air monitoring data. A map of metropolitan Atlanta was used to visually determine the closest monitoring station to individuals' reported locations during each hour of the previous 2 days. Air quality data from that particular station were then abstracted from the Georgia Department of Natural Resources monitoring website (<http://www.air.dnr.state.ga.us/amp/>).

Statistical Analyses

Nonparametric tests (Wilcoxon Rank Sum) were used to evaluate differences between healthy control subjects and asthmatics with respect to quantitative measures. Linear regression modeling with biologically plausible and consistent models, excluding collinear variables, was used to measure associations of clinical parameters with ozone exposure, controlling for potential confounders. Model variables were selected due to the plausible and likely independent contributions of each variable to the outcome measure. These variables, such as subjective levels of exposures to air pollution and common allergens, 8-hour averaged levels of ozone in the previous days before evaluation, and peripheral eosinophil and IgE levels, were used as consistently as possible in models evaluated. Potential effect of enrollment year was addressed by including a "year" variable in all multivariate models; however, primary outcomes were not appreciably changed. Statistical analyses were performed with SAS version 9.1 (SAS Institute Inc, Gary, NC, USA), with p values ≤ 0.05 considered significant. Non-significant trends are also reported in certain instances where $p \leq 0.10$.

Results

Patient Characteristics

The study sample included 38 asthmatic patients and 13 healthy control subjects between May and September. Twelve asthmatic patients and 5 control subjects were studied during the 2003 season (during which 13 days exceeded air quality standards); 20 asthmatic patients and 6

control subjects were studied in 2005 (during which 17 days exceeded air quality standards), and 6 asthmatics and 2 healthy control subjects were studied in 2006 (during which 30 days exceeded air quality standards). One asthmatic individual was not able to reliably perform spirometry but did provide all other biosamples and clinical information and was therefore included in the analyses.

Clinical features of enrolled individuals are presented in Table 1. Since a significant number of men screened for enrollment were smokers, a higher number of women participated in the study. As expected, participants with asthma demonstrated worse airflow obstruction. Markers of allergy and inflammation were more prevalent in the asthmatics. Over half of asthmatics had confirmed allergies by skin testing. However, three control subjects had skin tests positive as well (Table 1). Fifty percent of the asthmatic group had mild disease, and the rest had moderate or severe asthma based on Global Initiative for Asthma-GINA criteria (Table 2) (23). Medication history demonstrated that frequency of controller therapy increased with severity. A small number of individuals with moderate or severe asthma (8%) were not on any controller therapy at the time of evaluation.

Ambient Ozone and Activity Levels

Peak 1-hour and peak 8-hour averaged levels recorded by ambient posted monitors nearest to enrolled individuals in the 24 and 48 hours before study visits were similar in the asthmatic and non-asthmatic groups (Appendix Table A1). Subjective estimation of high pollution exposures (“medium” or “high” over prior 48 hours) also did not significantly differ between the asthma and healthy control group. In any given time period (day of visit, 1 day before, or 2 days before participant evaluation), 8-hour averaged atmospheric ozone levels were inversely related to relative humidity ($R < -0.60$, $p < 0.001$) and correlated with temperature ($R > 0.45$, $p < 0.01$). Reported levels of activity and exertion were also similar between groups over the previous 2 days (all $p > 0.10$, data not shown).

Symptoms

Frequency of upper airway symptoms such as itchy eyes, runny nose, sneezing, and congestion was higher in the asthmatic group, and headaches also tended to be higher (Table 1). Stepwise logistic regression analyses with symptoms as the outcome variables were performed. Independent variables included in the model were presence of asthma, use of nasal steroids, measured ozone levels, and subjective levels of exposure to allergen and pollution. Presence of asthma and high pollution exposures were significantly associated with itchy eyes (asthma OR = 12.62 [1.14–139.99], allergen exposure OR = 10.18 [2.12–48.88]). High allergen exposures was the only independent variable significantly associated with headaches, sneezing, and congestion (OR 1.67 [1.06–2.61], 1.72 [1.09–2.72], and 2.05 [1.26–3.33], respectively).

Quality of Life

Of a best possible score of seven, the median total quality-of-life score in our panel of asthmatics was 5.22, with a range from 2.16 to 7.00. The forced expiratory volume in 1 second (FEV₁) percent predicted was associated with the overall score ($R = 0.35$, $p = 0.04$), and the emotional domain score was especially related ($R = 0.50$, $p < 0.01$). Linear regression models controlling for recent high allergen exposures, activity levels, and atmospheric ozone levels in the previous 2 days estimated that a 50% increase in baseline % FEV₁ would be associated with an average increase of the quality-of-life score by 1. In contrast, increases in atmospheric peak ozone levels by only 0.020 ppm (a plausible difference from high to low ozone days) was associated with an average decrease in the quality of life score by a half a point (Table 3).

Markers of Allergic Inflammation: Eosinophils, (IgE), and Exhaled NO

Peripheral eosinophilia in asthmatics was higher in association with peak ozone levels the day before evaluation (Figure 1A), whereas no such association was seen in the healthy control group (Figure 1B). Linear regression modeling of peripheral eosinophilia and ozone levels, controlling for FEV₁ % predicted tended to demonstrate a lag effect for ozone levels. Increases in peak 8-hour averaged ozone levels every 0.020 ppm was associated with a 1.6 percent higher number of eosinophils (Table 4). A similar tendency for a 2-day lag effect was also noted, such that a 0.020 ppm increase in 8-hour averaged ozone levels was associated with an average 1% higher peripheral eosinophilia ($p \leq 0.10$ for model and ozone variable coefficient).

Exhaled NO values were higher in the asthmatics ($n = 29$) than healthy control subjects ($n = 11$) in whom off-line samples were collected (Table 1). Exhaled NO was inversely related to airflow (FEV₁/forced vital capacity [FVC] ratio % predicted, and FEV₁ and peak flow % predicted) and directly correlated with post-bronchodilator changes in FEV₁ and FVC (Appendix Table A2). Interestingly, in asthmatics, exhaled NO also correlated with peak ozone levels 1 to 2 days before evaluation and tended to correlate with subjective exposures to pollution (Table 5). Adding allergen exposures did not contribute to the significance of the model. Correlation between ozone and exhaled NO levels was particularly strong among the known atopic asthmatics (Figure 2) Linear regressions of exhaled NO and ozone levels, controlling for FEV₁ % predicted, demonstrated that for every 0.020 ppm increase in 8-hour averaged ozone levels, exhaled NO levels would be higher by nearly 3 ppb (model $p = 0.07$, coefficient for zone = 2.78 [C.I. 0.09–5.47], $p = 0.05$) (Appendix Table A3). Introducing peripheral eosinophils into the model predicted similar increases in exhaled NO with ozone levels (data not shown). Adding variables for subjective exposure to high pollution and eosinophils to the model estimated that a positive response to “high pollution” was associated with a 7 ppb higher levels of exhaled NO, thereby supporting the premise that individual estimations of pollutant exposures are likely to be accurate (Table 6).

Associations of Ozone Exposure with Lung Function and Inflammation

Percent predicted FEV₁/FVC ratio was inversely related to peak 8-hour ozone levels 2 days before testing evaluation ($R = 0.34$, $p = 0.04$). When controlling for IgE, peripheral eosinophilia, subjective level of recent allergen and pollution exposures, and 8-hour averaged ozone levels, eosinophilia and pollution exposures were related to reduced airflow (Table 7). Subjective exposures to high levels of air pollution, controlling for allergen exposures, was associated with, on average, a 14-point lower (95% C.I. 3, 25) predicted FEV₁/FVC ratio. Similar associations were seen in the group of all enrolled (asthma and non-asthma), especially in those with allergies (Appendix Table A4) where subjective exposures to pollution was associated with an average 16-point lower predicted ratio. In addition, increases in peak 8-hour averaged levels of ozone by 0.020 were associated with an average 8-point lower % FEV₁/FVC ratio (95% C.I. 3,14). Linear regression modeling demonstrated a trend of reduced FEV₁ percent predicted with subjective reporting of high exposures to air pollution (Table 8). Adding temperature and relative humidity variables into models predicting FEV₁/FVC or FEV₁ % predicted did not demonstrate significant associations or alter the relationships of the original model. Only in the group of all atopic individuals, it was estimated that every 5% higher level of relative humidity was associated with a 4-point higher % FEV₁/FVC ratio.

Discussion

This study demonstrates an association of ozone lag effects on the morbidity of asthma, specifically quality-of-life scores, airway inflammation, and lung function. Air quality indices the days previous to our subjects' study visits indicate that ozone levels were adequately high to test our hypothesis, although the predominance of exposure levels were within the current

and revised standards for peak 1-hour and 8-hour averaged ozone (29). In this panel study, airway inflammation was associated with higher ozone levels, controlling for factors that are expected to contribute to airway inflammation. Airways obstruction also appeared to be associated with higher subjective pollution levels, regardless of estimated levels from posted ambient ozone monitors. Similar to findings in other studies (30), these asthmatics had more frequent upper airway symptoms. Also, air pollution exposures were negatively associated with quality-of-life scores. Prior protocolized experimental studies that have demonstrated increased allergenicity from ozone priming were underscored by this real world translational pilot study that demonstrated increased eosinophilia and airway inflammation (as measured by exhaled NO) in relation to recent higher ozone exposures.

Ground level ozone pollution is formed from motor vehicle exhaust, sunlight, and high temperatures (2). Ambient ozone levels usually vary between 0.020 and 0.040 parts per million (ppm). Moderate elevations in levels are usually 0.070 to 0.120 ppm, usually during the peak ozone months of May through September. The air quality standard for ozone, which is designed to protect public health with an adequate margin of safety, is 0.080 ppm, averaged over 8 hours, specifically “the 3-year average of the fourth-highest daily maximum 8-hour average ozone concentrations measured at each monitor within an area over each year must not exceed 0.080 ppm” (29).

Epidemiologic studies have demonstrated that asthmatics are particularly susceptible to poor air quality, and multiple mechanisms may account for that association. The immediate effect appears to be transient and neurally mediated. FEV₁ and FVC return to pre-exposure levels within 12 to 24 hours. However, airway resistance remains elevated, indicating a longer-term consequence of exposure (31,32). The association with air pollution appears to be with a lag-effect, particular with ozone (5,6,8,9). Changes in lung function after ozone exposure are associated with an exuberant airway inflammatory response. The magnitude of this response is heightened in individuals with asthma, with infiltration of both neutrophils and eosinophils into the airways (2,32–35). Furthermore, the asthmatic response to inhaled allergen is amplified with pre-exposure to ozone (17,36,37).

The similar peak ozone exposures in our healthy and asthmatic cohorts during their routine every-day activities in Atlanta provided an ideal real-life opportunity to evaluate symptoms, lung function, and airway inflammation between the two groups. In addition, this study was performed in the natural environment with particular attention to activity level and provides complementary data and validation of findings in controlled experimental studies. Other investigators have suggested that the mechanism of chronic allergic inflammation and susceptibility to ozone-related airway inflammation are related (38). In addition, responses to inhaled allergen are also enhanced after exposure to ozone (36). In our study, individuals with asthma tended to demonstrate worse airflow obstruction and enhanced biomarkers of allergic inflammation associated with ozone exposure, with increased eosinophilia and exhaled NO levels, even controlling for lung function.

The importance of self-reported exposures was evident in this study and may be partially explained by heightened perception of exposures by those who are most affected (39). However, in this study, objective clinical markers underscored the validity of their reports. Therefore, personal immediate exposures and subjective sense of exposures are important and cannot be estimated by distant and generalized measures alone. Asthmatics' exhaled NO levels were higher in association to perceptions of high pollution exposure (even controlling for allergen exposure). In addition, individuals' perceptions of recent exposures to high pollution levels were related to lung function. Prior studies have described changes in FEF_{25–75}, peak expiratory flows, and %FEV₁ after ambient exposure (10,40,41). Recent measured ozone levels did not demonstrate a direct relationship to %FEV₁ in asthmatic participants in this study.

However, individual perception of pollution exposures appeared to play a role. The lack of correlation of larger airways function to ozone levels in our cohort may be due to tolerance developed over the ozone season or insufficient subjects. Ozone chamber studies show that changes in FEV₁ and FVC normalize after 12 to 24 hours of ozone exposure but that the increase in airway resistance often persists (31). No airway resistance measurements were made in this study; therefore, such associations may have been missed, perhaps accounting for worse quality-of-life scores with higher ozone levels, even after controlling for lung function.

The role of other pollutants in increasing morbidity related to chronic lung diseases should not be underestimated. Particulate matter (PM), which is the most common type of pollution associated with industrial and traffic-related sources, refers to solid particles or droplets that are suspended in the air. These particles can come from natural sources or industrial sources and are categorized by size (fine particles are those less than 2.5 microns or less in diameter) (42,43). Epidemiologic and experimental evidence has supported associations of PM and asthma morbidity (42–46). For example, children attending schools within 400 meters of major highways and freeways had increased respiratory symptoms suggestive of asthma (42). Although this current study was not designed to evaluate the role of PM pollution with asthma symptoms and airway inflammation, there were some individuals in whom PM contribution could be estimated with geographic modeling and home addresses. The spatial relationships of PM are more relevant than in ozone due to dispersion and individuals' immediate environments; therefore, firm conclusions cannot be drawn. Despite those limitations, 24- and 48-hour prior measures of ozone and PM were significantly correlated ($R > 0.65$, $p < 0.001$) for all instances in which PM were able to be determined. All models for lung function with ozone were run including PM values, and no significant plausible contributions were noted.

As stated in the hypothesis, this study was designed to evaluate associations between air quality (ozone levels) and asthma activity and inflammation. However, there are certainly limitations to this panel study. Although air pollution levels between groups were similar, individuals were recruited over three high ozone seasons and were evaluated in a cross-sectional and not serial manner. Another limitation is that despite attempts to keep enrollment equal between genders, there was a predominance of women enrolled in the study. In addition, the asthmatic cohort was predominantly African American. We may in part be demonstrating some phenomenon that may be enhanced in certain ethnic populations and that requires further study. As of now, there is no literature of antioxidant defenses and racial background affecting responses to air pollution. However, it is known that areas of increased pollution owing to traffic and urban development are frequently worse in areas where minorities reside (47–49). Next, most, but not all, asthmatics were atopic; therefore some misclassification did occur. However, models did allow for atopy to be evaluated, and subjective levels of allergen exposure were included in most models. Atopy did not play a significant role in any associations except for exhaled NO levels. A reasonable conclusion is that even if smaller groups are evaluated in such translational exposure studies, real-world ozone levels did demonstrate biologically and scientifically plausible associations, underscoring other investigators' premise that controlled chamber studies likely underestimate the effects of true outdoor air pollutants (11,17). Ambient posted monitors have been used thus far to evaluate lung health and ozone levels (20,50). Based on our results, it appears that a combination of exposure measures, such as measured outdoor, personal monitor levels, and subjective levels of exposure, will show the most accurate estimates of associations of pulmonary effects of air quality in real-world field studies.

Conclusion

In conclusion, in this real-world observational study, we demonstrate that asthmatics exposed to ambient atmospheric ozone levels during high ozone season experienced worse symptoms and had lower quality-of-life scores, worse lung function with increased airflow obstruction,

and enhanced allergic inflammation. Although other factors potentially play a role in pollutant-related airway inflammation, such as baseline lung function or individual antioxidant capability, the associations seen in this study were evident with mean pollutant levels within the current air quality standards (22,29).

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Appendix

Table A1

Peak levels of ozone (ppm) days prior to evaluation.

	Asthmatics		Controls		p value*
8-hour averaged ozone levels 2 days prior	0.061	(.032–.074)	0.056	(.037–.064)	0.97
1-hour ozone levels 2 days prior	0.072	(.045–.087)	0.066	(.050–.082)	0.99
8-hour averaged ozone levels 1 day prior	0.059	(.044–.073)	0.065	(.057–.083)	0.35
1-hour ozone levels 1 day prior	0.070	(.049–.080)	0.077	(.070–.094)	0.28
Air Quality index [†]					
2 days prior	75	(53–101)	76	(61–111)	0.53
1 day prior	75	(55–103)	97	(70–121)	0.18
Subjective exposure to high pollution [‡]	26	(68%)	6	(46%)	0.17

Data presented as median (IQR);

* p-value based on wilcoxon rank sum testing

[†] AQI defined as the index of air quality as determined by the highest level of criteria pollutants: PM, Ozone, sulfur dioxide, nitrogen dioxide, and carbon monoxide.

[‡] Those reporting exposure to medium to high levels of pollution during prior 2 days.

Table A2

Exhaled NO and lung function in asthma.

Exhaled NO and	R (Spearman)	p	Numbers
FEWFVC Ratio	−0.567	0.002	28
% Pred Ratio	−0.53	0.003	28
%FEV ₁	−0.436	0.021	28
%PEFR	−0.437	0.020	28
%FEF	−0.567	0.002	28
Change FEV ₁ post BD	0.649	0.0006	24
Change FVC post BD	0.468	0.021	24

Table A3

Exhaled NO and air pollution in asthma (model $p = 0.07$).

	Parameter estimate	95% C.I.	p*
Intercept	16.69	1.83, 31.55	0.04
FEV ₁ % predicted	−0.12	−0.28, .04	0.18
Change of 0.02 ppm of 8-hour averaged ozone levels 2 days prior	2.78	0.09, 5.47	0.05

* p value: significance of parameter estimate in linear regression model.

Table A4

Predicted ratio in allergic asthmatics and nonasthmatics (model $p = 0.02$).

	Parameter estimate	Std Err	p
Intercept	148.64	16.46	<0.01
Subjective exposure to high allergen *	-1.60	1.87	0.41
Subjective exposure to high pollution *	-16.44	6.81	0.04
Change of 0.02 ppm of 8-hour averaged ozone levels 2 days prior	-8.33	2.80	0.02
Peripheral blood % eosinophils	-215.00	1.94	0.30
Serum IgE level	-0.006	0.003	0.07
Asthmatic	-12.57	11.46	0.30

* Those reporting exposure to “medium” to “high” levels of pollution or allergen during prior 2 days.

* p value: significance of parameter estimate in linear regression model.

Figure 1. Eosinophilia and prior ozone levels in asthmatics (a) $R = 0.41$, $p = 0.03$ vs. controls (b) $R = 0.12$, $p = 0.73$.

Figure 2.
Correlation in atopic asthmatics between ozone levels 2 days prior and exhaled NO levels, $R = 0.60$, $p = 0.014$.

Table 1

Subject characteristics.

	Asthmatics	Controls	<i>p</i> value*
N	38	13	
Age	44 (31–50)	40 (29–55)	1.00
Gender F:M	32:6	12:1	
BMI	32 (25–40)	30 (23–41)	0.49
Race-ethnicity			
Caucasian	11	4	
African- American	26	8	
Asian	0	1	
Other	1	0	
Lung function			
% FEV ₁ /FVC ratio	91 (81–101)	97 (96–102)	0.04
% FEV ₁	79 (69–90)	88 (82–94)	0.07
%PEF	78 (66–99)	83 (66–88)	0.52
Markers of blood leukocytes ($\times 10^3$)	6.8 (5.7–8.6)	5.3 (4.8–6.4)	0.07
Eosinophils (%)	2 (2–4)	2 (1–2)	0.07
Neutrophils (%)	62 (53–68)	53 (41–62)	0.09
IgE (IU/mL)	134 (50–324)	34 (14–74)	<0.01
eNO (ppb)	11.4 (9.8–17.0)	7.1 (6.3–12.5)	0.03
Results of allergen skin tests:			
>1/12 positive	20	1	
1/12 positive	3	2	
0/12 positive	8	5	
History of atopy (no skin test)	7	5	
Frequency of upper respiratory symptoms			
Itchy eyes	22/34	64 1/12	8 <0.01
Runny nose	16/34	47 0/12	0 <0.01
Sneezing	17/34	50 1/12	8 0.02
Congestion	15/38	39 1/13	8 0.04

	Asthmatics	Controls	<i>p</i> value [*]
Headaches	13/38	34 1/13	8 0.08

Data presented as median (IQR);

^{*} *p* value based on Wilcoxon Rank Sum,

⁺ *p* value based on Fischer Exact Test.

Table 2

Asthmatic Global Initiative for Asthma (GINA) Classification (n = 37)* by severity and medication.

Severity	N	ICS only	ICS +LTRA	LTRA only	Neither ICS nor LTRA
Mild (%FEV ₁ > 80)	18	6	3	4	5
Moderate asthma (%FEV ₁ 60–80)	13	5	5	1	2
Severe (%FEV ₁ < 60)	6	4	1	0	1
Total	37	15	9	5	8

* One asthmatic patient was unable to adequately perform spirometry.

ICS = inhaled corticosteroids; LTRA = leukotriene receptor antagonist.

Table 3Asthma quality-of-life scores and exposures (model $p = 0.07$).

	Parameter estimate	95% C.I.	<i>p</i>
Intercept	2.97	−0.58, 6.52	0.11
FEV ₁ % predicted	0.02	0.00, 0.04	0.05
Subjective exposure to high allergen* activity levels 2 days prior	0.08	−0.19, 0.35	0.57
	0.05	−0.05, 0.15	0.40
Change of 0.02 ppm of 8-hour averaged ozone levels 2 days prior	−0.46	−0.85, −0.07	0.03

* Those reporting exposure to “medium” to “high” levels of pollution or allergen during prior 2 days.

Table 4Eosinophilia and air pollution in asthmatics (model $p = 0.03$).

	Parameter estimate	95% C.I.	<i>p</i>
Intercept	1.95	−4.79, 8.69	0.58
FEV ₁ %predicted	−0.04	−0.12, 0.04	0.26
Change of 0.02 ppm of 8-hour averaged ozone levels 1 day prior	1.63	0.41, 2.85	0.02

Table 5

Exhaled NO and air quality in asthmatics, atopic asthmatics.

Correlations of exhaled NO and air quality	R (Spearman ^ρ)	<i>p</i>	Numbers
All asthmatics:			
Peak 8-hour avg ozone levels 2 days prior	0.428	0.21	29
Peak 1-hour ozone levels 2 days prior	0.415	0.025	29
Subjective exposure to high pollution*	0.34	0.07	29
Atopic asthmatics:			
Peak 8-hour avg ozone levels 2 days prior	0.601	0.014	16
Peak 1-hour ozone levels 2 days prior	0.546	0.028	16
Subjective exposure to high pollution*	0.39	0.13	16

* Those reporting exposure to “medium” to “high” levels of pollution during prior 2 days.

Table 6Exhaled NO and air pollution in asthma (model $p=0.02$).

	Parameter estimate	Std Err	<i>p</i>
Intercept	-5.70	11.31	0.62
FEV ₁ % predicted	0.0005	0.0969	1
Change of 0.02 ppm of 8-hour averaged ozone levels 2 days prior	3.10	1.40	0.04
Subjective exposure to high pollution * peripheral blood % eosinophils	7.88	3.56	0.04
	2.56	1.17	0.04

* Those reporting exposure to “medium” to “high” levels of pollution during prior 2 days.

* *p* value: significance of parameter estimate in linear regression model.

Table 7FEV₁/FVC ratio (% predicted) in asthmatics (model $p < 0.01$).

	Parameter estimate	95% CI	<i>p</i>
Intercept	119.22	104.77, 133.67	<0.01
Subjective exposure to high allergen *	-0.70	-4.52, 3.12	0.72
Subjective exposure to high pollution *	-13.65	-24.76, -2.54	0.03
Change of 0.02 ppm of 8-hour averaged ozone levels 2 days prior	-3.14	-7.78, 1.48	0.20
Peripheral blood % eosinophils	-2.24	-3.96, -0.52	0.02
Serum IgE level	-0.007	-0.013, -0.001	0.04

* Those reporting exposure to “medium” to “high” levels of pollution or allergen during prior 2 days.

* *p* value: significance of parameter estimate in linear regression model.

Table 8FEV₁ percent predicted in asthmatics (model $p = 0.04$).

	Parameter estimate	95% CI	p^*
Intercept	97.00	77.30, 116.70	<0.01
Subjective exposure to high allergen *	1.65	-45.4, 48.69	0.50
Subjective exposure to high pollution *	-11.84	-25.21, 1.53	0.10
Change of 0.02 ppm of 8-hour averaged ozone levels 2 days prior	1.28	-4.55, 7.10	0.67
Peripheral blood % eosinophils	-0.95	-3.09, 1.19	0.39
Serum IgE level	-0.010	-0.018, -0.002	0.02
On inhaled corticosteroid treatment	-12.08	-24.45, -0.29	0.07

* Those reporting exposure to “medium” to “high” levels of pollution or allergen during prior 2 days.

* p value: significance of parameter estimate in linear regression model.