Published in final edited form as:

Arch Environ Occup Health. 2009 ; 64(3): 168–176. doi:10.1080/19338240903240616.

Interaction of Ambient Air Pollution With Asthma Medication on Exhaled Nitric Oxide Among Asthmatics

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Abstract

The interaction between ambient air pollution and asthma medication remains unclear. The authors compared airway inflammation response to air pollution among asthmatics. Increases of 10 ppb of nitrogen dioxide (NO₂) and of 10 µg/m³ of particulate matter < 10 micron in diameter (PM₁₀) daily concentrations were associated with an increase in exhaled nitric oxide (eNO) of 0.13 ppb (95% confidence interval = 0.06, 0.19) and of 0.07 ppb (95% confidence interval = 0.02, 0.12), respectively, in models adjusted for important covariates. The results show that the medication could not counteract airway inflammation effects of air pollution. Specifically, the patients on triamcinolone decreased the sensitivity to PM₁₀ but increased the sensitivity to NO₂. The patients on salmeterol were more vulnerable to both NO₂ and PM₁₀. This study indicates that the current pollution levels may still enhance airway inflammation among patients with persistent asthma even when they are on asthma medications.

Keywords

air pollution; asthma; exhaled nitric oxide; salmeterol; triamcinolone

Ambient nitrogen dioxide (NO₂), particulate matter < 10 µm (PM₁₀), ozone (O₃), and sulfur dioxide (SO₂) adversely affect asthma.^{1–5} However, there is still disagreement on the levels of pollution that would significantly affect asthmatics and on the magnitudes of the estimated health effects.^{6–7} These disagreements are due to an insufficient understanding of the underlying mechanisms responsible for the effects of exposure to specific pollutants; they may also be due to measurements being combined with varying levels of exposure errors, a fundamental flaw in most traditional epidemiological studies. Accumulating data suggest that inflammatory processes play a key role in air pollution health effects.

Nitric oxide is one of the smallest biologically active substances in the human body. This short-lived intercellular messenger has been recognized as a key signaling molecule in a

wide variety of biological functions including modification of the inflammatory responses, airway vascular tone, and nonadrenergic noncholinergic (NANC) transmission.⁸ The production of endogenous NO is controlled by the various isoforms of nitric oxide synthase (NOS).⁹ The likeliest source for the majority of exhaled NO (eNO) in asthma seems to be inducible oxide synthase (iNOS). In the lung, activation of the iNOS by proinflammatory cytokines, macrophages, neutrophils, epithelial cells, endothelial cells, and nonadrenergic noncholinergic neuroges can produce NO.¹⁰

Recent epidemiologic studies have shown that eNO is a promising biomarker in assessing health effects of air pollution on airway inflammation.^{11–13} However, few studies have examined the association between air pollution and eNO in patients with persistent asthma on well-defined medication regimens that are administered in an asthma clinical trial.^{14–15} The objectives of the present study are to examine whether changes in eNO are associated with short-term changes in air pollution among patients with persistent asthma on a regimen of an inhaled corticosteroid, an inhaled long-acting β_2 -agonist, or a placebo and to examine whether the asthma medication regimens modify the associations. Our global hypothesis is that ambient levels of pollutants adversely affect the health of asthmatics despite medication use. We further hypothesize that anti-inflammatory agents reduce the exposure–response relationship by blocking airway inflammation and that bronchodilators enhance exposure–response relationships by increasing the depth of the delivered dose of a pollutant.

Methods

Study Cohort and Protocol

In the present study, we used a panel study design. The participants were in the National Heart, Lung and Blood Institute sponsored Salmeterol Off Corticosteroids Study (SOCS) of the Asthma Clinical Research Network (ACRN).^{16–17} SOCS was a 28-week clinical trial conducted at 6 university-based ambulatory care centers from February 1997 to January 1999.¹⁸ The 6 centers were located in 6 cities: Boston; New York; Denver; Philadelphia; San Francisco; and Madison, WI. The participants were nonsmokers aged 12–65 years. After a 6-week run-in period during which the 164 participants received inhaled corticosteroid (triamcinolone, 4 puffs BID) and as-needed rescue albuterol, the participants were randomized into 3 groups: 54 participants in the group on an inhaled corticosteroid (4 puffs twice per day of triamcinolone acetonide), 54 participants in the group on an inhaled long-acting β_2 -adrenergic agonist (2 puffs twice per day of salmeterol xinafoate), and 56 participants in the group on a placebo (2 puffs twice per day) for 16 weeks. All participants then stopped medication for an additional 6-week study period (run-out).

In the present study, we used eNO collected from 119 participants who had repeat eNO measures at their scheduled clinical visit dates during the 16 weeks of active treatment (randomization) in the SOCS trial. The SOCS study had developed a standardized protocol and definitions common to the 6 participating asthma clinical centers for the measurements. The ACRN Steering Committee oversaw the quality control and the conduct of the study. All the 6 centers had internal review board approval. The Human Subject Protection Office of the Penn State College of Medicine also approved the study protocol. The protocol has been reported elsewhere.¹⁶

eNO Measurements

All eNO tests were performed at each center with the use of equipment and procedures that were standardized for the entire ACRN.¹⁹ The research staff were trained and certified to ensure proficiency and uniformity in all procedures. Each center strictly followed standardized quality control and assurance procedures.¹⁸ Participants performed the

maneuver in a sitting position with a nose clip. Initially, participants maintained a tidal breath for 30 seconds through the ambient nitric oxide removal apparatus (ANORA). The ANORA removes nitric oxide from the ambient air while a subject is tidal breathing to control for the effect of variation in ambient nitric oxide on the mixed expired nitric oxide determination. Then, the participants exhaled a single vital capacity into a mylar balloon. The balloon remained closed until the exhaled air was analyzed within 48 hours. The Sievers 280 Nitric Oxide Analyzer (Sievers Instruments, Boulder, CO) was used to measure eNO.¹⁹

Pollution and Climate Variables

Air pollution data were derived from the US Environmental Protection Agency (EPA) Air Quality System (AQS) Database.²⁰ These measurements are subject to uniform criteria for monitoring, siting, instrumentation, and quality assurance. In the AQS database, gaseous pollutants NO₂, O₃, and SO₂ are reported hourly, and 24-hour average concentrations of NO₂ and SO₂ and 8-hour maximum concentrations of O₃ can be produced conveniently. PM₁₀ is reported daily. We first identified all the monitors, which are closest (less than 20 miles between a monitor location and the centroid of a zip code) to the zip code centroid of the participants' home addresses. Then, monitor-specific mean concentrations of pollutants were calculated. We further calculated zip-code-specific mean concentrations by averaging all available monitor-specific concentrations in each zip code. Based on the dates of the eNO and pollutant measures, we determined the residential zip code centroid-specific daily mean concentrations for the 4 days prior to the eNO measurements for each participant. Thus, the variable was developed to serve as a surrogate for the short-term exposure to NO₂, PM₁₀, O₃, and SO₂, respectively, for each participant as day-0, day-1, day-2, day-3, and 4-day daily mean concentrations. Meteorological data were obtained from the National Climate Data Center for the same zip code areas.

Statistical Analysis

We performed an analysis that used a longitudinal linear regression model to determine the associations between the study pollutants and eNO while controlling for age, sex, race/ethnicity, asthma clinical center, season, week, daily average temperature, and daily average relative humidity. This model allowed for interval-independent variables and used the method of restricted maximum likelihood to estimate parameters.²¹ SAS PROC MIXED was used to carry out the computations required to fit this model. Our data analyses were carried out in 3 steps. First, we generated descriptive statistics to check the validity of the variables and their distributions and to identify potential outliers. We also obtained the descriptive statistics stratified by major covariates such as treatment group to provide insight into potential confounders and effect modifiers.

Second, we performed bivariate as well as multiple regression analyses. Using bivariate analysis and synthesizing the literature, we identified important covariates that belong in the subsequent regression models. Then, a pollutant effect was estimated by entering a pollutant separately into the model (single pollutant model). Daily pollutant concentrations lagged up to 4 days were examined to take into account air pollution exposure on the days preceding the eNO measures. We also examined the effects of cumulative exposure to the pollutants by entering the exposure variable (up to 4 days) into the models.

Third, we tested interaction terms among independent variables in the models. We paid special attention to separate interactions of pollutants with treatment group. We conducted stratified analyses by dividing the study participants into the 3 medication regimens to test whether the slope was different from zero in each medication regimen group.²²

Results

Description of Study Population

Repeated samplings of eNO were collected on 119 SOCS study participants at scheduled visits. The characteristics of the 119 participants were not statistically different from those of the excluded 45 SOCS participants who did not have samplings of eNO at scheduled visits (results not shown). Of the 119 participants, a total of 555 breath samples were available for this study. These eNO samples were collected when participants were scheduled for clinical visits every 2–4 weeks during the 16-week period. Also, 159 samplings were missing because of participant withdrawals. Further exclusions were made because of missing pollution data (ie, $n = 75$ for O_3). Thus, the remaining sample size in the present study was 480 person-days of observations (see Table 1). A majority of the participants were female and aged 20–40 years. In addition, 32% of the participants were minorities. The mean eNO level in the triamcinolone group was lower than those in the other 2 groups.

Air pollution levels

The pollution levels of NO_2 , PM_{10} , and SO_2 were all below the current US National Ambient Air Quality Standard (NAAQS; see Table 2).²³ No days had NO_2 concentrations higher than 53 ppb, which is the current standard for the NO_2 annual arithmetic mean. The highest daily mean PM_{10} concentration was lower than the current NAAQS of $150 \mu g/m^3$, and the highest daily mean SO_2 concentration was far lower than the current NAAQS of 140 ppb. For ozone, however, the highest 8-hour mean concentration was higher than the current standard of 80 ppb. There was no statistically significant difference in the mean concentrations of NO_2 , PM_{10} , O_3 , and SO_2 between the 3 medication groups.

Regression Analysis

An increase of 10 ppb of NO_2 was associated with an increase in eNO on the same day (0.13 ppb, 95% confidence interval [CI] = 0.06, 0.19; see Table 3). The significant associations of NO_2 with eNO were also observed on previous one day (0.08 ppb, 95% CI = 0.02 to 0.14) and the average of the previous 4 days (0.11 ppb; 95% CI = 0.01, 0.20). Similarly, an increase in PM_{10} concentration of $10 \mu g/m^3$ was associated with an increase in eNO of 0.07 ppb (95% CI = 0.02, 0.12). In contrast, a negative association was observed for the O_3 concentration of 10 ppb with eNO (−0.09 ppb; 95% CI = −0.13, −0.05) on the same day. The negative associations also existed on lag 2 day (−0.05 ppb; 95% CI = −0.09, −0.01) and the average of the previous 4 days (−0.08 ppb; 95% CI = −0.14, −0.02). No significant associations were observed for SO_2 . In general, the largest effects were found on the same day (lag 0) of the eNO measured. Two pollutant models of NO_2 and PM_{10} led to attenuation of both regression coefficients to nonsignificant levels (see Table 4). These reductions were likely the result of multicollinearity. However, the effect of O_3 remained statistically significant in models that included NO_2 , PM_{10} , or SO_2 .

Across the 3 medication regimen groups, stronger associations between NO_2 and eNO were observed among the patients treated with triamcinolone or salmeterol. The results indicate that the 2 medication regimens did not protect against the effects of NO_2 . The strongest association between PM_{10} and eNO was observed among the patients treated with salmeterol; however, PM_{10} was not associated with eNO among the patients treated with triamcinolone. These results indicate that salmeterol increased the sensitivity to PM_{10} , and triamcinolone decreased the sensitivity.

Comment

Although studies have been conducted to evaluate the extent to which the use of asthma medication modifies the health effects of air pollution, the results have not been clear. The findings from previous studies are contradictory, and the magnitudes of reported effects have been questioned. For example, Hiltermann et al²⁴ reported that stratification by steroid use did not affect the magnitude of the observed associations between the shortness of breath with NO₂ and PM₁₀. Contrary to this observation, Peters et al²⁵ reported that medication use was not a confounder but attenuated the associations between particle pollution and peak expiratory flow and symptoms in asthmatics. Using improved exposure data, recent studies on eNO and air pollution have supported the findings reported by Peters et al that the use of ICS modified the association between eNO and particle pollution.^{12–13,15} However, the reported direction of modification was different. Delfino et al¹² reported that the associations between (a) eNO and ambient particle matter with a diameter of less than or equal to 2.5 microns (PM_{2.5}) and (b) personal and ambient organic carbon were only significant in participants taking ICS alone. Koenig's group found no associations between eNO and PM_{2.5} in the children on ICS therapy.^{13–15} Our study demonstrates that ambient air pollution is associated with eNO among patients with persistent asthma on well-defined asthma medication regimens. The significant associations were observed in all three asthma medication regimens. These results appear to show that the typical asthma medication could not counteract airway inflammation effects of air pollution among patients with persistent asthma. Specifically, the patients on triamcinolone decreased the sensitivity to PM₁₀ but increased the sensitivity to NO₂. We further observed that the patients on salmeterol appeared to be more vulnerable to NO₂ and PM₁₀, compared with the placebo group.

While exposure metrics and study populations were different, the differences between our findings and previous ones may be due to the major difficulties involved in the study of the interaction between the exposure and asthma medication, such as the difficulty of collecting precise information about various asthma medication regimens as well as about homogeneity of asthma status. These difficulties stem largely from the study of people who are living in uncontrolled environments. By using asthma clinical trial data, we could make full use of more specific and more precise information on asthma medication regimens, extensive and high quality health outcome measures, and on documentation of homogeneous asthma status at the baseline. In addition, the 119 study participants provided 480 person-days of observations. This sample size is larger than those in the previous similar studies.^{12–13} Therefore, we were able specifically to assess health effects of air pollution among asthmatics on the 3 asthma medications.

Previous studies have shown that long-acting beta-adrenergic agonists are effective as bronchodilators.²⁶ However, overuse of the agonists, or their use as monotherapy, may contribute to a worsening of asthma control.²⁷ McIvor et al²⁷ showed that patients with persistent asthma well controlled by low doses of triamcinolone cannot be switched to salmeterol monotherapy without risk of clinically significant loss of asthma control. They further proposed that treatment with salmeterol may mask worsening airway inflammation and delay awareness of worsening asthma. In addition, Lazarus et al¹⁸ showed that salmeterol was not more effective than a placebo at suppressing airway inflammation. Under conditions of air pollution, it is plausible that asthma patients treated with β_2 -agonists may experience more inhalation exposure to pollutants because the bronchodilation may result in greater pollutant deposition in the lower airways. This may be particularly true for the respiratory effect of the coarse particles. In the inhaled corticosteroid group, PM₁₀ was not significantly associated with eNO. Overall, we found less significant associations in the inhaled corticosteroid group, with the exception of the effects of NO₂. These results may

indicate that inhaled corticosteroid could effectively counteract airway inflammation by PM₁₀.

We observed consistent effects of NO₂ on airway inflammation in the present study. Although the effects of NO₂ have been proven to be plausible in experimental studies, only mild airway inflammation has been demonstrated in human experimental studies of exposure to a concentration of NO₂ (2000 ppb) that is nearly 2 orders of magnitude higher than the estimated mean ambient exposure of the participants in this study.^{28–29} Furthermore, neutrophilic airway inflammation has not been consistently shown to be associated with increased eNO.^{28–30} NO₂ can be a marker of motor vehicle emissions. The observed NO₂ effects may be attributed to traffic-related air pollution in the 6 study cities in which local traffic was much heavier than that in a suburban area. This speculation is supported by recent panel studies. Steerenberg et al³⁰ reported a statistically significant association between ambient NO₂ and eNO and concluded that children living in areas with moderate levels of traffic-related air pollution had increased levels of eNO. Adamkiewicz et al¹¹ also reported that exposure to ambient NO₂ among an elderly population living in Steubenville, Ohio was associated with an increase in eNO, although the association was not statistically significant.

We did not observe any statistically significant association between ambient SO₂ and eNO. However, researchers should be cautious when interpreting this. In this study, the daily average concentration was 5.3 ppb. The EPA air quality standards for the SO₂ 24-hour average is 140 ppb. The low levels of ambient SO₂ may have provided a narrow pollution range for this study, thus limiting our ability to fully explore the relation between exposure to ambient SO₂ and eNO.

We observed negative associations between ambient O₃ and eNO. This observation is in line with those reported from a recent study. Adamkiewicz et al¹¹ have observed a negative association (not statistically significant) between 24-hour O₃ exposures and eNO in an elderly population. Several other studies have also confirmed that eNO levels are reduced in habitual smokers as well as passive smokers.³¹ The mechanism for the negative associations between ambient O₃ exposure and eNO is unclear, and little is known about the effect of O₃ exposure on eNO in patients with persistent asthma. However, it is possible for O₃, as a potent oxidant, to react rapidly with NO, yielding the harmful oxidant peroxynitrite.³² This rapid chemical reaction between O₃ and NO could occur both in airways and in the system of the ANORA. The ambient O₃ may also increase breakdown of NO or damage to NO producing epithelial cells.³¹ However, we could not clarify these mechanisms in this study. Last, the inverse association with O₃ is likely to be spurious because of modeling year-round exposure, although our model analyses controlled for season. Ozone is a seasonal pollutant (ie, not present in high levels from November to April in many parts of the country), and previous analyses of ozone and potential health effects have often shown much stronger effects of ozone when the analyses were restricted to the warm months (May through October).

This study is limited in several respects. First, we did not have data on factors that are known or suspected to affect respiratory health. These factors are environmental tobacco smoke, indoor air pollution sources, working environment, and time-activity pattern. In addition, time-activity patterns can vary with pollutant levels (eg, more time outdoors in high-ozone summer months than during the low-ozone winter months). Similarly, there may be greater indoor air pollution exposure during winter months. Nevertheless, the estimated effects are unlikely to be confounded significantly by these factors because they do not vary with daily pollution exposures.³³ Second, exposure misclassification is a major limitation of environmental epidemiologic research and is clearly applicable to this study. This is because

we did not have personal exposure data and because, shared by most panel studies, we used stationary air monitoring data as a surrogate of personal exposure. However, it has been well documented that ambient particles are distributed uniformly within a region, especially extra fine particles but not coarse particles (PM_{10} is a measure of both coarse and fine particles).³⁴ Other researchers have also found that indoor NO_2 correlated highly with outdoor NO_2 .³⁵ Third, time-dependent covariates, aeroallergens (ie, fungi and pollen) and viral respiratory infections, are potential confounders. However, previous studies did not find confounding by outdoor fungi and pollen.^{36–37} In addition, we expect that we can, at least partially, remove such confounding by temporal filtering of the regression model analyses because the levels of aeroallergens generally follow seasonal weather patterns.³⁸ As for the viral respiratory infections, introducing it into the regression model did not substantially alter the associations between pollutants and health outcomes.^{25,39} Nevertheless, we conducted supplement analyses by constricting data analyses on the nonepidemic winter seasons and did not find any significant change of the estimated effects.

Conclusions

Ambient air pollution levels below the current US National Ambient Air Quality Standard may still enhance airway inflammation among patients with persistent asthma, even on typical asthma medications.

Acknowledgments

This study was supported by grants U10 HL-51810, U10 HL-51834, U10 HL-51831, U10 HL-51823, U10 HL-51845, U10 HL-51843, U10 HL-56443, and M01 RR-03186 from the NHLBI. The authors acknowledge the assistance and support from ACRN Steering Committee, Clinical coordinators and technical personnel, Data Coordinating Center personnel, and Protocol Review Committee.

References

1. Rabinovitch N, Strand M, Gelfand EW. Particulate levels are associated with early asthma worsening in children with persistent disease. *Am J Respir Crit Care Med*. 2006; 173:1098–1105. [PubMed: 16484676]
2. Cookson WOCM, Moffatt MF. Asthma: an epidemic in the absence of infection? *Science*. 1997; 275:41–42. [PubMed: 8999535]
3. Schildcrout JS, Sheppard L, Lumley T, Slaughter JC, Koenig JQ, Shapiro GG. Ambient air pollution and asthma exacerbations in children: an 8-city analysis. *Am J Epidemiol*. 2006; 164:505–517. [PubMed: 16798793]
4. Peters JM, Avol E, Gauderman WJ, et al. A study of 12 southern California communities with differing levels and types of air pollution. II. Effects on pulmonary function. *Am J Respir Crit Care Med*. 1999; 159:768–775. [PubMed: 10051249]
5. Pikhart H, Bobak M, Kriz B, et al. Outdoor air concentrations of nitrogen dioxide and sulfur dioxide and prevalence of wheezing in school children. *Epidemiology*. 1999; 11:153–160. [PubMed: 11021612]
6. Desqueyroux H, Pujet JC, Prosper M, et al. Short-term effects of low-level air pollution on respiratory health of adults suffering from moderate to severe asthma. *Environ Res*. 2002; 89:29–37. [PubMed: 12051782]
7. Gilliland FD, Berhane K, Rappaport EB, et al. The effects of ambient air pollution on school absenteeism due to respiratory illnesses. *Epidemiology*. 2001; 12:43–54. [PubMed: 11138819]
8. Moncada S, Higgs A. The L-arginine-nitric oxide pathway. *N Engl J Med*. 1993; 329:2002–2012. [PubMed: 7504210]
9. Nathan C, Xie QW. Nitric oxide synthases: roles, tolls and controls. *Cell*. 1994; 78:915–918. [PubMed: 7522969]
10. Barnes PJ, Liew FY. Nitric oxide and asthmatic inflammation. *Immunol Today*. 1995; 16:128–130. [PubMed: 7536418]

11. Adamkiewicz G, Ebelt S, Syring M, et al. Association between air pollution exposure and exhaled nitric oxide in an elderly population. *Thorax*. 2004; 59:204–209. [PubMed: 14985553]
12. Delfino RJ, Staimer N, Gillen D, et al. Personal and ambient air pollution is associated with increased exhaled nitric oxide in children with asthma. *Environ Health Perspect*. 2006; 114:1736–1743. [PubMed: 17107861]
13. Koenig JQ, Mar TF, Allen RW, et al. Pulmonary effects of indoor- and outdoor-generated particles in children with asthma. *Environ Health Perspect*. 2005; 113:499–503. [PubMed: 15811822]
14. Koenig JQ, Jansen K, Mar TF, et al. Measurement of offline exhaled nitric oxide in a study of community exposure to air pollution. *Environ Health Perspect*. 2003; 111:1625–1629. [PubMed: 14527842]
15. Mar TF, Jansen K, Shepherd K, et al. Exhaled nitric oxide in children with asthma and short-term PM_{2.5} exposure in Seattle. *Environ Health Perspect*. 2005; 113:1791–1794. [PubMed: 16330366]
16. Chinchilli VM, Drazen J, Fish J, et al. The clinical trials in the initial 5-year award period of the Asthma Clinical Research Network. *Control Clin Trials*. 2001; 22(suppl):126S–134S. [PubMed: 11728619]
17. Lemanske J, Sorkness C, Mauger E, et al. Inhaled corticosteroid reduction and elimination in patients with persistent asthma receiving salmeterol: a randomized controlled trial. *JAMA*. 2001; 285:2594–2603. [PubMed: 11368733]
18. Lazarus S, Boushey H, Fahy J, et al. Long-acting β_2 -agonist monotherapy vs continued therapy with inhaled corticosteroids in patients with persistent asthma: a randomized controlled trial. *JAMA*. 2001; 285:2583–2593. [PubMed: 11368732]
19. Silkoff PE, McClean PA, Slutsky AS, et al. Marked flow-dependence of exhaled nitric oxide using a new technique to exclude nasal nitric oxide. *Am J Respir Crit Care Med*. 1997; 155:260–267. [PubMed: 9001322]
20. U.S. Environmental Protection Agency(EPA). Aerometric Information Retrieval System. Vol. 2. Research Triangle Park, NC: US Environmental Protection Agency, Office of Air Quality Planning and Standards; 1993.
21. Jennrich RI, Schluchter MD. Unbalanced repeated-measures models with structured covariance matrices. *Biometrics*. 1986; 42:805–820. [PubMed: 3814725]
22. Schwartz J, Dockery DW, Neas LM. Is daily mortality associated specificity with fine particles? *J Air Waste Manag Assoc*. 1996; 46:927–939. [PubMed: 8875828]
23. U.S. Environmental Protection Agency (EPA). National Air Quality and Emissions Trends Report. Research Triangle Park, NC: US Environmental Protection Agency, Office of Air Quality Planning and Standards; 1990.
24. Hiltermann TJN, Stolk J, van der Zee SC, et al. Asthma severity and susceptibility to air pollution. *Eur Respir J*. 1998; 11:686–693. [PubMed: 9596122]
25. Peters A, Dockery DW, Heinrich J, et al. Medication use modifies the health effects of particulate sulfate air pollution in children with asthma. *Environ Health Perspect*. 1997; 105:430–435. [PubMed: 9189709]
26. Spitzer WO, Suissa S, Ernst P, et al. The use of beta-agonists and the risk of death and near death from asthma. *N Engl J Med*. 1992; 326:501–506. [PubMed: 1346340]
27. McIvor RA, Pizzichini E, Turner MO, et al. Potential masking effects of salmeterol on airway inflammation in asthma. *Am J Respir Crit Care Med*. 1998; 158:924–930. [PubMed: 9731027]
28. Bascom R, Bromberg P, Costa D, et al. Part 2. State of the art review: health effects of outdoor air pollution. *Am J Respir Crit Care Med*. 1996; 153:477–498. [PubMed: 8564086]
29. Blomberg A, Krishna MT, Helleday R, et al. Persistent airway inflammation but accommodated antioxidant and lung function responses after repeated daily exposure to nitrogen dioxide. *Am J Respir Crit Care Med*. 1999; 159:536–543. [PubMed: 9927370]
30. Steerenberg PA, Nierkens S, Fischer PH, et al. Traffic-related air pollution affects peak expiratory flow, exhaled nitric oxide, and inflammatory nasal markers. *Arch Environ Health*. 2001; 56:167–174. [PubMed: 11339681]
31. Yates DH, Breen H, Thomas PS. Passive smoke inhalation decreases exhaled nitric oxide in normal subjects. *Am J Respir Crit Care Med*. 2001; 164:1043–1046. [PubMed: 11587994]

32. Anderson R, Theron AJ, Richards GA, et al. Passive smoking by humans sensitizes circulating neutrophils. *Am Rev Respir Dis*. 1991; 144:570–574. [PubMed: 1892296]
33. Dockery DW, Brunekreef B. Longitudinal studies of air pollution effects on lung function. *Am J Respir Crit Care Med*. 1996; 154(suppl):S250–256. [PubMed: 8970397]
34. Janssen NAH, Hoek G, Brunekreef B, et al. Personal sampling of particles in adults: relation among personal, indoor, and outdoor air concentrations. *Am J Epidemiol*. 1998; 147:537–547. [PubMed: 9521180]
35. Rutishauser M, Ackermann U, Braun C, et al. Significant associations between outdoor NO₂ and respiratory symptoms in preschool children. *Lung*. 1990; 168(suppl):347–352. [PubMed: 2117135]
36. Delfino RJ, Coate B, Zeiger RS, et al. Daily asthma severity in relation to personal ozone exposure and outdoor fungal spores. *Am J Respir Crit Care Med*. 1996; 154:633–641. [PubMed: 8810598]
37. Anderson HR, Ponce de Leon A, Bland JM, et al. Air pollution, pollens, and daily admissions for asthma in London 1987–1992. *Thorax*. 1998; 53:842–848. [PubMed: 10193370]
38. Lewis WH, Dixit AB, Ward WA. Distribution and incidence of North American pollen aeroallergens. *Am J Otolaryngol*. 1991; 12:205–226. [PubMed: 1767872]
39. Neukirch F, Segala C, Moullec YL, et al. Short-term effects of low-level winter pollution on respiratory health of asthmatic adults. *Arch Environ Health*. 1998; 53:320–328. [PubMed: 9766476]

Table 1
Participant Characteristics and Their Relation With the Exhaled Nitric Oxide During 16 Weeks of Active Medication Treatment, February 1997 to January 1999

Characteristic	Participants			Samples		eNO measurements (ppb)							
	N	%	N	%	Total	Triamcinolone		Salmeterol		Placebo		M	SD
						M	SD	M	SD	M	SD		
All subjects	119	100	480	100	23.3	16.5	19.4	12.3	25.6	19.7	25.3	16.3	
Sex													
Male	37	31	147	31	20.4	20.0	12.0	6.6	24.5	25.8	23.5	18.7	
Female	82	69	333	69	24.7	14.5	22.1	12.8	26.3	15.2	26.0	15.3	
Age													
<20	16	13	47	10	22.6	9.3	22.5	10.1	23.1	7.3	21.8	12.6	
20 to 40	78	66	338	70	23.8	15.6	20.4	12.7	24.2	15.9	26.6	17.2	
>40	25	21	95	20	22.1	21.5	15.2	10.5	32.0	32.0	20.3	12.0	
Minority													
Yes	38	32	152	32	24.5	19.5	19.0	11.9	34.7	27.4	21.3	13.3	
No	81	68	328	68	22.8	14.9	19.6	12.5	21.6	13.3	26.9	17.2	
Study center													
Boston, MA	25	21	106	22	13.9	7.2	11.6	7.3	13.8	7.0	16.0	6.2	
Denver, CO	29	24	130	27	30.5	22.2	27.1	15.1	29.2	29.1	34.9	20.4	
Madison, WI	19	16	41	9	22.0	13.8	14.3	9.0	23.6	13.5	25.7	15.1	
Philadelphia, PA	20	17	96	20	22.5	11.5	20.0	8.4	27.6	14.1	18.6	8.1	
San Francisco, CA	23	19	103	21	25.4	15.0	19.4	11.0	29.0	16.0	29.2	16.4	
New York, NY	3	3	4	1	22.3	17.8	14.9	11.5	12.3	—	47.0	—	

Table 2
Pollution and Meteorological Variables, 0-Day Lag for the Daily Concentrations of Pollutants During 16 Weeks of Active Treatment for Asthmatics, by Measures of eNO

Measure	N	M	SD	Min	Max	Percentile		
						25th	50th	75th
Total Samples								
NO ₂ (ppb)	441	23.6	8.0	3.9	48.1	18.2	23.7	28.8
PM ₁₀ (μg/m ³)	282	27.3	12.9	6.1	81.5	18.0	24.9	34.4
O ₃ (ppb)	480	33.6	18.4	1.6	91.5	21.1	30.6	44.4
SO ₂ (ppb)	471	5.3	4.4	1.0	27.2	1.7	4.2	7.6
Temperature (°F)	480	52.9	15.3	14.8	85.4	40.4	54.6	64.3
Relative humidity (%)	480	70.2	17.0	19.3	100.0	57.5	72.1	83.8
Triamcinolone								
NO ₂ (ppb)	147	23.1	8.2	5.3	41.6	17.6	23.9	28.6
PM ₁₀ (μg/m ³)	90	27.5	13.7	7.5	81.5	17.5	25.4	34.5
O ₃ (ppb)	166	32.3	17.7	2.4	89.2	20.6	29.7	39.7
SO ₂ (ppb)	155	5.0	4.0	1.0	20.6	1.9	3.9	7.1
Temperature (°F)	166	51.0	14.9	14.8	81.2	38.0	53.0	60.1
Relative humidity (%)	166	71.4	15.9	27.2	100.0	59.6	72.7	85.1
Salmeterol								
NO ₂ (ppb)	147	24.0	7.8	3.9	48.1	18.4	23.8	29.2
PM ₁₀ (μg/m ³)	99	26.9	11.4	7.2	56.6	18.0	26.6	34.5
O ₃ (ppb)	149	33.9	19.2	2.0	91.5	21.4	29.6	45.9
SO ₂ (ppb)	151	5.6	4.8	1.0	24.8	1.7	4.4	8.7
Temperature (°F)	149	54.2	15.6	14.8	83.6	42.4	56.0	65.7
Relative humidity (%)	149	70.3	17.1	25.1	100.0	57.2	74.6	83.7
Placebo								
NO ₂ (ppb)	147	23.8	8.1	3.9	43.6	18.6	23.4	27.9
PM ₁₀ (μg/m ³)	93	27.4	13.6	6.1	81.5	18.2	24.4	32.7
O ₃ (ppb)	165	34.5	18.4	1.6	86.3	20.9	31.9	45.4

Measure	N	M	SD	Min	Max	Percentile		
						25th	50th	75th
SO ₂ (ppb)	165	5.4	4.3	1.0	27.2	1.7	4.4	8.0
Temperature (°F)	165	53.8	15.5	19.2	85.4	40.6	56.0	66.8
Relative humidity (%)	165	69.0	17.8	19.3	100.0	57.2	71.1	80.9

Table 3
Exposure Effect Estimates and 95% Confidence Intervals (CIs) on eNO, Single Pollutant Models

Pollutant and lag ^b	Change in exhaled NO (ppb) for a 10-unit change in pollutant measures ^a					
	Total		Triamcinolone		Salmeterol	
	??	CI	??	CI	??	CI
NO ₂						
0	0.13	0.06, 0.19 [*]	0.17	0.06, 0.27 [*]	0.11	0.01, 0.22 [*]
1	0.08	0.02, 0.14 [*]	0.09	0.00, 0.18 [†]	0.11	0.01, 0.21 [*]
2	0.00	-0.06, 0.07	0.03	-0.08, 0.13	0.05	-0.06, 0.15
3	-0.03	-0.09, 0.04	-0.01	-0.11, 0.09	0.00	-0.11, 0.11
0-3	0.11	0.01, 0.20 [*]	0.15	0.01, 0.30 [*]	0.15	0.00, 0.30 [†]
PM ₁₀						
0	0.07	0.02, 0.12 [*]	0.05	-0.03, 0.12	0.09	0.00, 0.18 [†]
1	0.02	-0.02, 0.07	0.05	-0.02, 0.13	0.10	0.01, 0.19 [*]
2	0.01	-0.04, 0.06	0.00	-0.07, 0.08	0.04	-0.05, 0.13
3	0.00	-0.04, 0.05	0.01	-0.06, 0.07	0.00	-0.08, 0.08
0-3	0.02	-0.04, 0.07	0.05	-0.04, 0.14	0.04	-0.05, 0.14
O ₃						
0	-0.09	-0.13, -0.05 [*]	-0.05	-0.11, 0.01	-0.11	-0.17, -0.05 [*]
1	0.00	-0.04, 0.04	0.01	-0.04, 0.07	0.00	-0.06, 0.06
2	-0.05	-0.09, -0.01 [*]	-0.03	-0.10, 0.03	-0.06	-0.12, -0.01
3	-0.02	-0.06, 0.02	0.00	-0.06, 0.06	-0.02	-0.08, 0.04
0-3	-0.08	-0.14, -0.02 [*]	-0.05	-0.12, 0.03	-0.10	-0.17, -0.02 [*]
SO ₂						
0	0.09	-0.07, 0.25	0.17	-0.11, 0.44	0.04	-0.18, 0.27
1	0.07	-0.09, 0.23	0.14	-0.12, 0.40	0.17	-0.05, 0.40
2	-0.02	-0.15, 0.11	0.07	-0.13, 0.27	0.00	-0.22, 0.22
3	0.01	-0.13, 0.15	0.08	-0.14, 0.30	-0.06	-0.30, 0.18
0-3	0.07	-0.12, 0.26	0.15	-0.13, 0.43	0.10	-0.19, 0.38

^aControlling for age, sex, race/ethnicity, asthma clinical center, season, week, daily average temperature, and daily average relative humidity.

^bLag 0 = lag of 0 day, lag 1 = lag of 1 day, lag 2 = lag of 2 days, lag 3 = lag of 3 days, and lag 0-3 = lag of 4 days (lag 0, 1, 2, and 3).

* $p < .05$.

[†] $p < .07$.

Table 4
Effect Estimates and 95% Confidence Intervals (CIs) on Exhaled NO, 2 Pollutant Models, 0-Day Lag

Pollutant and co-pollutant	Change in exhaled NO (ppb) for a 10-unit change in pollutant measures ^d											
	Total			Triamcinolone			Salmeterol			Placebo		
	??	CI	??	??	CI	??	??	CI	??	??	CI	??
NO ₂												
PM ₁₀	0.08	-0.01, 0.17	0.13	-0.03, 0.28	0.04	-0.09, 0.17	0.09	-0.05, 0.24				
O ₃	0.11	0.05, 0.18*	0.16	0.06, 0.26*	0.08	-0.03, 0.19	0.10	-0.01, 0.20 [†]				
SO ₂	0.14	0.06, 0.22*	0.18	0.06, 0.30*	0.10	-0.03, 0.23	0.13	0.00, 0.24*				
PM ₁₀												
O ₃	0.09	0.04, 0.14*	0.06	-0.02, 0.13	0.10	0.00, 0.20 [†]	0.11	0.03, 0.19*				
NO ₂	0.05	-0.01, 0.11	0.02	-0.06, 0.11	0.05	-0.05, 0.15	0.07	-0.01, 0.16				
SO ₂	0.06	0.00, 0.12*	0.05	-0.04, 0.14	0.05	-0.05, 0.15	0.08	-0.01, 0.17*				
O ₃												
PM ₁₀	-0.08	-0.13, -0.02*	-0.04	-0.11, 0.03	-0.13	-0.20, -0.06*	-0.04	-0.13, 0.04				
NO ₂	-0.06	-0.10, -0.02*	-0.02	-0.08, 0.04	-0.10	-0.15, 0.05*	-0.04	-0.10, 0.02				
SO ₂	-0.09	-0.13, -0.04*	-0.05	-0.11, 0.02	-0.07	-0.13, -0.01*	-0.15	-0.22, -0.08*				
SO ₂												
PM ₁₀	0.16	-0.08, 0.40	0.26	-0.19, 0.71	0.04	-0.27, 0.36	0.26	-0.12, 0.65				
O ₃	0.05	-0.12, 0.22	0.16	-0.12, 0.44	-0.04	-0.30, 0.23	0.06	-0.20, 0.31				
NO ₂	0.00	-0.18, 0.18	0.07	-0.22, 0.36	-0.05	-0.28, 0.18	0.03	-0.23, 0.28				

^dControlling for age, sex, race/ethnicity, asthma clinical center, season, week, daily average temperature, and daily average relative humidity.

* $p < .05$.

[†] $p < .07$.