Respiratory Medicine (2011) 105, 869–876

available at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/rmed

Prenatal exposure to polycyclic aromatic hydrocarbons, environmental tobacco smoke and asthma

Maria José Rosa [a,b], Kyung Hwa Jung [a], Matthew S. Perzanowski [b], Elizabeth A. Kelvin [c], Katherine W. Darling [a], David E. Camann [e], Steven N. Chillrud [d], Robin M. Whyatt [b], Patrick L. Kinney [b], Frederica P. Perera [a,b], Rachel L. Miller [a,b,f,*]

[a] Division of Pulmonary, Allergy, Critical Care Medicine, Department of Medicine, Columbia University College of Physicians and Surgeons, NY, USA
[b] Department of Environmental Health Sciences, Mailman School of Public Health, Columbia University, NY, USA
[c] CUNY School of Public Health at Hunter College, NY, USA
[d] Lamont-Doherty Earth Observatory, Columbia University, Palisades, NY, USA
[e] Southwest Research Institute, San Antonio, TX, USA
[f] Department of Pediatrics, Columbia University College of Physicians and Surgeons, NY, USA

Received 11 August 2010; accepted 19 November 2010
Available online 15 December 2010

KEYWORDS
Asthma;
Polycyclic aromatic hydrocarbons;
Environmental tobacco smoke;
Atopy

Summary
Background: Previously, we reported that prenatal exposures to polycyclic aromatic hydrocarbons (PAH) and postnatal environmental tobacco smoke (ETS) in combination were associated with respiratory symptoms at ages 1 and 2 years. Here, we hypothesized that children exposed to both prenatal PAH and ETS may be at greater risk of asthma and seroatopy at ages 5–6 years, after controlling for current pollution exposure.
Methods: Prenatal PAH exposure was measured by personal air monitoring over 48 h. ETS exposure, respiratory symptoms and asthma at ages 5–6 years were assessed through questionnaire. Immunoglobulin (Ig) E was measured by Immunocap.

Abbreviations: BRQ, brief respiratory questionnaire; CCCEH, Columbia Center for Children's environmental health; DEP, diesel exhaust particles; ER, emergency room; ETS, environmental tobacco smoke; ISAAC, international study of asthma and allergies in childhood; NYC, New York city; PAH, polycyclic aromatic hydrocarbons; PM, particulate matter; PUF, polyurethane foam; ROS, reactive oxygen species.

* Corresponding author. Division of Pulmonary, Allergy, Critical Care Medicine, Department of Medicine, Columbia University College of Physicians and Surgeons, PH8E, 630 W. 168th St, NY 10032, USA. Tel.: +1 212 305 7759; fax: +1 212 305 2277.
E-mail addresses: mr2805@columbia.edu (M.J. Rosa), kj2237@columbia.edu (K.H. Jung), mp2217@columbia.edu (M.S. Perzanowski), kelvine@pi.cpmc.columbia.edu (E.A. Kelvin), katherine.darling@uscf.edu (K.W. Darling), dcamann@swri.org (D.E. Camann), chilli@ldeo.columbia.edu (S.N. Chillrud), rmw5@columbia.edu (R.M. Whyatt), plk3@columbia.edu (P.L. Kinney), fpp1@columbia.edu (F.P. Perera), rlm14@columbia.edu (R.L. Miller).

0954-6111/$ - see front matter
doi:10.1016/j.rmed.2010.11.022

Results: A significant interaction between prenatal PAH and prenatal (but not postnatal) ETS exposure on asthma ($p < 0.05$), but not IgE, was detected. Among children exposed to prenatal ETS, a positive nonsignificant association was found between prenatal PAH exposure and asthma (OR 1.96, 95% CI [0.95—4.05]). Among children without exposure to prenatal ETS, a negative nonsignificant association was found between prenatal PAH exposure and asthma (OR 0.65, 95% CI [0.41—1.01]). Prenatal PAH exposure was not associated with asthma or IgE at age 5—6 years.

Conclusions: Combined prenatal exposure to PAH and ETS appears to be associated with asthma but not seroatopy at age 5—6. Exposure to PAH alone does not appear associated with either asthma or seroatopy at age 5—6 years. Discerning the differential effects between ETS exposed and ETS nonexposed children requires further study.

Introduction

Exposure to diesel exhaust particles (DEP) has been associated with greater morbidity and mortality due to respiratory illnesses.[1–5] DEP consists of a carbonaceous core onto which metals and other chemicals such as polycyclic aromatic hydrocarbons (PAHs), heterocyclics, quinones, aldehydes, and aliphatic hydrocarbons are adsorbed.[6] The effects of these individual components on respiratory illness remain understudied. However, studies suggest that exposure to PAH in particular, emitted from traffic sources, cigarette smoking, cooking and space heating, may be associated with the development and exacerbation of asthma and seroatopy.[7,8] Previously, our group at the Columbia Center for Children's Environmental Health (CCCEH) reported significant interactions between PAH exposure measured prenatally and early postnatal exposure to environmental tobacco smoke (ETS) on the outcomes of cough and wheeze at age 1 year and difficulty breathing and probable asthma at age 2 years in children in NYC.[9] Similar results were reported in Poland, where PAH levels were several times higher than the ones measured in NYC.[10]

Despite these reports, the importance of prenatal PAH exposure, independently and in association with ETS, on persistent wheeze and asthma still needs to be ascertained. Early wheeze and respiratory symptoms may be transient and not necessarily be associated with persistent wheeze or asthma at later ages.[11] Associations of prenatal PAH and ETS exposure with asthma also would need to consider the effect of later postnatal exposures that may correlate with prenatal levels. We hypothesized that children exposed to higher levels of PAH prenatally combined with ETS (either prenatal or postnatal) may be at greater risk of wheeze, asthma and seroatopy at ages 5—6 years, even after controlling for current exposure to PAH and ETS. Our strategy, using a longitudinal design based within the CCCEH birth cohort, was to examine the association of repeat measures of PAH and ETS exposure with symptoms ascertained when the children reached 5 through 6 years of age.

Methods

Study population

Briefly, 725 nonsmoking, healthy women who self-identified as Dominican ethnicity or African American race and living in Northern Manhattan or the South Bronx were enrolled during pregnancy and their children followed prospectively.[12–14] Questionnaires were administered prenatally, and at ages one, two, three and 5—6 years. Data were analyzed for those children ($n = 290$) for whom airborne PAH measures at two time points (prenatal, age 5—6 years) and complete questionnaire data were available. The study was approved by the Columbia University Institutional Review Board.

PAH exposure

During the prenatal period, personal air monitoring over 48 h was conducted during the third trimester as described.[9] Personal pumps ran continuously and collected particles up to 2.5 μm in diameter on a quartz microfiber filter and gas phase PAH were collected on a polyurethane foam (PUF) sampler. At age 5 through 6, a two-week home-based indoor monitoring was used to determine PAH exposure as described.[15] Briefly, monitors were set up in the room where the child spent most of his/her time at a height of about 1.2 m and at least 0.3 m from the walls. Particles and gas phase PAH collected from the first two-week monitoring period were extracted together. Filters from both time points were analyzed for 9 PAH compounds: benz(a)anthracene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(ghi)perylene, benzo(a)pyrene, chrysene/isocrysene, dibenz(a,h)anthracene, indeno(1,2,3) pyrene and pyrene at the Southwest Research Institute. Anthracene-d_{10} and p-terphenyl-d_{14} were used as surrogate standards for recovery and chrysen-d_{12} and perylene-d_{12} were used as internal standards for quantification. Data were flagged if any monitoring problems occurred (i.e. power outages, flow rate changes, tube disconnection, pump failure, etc) and additional analysis flags were assigned to PAH compounds below the limit of detection (LOD), elevated peaks due to presence of an interference peak, or questionable peaks. Flagged samples were excluded from statistical analysis based on a previously established protocol.[15]

ETS exposure

Prenatal ETS exposure among children of mothers who did not smoke during pregnancy was defined as the report of any smoker in the home during pregnancy. Postnatal ETS

was defined as the report of any smoking household member when the child was aged 6 months, 1, 2, 3 or 5 years. Plasma cotinine was measured in cord blood and at ages 2, 3 and 5 years at the Center for Disease Control and Prevention and used to validate data obtained from questionnaire. Chi-square tests for trend were run to validate the association between plasma cotinine levels from cord blood or ages 2 and 3 years (insufficient sample size for age 5) and report of ETS in the home at the corresponding age. Analyses were repeated examining report of ETS in the home or at work. Cotinine levels were split into quartiles and the lowest quartile included those cotinine levels below limit of detection (LOD). The association between ETS exposure and cotinine levels was highly significant at all ages (χ^2 for trend for prenatal ETS, ETS at age 2 years, ETS at age 3 years, $p < 0.001$ for each). Similar results were found when ETS exposure was defined as either report of smoker at home or at work for prenatal ETS, and report of a smoker at home or outside smoke exposure for postnatal ETS at ages 2 and 3 years (data not shown).

Questionnaires and IgE

The Brief Respiratory Questionnaires (BRQ), validated in a similar local urban population,[16] and the International Study of Asthma and Allergies in Childhood questionnaires (ISAAC), validated internationally,[17] were administered at both ages 5 and 6 years. The BRQ was used to assess report of doctor diagnosis of asthma, emergency room visits due to breathing problems, and use of asthma medications in the past 12 months at either age. The ISAAC questionnaire was used to assess current wheeze, defined as any report of wheeze in the past 12 months, and nighttime wheeze, defined as sleep disturbed by wheeze at least one night a week in the past 12 months at either age 5 or 6 years.

Total serum IgE and specific IgE to cat, mouse, *Dermatophagoides* farinae and German cockroach at age 5 years were measured in duplicate using Immunocap (Phadia, Uppsala, Sweden).[18] Children were considered seroatopic if they had a specific IgE $\geq$0.35 IU/mL to any of the indoor allergens tested. Endotoxin was measured in bedroom floor dust collected at age 1 or 3 years.[14]

Statistical analyses

Total gas and particle PAH levels were analyzed as the sum of eight individual PAH as described.[9] PAH levels also were analyzed as the sum of individual PAH with the exclusion of chrysene, an ETS-derived PAH. Because airborne pyrene levels were not well-correlated to the other 8 PAH measured, pyrene levels were analyzed in separate models. Total sum of 8 PAH, individual PAH measures and IgE concentrations were natural log transformed due to non-normal distribution.

Maternal education, dichotomized on completion of high school at the time of the child's birth, was used as a proxy for socioeconomic status. From the BRQ questionnaire, report of the number of emergency room (ER) visits due to asthma, wheezing, cough bronchitis or other breathing problems was dichotomized as zero versus any ER visits. To reduce the number of tests performed and to increase sample size, age 5 and age 6 respiratory data were combined and a positive outcome was defined as report of that outcome at either age 5 or age 6 years. Chi-square tests were run to test the association between report of symptoms at age 5 or age 6 years.

Respiratory and specific IgE outcomes were analyzed using logistic regression models. Total IgE levels were analyzed using a linear regression model. Data were analyzed using SPSS Version 17 (Chicago, IL). Interactions between individual PAH and report of prenatal ETS and postnatal ETS also were analyzed by logistic regression. All logistic regression models included maternal education, maternal asthma, race/ethnicity and sex as covariates.

Results

The demographic characteristics of the cohort are summarized in Table 1. Results for the interaction between prenatal PAH and prenatal ETS exposure, as well as main effects of prenatal and postnatal PAH and ETS exposure, are presented in Table 2. A significant interaction between prenatal PAH levels and report of prenatal ETS exposure on asthma at age 5 or 6 years was found ($p = 0.02$). The interaction approached significance for report of any visit to the emergency room due to breathing problems at age 5 or 6 years (Table 2). Despite the strong association between

Table 1 Demographics of cohort subset included in these analyses.

Male sex, n (%)	130/290 (44.8)
Ethnicity/Race	
Dominican, n (%)	178/290 (61.4)
African American, n (%)	112/290 (38.6)
Mother not completed high school, n (%)	109/290 (37.6)
Any report of maternal asthma, n (%)	75/290 (25.9)
Child current wheeze, n (%)	98/290 (33.8)
Doctor diagnosed asthma, n (%)	98/290 (33.8)
Nighttime wheeze, n (%)	68/290 (23.4)
Asthma medication use, n (%)	96/290 (33.1)
ER visits, n (%)	84/290 (29.0)
Child total IgE$\geq$ 50 IU/mL	91/201 (45.3)
At least one SIgE$\geq$ 0.35 IU/mL	63/208 (30.3)
Prenatal ETS	93/290 (33.8)
Postnatal ETS	123/290 (42.4)
Prenatal PAH median(25th—75th)	2.28 (1.43—3.64)
Postnatal PAH median(25th—75th)	1.33 (0.81—2.24)

Ethnicity/race, maternal education and history of maternal asthma determined at the time of mother's enrollment. Doctor diagnosed asthma is defined as maternal report of doctor's diagnosis of asthma at ages 5 or 6 years. Current wheeze is defined as wheeze reported by mother/caretaker in the past 12 months using ISAAC questionnaire at age 5 or 6. Asthma medication use defined as report of asthma medication use in the past 12 months by BRQ at age 5 or 6 years. Emergency room visits are defined as at least one visit to the ER in the past 12 months due to breathing problems by BRQ at age 5 or 6 years. Nighttime wheeze are defined as sleep disturbed by wheeze at least one time a week in the past 12 months by ISAAC questionnaire at age 5 or 6 years.

Table 2 Association of PAH and ETS exposure with wheeze and asthma, at age 5 or 6 years.

	Total sample main effect	Model with addition of prenatal PAH*prenatal ETS interaction term
	OR (95% CI)	*p*-value for interaction
Doctor diagnosed asthma		
Prenatal PAH	0.88 (0.63—1.24)	0.020
Prenatal ETS	0.84 (0.44—1.61)	
Postnatal PAH	0.83 (0.60—1.15)	
Postnatal ETS	0.98 (0.53—1.84)	
Current wheeze		
Prenatal PAH	1.04 (0.74—1.45)	0.562
Prenatal ETS	1.19 (0.63—2.28)	
Postnatal PAH	0.72 (0.52—1.01)	
Postnatal ETS	1.26 (0.67—2.34)	
Asthma medication use		
Prenatal PAH	1.12 (0.80—1.56)	0.323
Prenatal ETS	1.06 (0.55—2.04)	
Postnatal PAH	0.66 (0.46—0.93)*	
Postnatal ETS	1.09 (0.58—2.05)	
ER visits		
Prenatal PAH	0.77 (0.53—1.12)	0.083
Prenatal ETS	0.82 (0.42—1.62)	
Postnatal PAH	0.75 (0.53—1.07)	
Postnatal ETS	0.99 (0.52—1.89)	
Nighttime wheeze		
Prenatal PAH	1.02 (0.71—1.49)	0.152
Prenatal ETS	1.20 (0.59—2.46)	
Postnatal PAH	0.78 (0.53—1.12)	
Postnatal ETS	0.80 (0.40—1.60)	

Models controlling for maternal asthma, maternal education, race/ethnicity and sex. Prenatal PAH refers to measurements collected from personal air monitor during the third trimester of pregnancy. Postnatal PAH refers to measurements collected from residential air monitor when child was aged 5—6 years old. Postnatal ETS is any reported ETS exposure from birth to age 5. Doctor diagnosed asthma, current wheeze, asthma medication, ER visits, nighttime symptoms as defined in Table 1. *p-value$<$0.05.

prenatal ETS and postnatal ETS (age 6 months, 1 year, 2 years, 3 years and 5 years), similar interactions were not replicated when examining the effects of exposure to prenatal PAH and postnatal ETS on any respiratory outcomes at age 5 or 6 years. Also, these results were not replicated when the sum of 8 PAH was replaced by pyrene in the model.

To determine the direction of these interactions, the adjusted models were stratified by the presence or absence of prenatal ETS exposure. Among those exposed to ETS prenatally, a nonsignificant positive association with prenatal PAH exposure was detected for reported asthma at age 5 or 6 years (OR 1.96, 95% CI [0.95—4.05]). Among those not exposed to ETS prenatally, a nonsignificant negative association with prenatal PAH exposure was detected for asthma (OR 0.65 95% CI [0.41—1.01]).

In main effects analyses, prenatal exposure to PAH was not significantly associated with report of asthma or any of the asthma symptoms under study ($p > 0.05$ in all analyses, Table 2). Report of prenatal and postnatal ETS was not associated significantly with any of the respiratory outcomes tested. Postnatal PAH exposure was associated negatively with report of asthma medication use. Logistic regression models that included endotoxin exposure as a covariate also did not reveal significant differences in the associations with respiratory symptoms (results not shown). The same results were also obtained when chrysene (i.e. PAH indicator of ETS) was included and excluded from the sum of PAH logistic regression models (results not shown).

In order to determine whether the interactions or main effects may be differentially attributable to exposure to individual PAH, logistic regression models were run separately with each of the eight individual prenatal PAHs and prenatal ETS. Significant differences in the associations across individual PAH were not detected (Table e1, online supplement).

Both early PAH and ETS exposures have been associated individually with the development of seroatopy.[19,20] To determine whether combined exposure to these pollutants was associated with seroatopy in the CCCEH cohort, prenatal PAH levels and report of ETS exposure were compared to total and specific IgE production at age 5 years. Postnatal ETS

was associated with greater odds of positive anti-mouse IgE (OR 5.29, 95% CI [1.58—17.71]). In contrast, prenatal PAH exposure was associated negatively with anti-*d. farinae* IgE (OR 0.23, 95% CI [0.09—0.62]). Significant interactions between prenatal PAH and prenatal ETS on total and specific IgE levels were absent.

Discussion

These results suggest a significant positive interaction between prenatal PAH and prenatal ETS on report of asthma at ages 5 or 6 years, even after controlling for current exposures. Prenatal PAH exposure alone was not associated with asthma or IgE at age 5 or 6 years. We suspect that the absence of a significant association of prenatal PAH exposure with asthma symptoms among children stratified by parents that reported the presence or absence of prenatal ETS exposure may be due to diminished statistical power. To our knowledge, this is the first study to compare prospectively the effects of prenatal PAH, and its interaction with residential ETS, after controlling for current exposure, on the subsequent report of asthma.

Growing literature both from experimental studies as well as epidemiological work suggests that the prenatal period is a time when lungs and other organs are particularly vulnerable to environmental hazards from exposure to not only PAH and ETS, but also ozone, nitrogen dioxide, carbon monoxide, and particulate matter (PM).[10,21—26] In the case of ETS, exposure to prenatal and early postnatal, but not later postnatal sidestream cigarette smoke, has been shown to increase airway hyperresponsiveness in mice.[21] The National Health and Nutrition Examination Survey (NHANES) clearly identified the risk of prenatal ETS on early wheeze, asthma and chronic bronchitis.[27] In a Polish cohort, children exposed to high PAH levels prenatally had significant higher risks of wheeze, sore throat, cough and barking cough during the first year of life.[10] Murine studies have shown that mice exposed to diesel exhaust particles (DEP) and PM during pregnancy produce offspring with greater airway allergic inflammation and ozone-induced airway hyperresponsiveness.[22,28] Prenatal exposure to PM_{10}, nitrogen dioxide and carbon monoxide also has been associated with decreased pulmonary lung function measures in asthmatics.[29] Several plausible mechanisms may explain these associations. Both PAH and ETS exposures have been associated experimentally with the induction of proinflammatory mediators. Cigarette smoke has been shown to induce mitochondrial production of reactive oxygen species (ROS) in human bronchial epithelial cells, and inhibit vascular endothelial growth factor receptor 2 (VEGFR-2), leading to endothelial cell airway dysfunction, decreased levels of eNOS, cell migration, and angiogenesis.[30,31] ROS can induce airway hyperresponsiveness and inflammation, two hallmarks of asthma, while endothelial airway dysfunction also has been implicated in the pathogenesis of airway disease.[32,33]

While synergistic effects of combined PAH and ETS exposures have not been demonstrated in the past, other models of exposure to air pollution suggest such a paradigm is plausible biologically. These include studies suggesting that rhinovirus infection interacts with the pollutants nitrogen dioxide and ozone to result in enhanced interleukin(IL)-8 release in human epithelial cells.[34] Influenza also has been shown to modify the association between ozone exposure and hospitalizations for respiratory disease.[35] Murine studies have shown increased ozone-induced airway hyperresponsiveness in offspring whose mothers were exposed to particulate matter during pregnancy.[22] Parental stress also has been shown to interact with exposure to traffic-related air pollution (i.e. NO_x) on asthma in humans.[36] Dog ownership (as a proxy for endotoxin exposure) has been associated with worse respiratory symptoms when occurring with greater exposure to air pollutants such as nitrogen dioxide, PM_{10}, $PM_{2.5}$ and ozone.[37]

In this study, we found one significant interaction between prenatal PAH and prenatal ETS exposure on the outcome of doctor diagnosed asthma at ages 5 or 6 years. Also, for children exposed to ETS the odds of doctor diagnosed asthma increased with higher prenatal PAH exposure while the association was negative among the children who were not exposed to ETS prenatally. The interaction with prenatal ETS, and not postnatal ETS, differs from our previous report on the outcomes of cough and wheeze at age 1 year and difficulty breathing and probable asthma at age 2 years.[9] However, in those analyses, only the interaction between prenatal PAH and postnatal ETS was examined, because virtually all mothers who reported prenatal ETS also reported postnatal ETS. In these current analyses, even though all ETS measures were correlated, there was not the same overlap permitting further investigation. Further, this study was able to control for the effects of postnatal PAH exposure on wheeze. These newer findings may reflect the true biological effect of the timing of ETS exposure on the different outcomes under investigation between studies (i.e. early wheeze[9] vs. later or persistent wheeze). This hypothesis also is supported by a recent Polish study that found prenatal exposure to PAH, measured as PAH-DNA adducts, to be associated only with onset wheeze in the first 2 years of life, but not at age 3 or 4 years.[38]

Prenatal PAH exposure alone was not associated with persistent wheeze and asthma at ages 5 or 6 years. There are several reasons why we may have failed to find these associations. First, there may be no effect of prenatal PAH exposure on persistent asthma, or the effect may be so small that we are insufficiently powered to detect it. Also, there may be other unmeasured environmental exposures that may be more important to the development of asthma than PAH. Another possibility is that the association between prenatal PAH exposure and respiratory health may be dependent on the timing of exposure within gestation. A recent study by Herr *et al*. showed that the association between prenatal PAH exposure on elevated cord blood IgE varied by gestational month of exposure.[39] PAH exposure during the first two months of pregnancy was associated with lower cord blood IgE; higher PAH exposure during the fourth through seventh month of pregnancy with elevated cord blood IgE. In our study, all the prenatal monitoring was done during the third trimester limiting such an analysis. During the third trimester, when most (~80%) of the prenatal monitoring was conducted in this study, Herr *et al*. did not observe significant associations between PAH levels and cord blood IgE.

Arguably, the association between prenatal PAH and asthma trends in the negative direction, though without statistical significance. Our analyses also showed a significant negative association between postnatal PAH exposure and the report of asthma medication use, but not current wheeze or ER visits due to respiratory problems, making

these results difficult to interpret. However, there are plausible mechanisms through which these exposures may be associated with decreased airway inflammation. PAH have been shown to induce thymus atrophy and thymocyte apoptosis in a murine model, and have been implicated in the suppression of human T cell mitogenesis.[40,41] Also, there is evidence that PAH exposure can inhibit B cell growth and/or induce pre-B cell apoptosis.[42,43] This latter process may downregulate IgE and other Ig production and these mechanisms could be associated with a decrease in respiratory symptoms.

Genetic variation also has been shown to modify the effect of exposure to pollutants on respiratory outcomes. Previous work has been done on the family of detoxification enzymes, the glutathione S-transferase (GST) genes and tobacco smoke exposure. Breton *et al*. reported a significant interaction between prenatal tobacco smoke exposure and the GSTM2 gene on lung function outcomes in children. In this study, a significant decrease in lung function was detected among children whose mothers smoked during pregnancy and possessed the GSTM2 risk haplotype.[44] Conversely, the null haplotype had a protective effect in the exposed children.[44] Also, a study in Germany reported that children with ETS exposure and GSTM1 deficiency had higher odds of current asthma and asthma symptoms than GSTM1 positive children without ETS exposure.[45] Another recent study also showed that the lack of GSTM1 was associated with a decrease in lung function and early onset asthma among children exposed to prenatal ETS.[46]

We hypothesized that combined prenatal PAH and ETS exposure may be associated with higher IgE levels among children participating in the CCCEH cohort because both PAH[47–51] and ETS[20,52,53] exposures have been associated with the production of T helper (Th) proallergic cytokines or allergic sensitization. In this study, the results were mixed. Postnatal ETS was associated with greater odds of positive anti-mouse IgE while prenatal PAH exposure was associated negatively with anti-cockroach and anti-*d. farinae* IgE. The latter finding may be consistent with our previous findings in mice where combined prenatal exposure to diesel and the mold allergen *Aspergillus fumigatus* was associated with decreased total IgE in the offspring.[54] Indeed, the effect of traffic-related air pollution on IgE appears to vary according to the specific allergen. For example, the PIAMA study found that exposure to $PM_{2.5}$, soot and nitrogen dioxide was associated with sensitization to food allergens, but not with sensitization to house dust mite, cat, dog, Dactylium, birch pollen or Alternaria.[55]

We acknowledge several limitations in the study. While the power was sufficient to detect some PAH by ETS interactions, the statistical power of the stratified analyses was more limited. In order to find statistically significant results with 80% power among these strata, we would need 424 children with prenatal ETS exposure and 1120 children without prenatal ETS exposure, an expensive study beyond the scope of this large cohort study. Therefore, the stratified results should be interpreted with caution. Also, exposure to ETS used in the final analyses was assessed using questionnaires. However, plasma cotinine levels were associated significantly with report of ETS exposure at every age measured, in agreement with our previously published results.[56] Because methods that rely on self-report tend to underreport ETS exposure, any bias would lead to an underestimation of the effects of ETS. Cotinine is also a marker of short term (past 48 h) ETS exposure and may not adequately capture recurrent exposure throughout pregnancy.[57,58] Further, the significant ETS-derived PAH contributions, *i.e*. chrysene, to the total PAH concentrations that were measured cannot be discounted. However, removal of chrysene from the regression models did not change the associations significantly suggesting otherwise.

Another caveat is the difficulty in separating measures of prenatal ETS exposure from postnatal ETS exposure given their strong association (the report of presence or absence of prenatal ETS exposure was the same for 78% of the participants at age 6 months, 82% at age 1 year, 74% at age 2 years, 72% at age 3 years and 74% at 5 years, all χ^2 p-values <0.001 for comparisons). However, significant interactions between postnatal ETS and prenatal PAH exposure on age 5 or 6 asthma-related outcomes were not detected. Finally, the postnatal PAH analyses relied on indoor measurements that may be more representative of indoor rather than traffic-related PAH sources. Outdoor PAH concentrations may represent more accurately the child's personal exposure to traffic-related air pollution. However, previously reported comparisons between indoor and outdoor sampling has shown that several of the PAHs measured indoors are derived primarily from outdoor sources; in particular the higher molecular weight, nonvolatile PAHs.[15]

In conclusion, a significant interaction between prenatal exposures to PAHs and ETS on reported asthma at age 5 or 6 years was detected. There were no significant associations between prenatal PAH exposure and respiratory symptoms or seroatopy at age 5 or 6 years. Further study is required to understand the effect modification of ETS exposure on the association between PAH exposure and respiratory outcomes.

Conflicts of interest

Authors has no conflicts of interest. Study sponsors had no role in the study design, in the collection, analysis and interpretation of data; in the writing of the manuscript; and in the decision to submit the manuscript for publication.

Acknowledgements

This work was performed at Columbia University. This work was supported by "Urban diesel exposure and inner city asthma" [R01 ES013163], "The role of airborne PAHs and DEP in the pathogenesis of childhood asthma" [P50ES015905], "Environmental health in a cohort of minority women/infants" [R01 ES008977], "The Columbia Center for Childrens Environmental Health" [P01ES09600] and the U.S. Environmental Protection Agency [R827027].

Supplementary material

The supplementary data associated with this article can be found in the online version at doi:10.1016/j.rmed.2010.11.022.

References

1. Jaakkol JJK, Vutanen M, Heinonen P. Low-Level air pollution and. *Am J Public Health* 1991;**81**:6—9.
2. Kagawa J. Atmospheric pollution due to mobile sources and effects on human health in Japan. *Environ health perspectives* 1994;**102**(Suppl. 4):93—9.
3. Samet JM, Dominici F, Curriero FC, Coursac I, Zeger SL. Fine particulate air pollution and mortality in 20 U.S. cities, 1987-1994. *N Engl J Med* 2000;**343**(24):1742—9.
4. Morgenstern V, Zutavern A, Cyrys J, Brockow I, Koletzko S, Kramer U, et al. Atopic diseases, allergic sensitization, and exposure to traffic-related air pollution in children. *Am J Respir Crit Care Med* 2008;**177**(12):1331—7.
5. Riedl M, Diaz-Sanchez D. Biology of diesel exhaust effects on respiratory function. *J Allergy Clin Immunol* 2005;**115**(2): 221—8. quiz 9.
6. WHO/IPCS. *Diesel fuel and exhaust emissions. World health Organization/International Programme on chemical Safety*. Environmental Health Criteria 171. Geneva: Switzerland; 1996.
7. Miller RL, Garfinkel R, Lendor C, Hoepner L, Li Z, Romanoff L, et al. Polycyclic aromatic hydrocarbon metabolite levels and pediatric allergy and asthma in an inner-city cohort. *Pediatr Allergy Immunol*; 2009.
8. Leem JH, Kim JH, Lee KH, Hong YC, Lee KH, Kang D, et al. Asthma attack associated with oxidative stress by exposure to ETS and PAH. The Journal of asthma: Official Journal Assoc for Care Asthma. 42: 463—467.
9. Miller RL, Garfinkel R, Horton M, Camann D, Perera FP, Whyatt RM, et al. Polycyclic aromatic hydrocarbons, environmental tobacco smoke, and respiratory symptoms in an inner-city birth cohort. *Chest* 2004;**126**(4):1071—8.
10. Jedrychowski W, Galas A, Pac A, Flak E, Camman D, Rauh V, et al. Prenatal ambient air exposure to polycyclic aromatic hydrocarbons and the occurrence of respiratory symptoms over the first year of life. *Eur J Epidemiol* 2005;**20**(9):775—82.
11. Martinez FD. Development of wheezing disorders and asthma in preschool children. *Pediatrics* 2002;**109**(2 Suppl):362—7.
12. Miller RL, Chew GL, Bell CA, Biedermann SA, Aggarwal M, Kinney PL, et al. Prenatal exposure, maternal sensitization, and sensitization in utero to indoor allergens in an inner-city cohort. *Am J Respir Crit Care Med* 2001;**164**(6):995—1001.
13. Goldstein IF, Perzanowski MS, Lendor C, Garfinkel RS, Hoepner LA, Chew GL, et al. Prevalence of allergy symptoms and total IgE in a New York City cohort and their association with birth order. *Int Arch Allergy Immunol* 2005;**137**(3):249—57.
14. Perzanowski MS, Miller RL, Thorne PS, Barr RG, Divjan A, Sheares BJ, et al. Endotoxin in inner-city homes: associations with wheeze and eczema in early childhood. *J Allergy Clin Immunol* 2006;**117**(5):1082—9.
15. Jung KH, Patel MM, Moors K, Kinney PL, Chillrud SN, Whyatt R, Hoepner L, Garfinkel R, Yan B, Ross J, Camann D, Perera F, Miller RL. Effect of heating season on residential indoor and outdoor polycyclic aromatic hydrocarbons, black carbon, and particulate matter in an urban birth cohort. *Revision Requested Atmos Environ*; 2010.
16. Bonner S, Matte T, Rubin M, Sheares BJ, Fagan JK, Evans D, et al. Validating an asthma case detection instrument in a head start sample. *J Sch Health* 2006;**76**(9):471—8.
17. Jenkins MA, Clarke JR, Carlin JB, Robertson CF, Hopper JL, Dalton MF, et al. Validation of questionnaire and bronchial hyperresponsiveness against respiratory physician assessment in the diagnosis of asthma. *Int J Epidemiol* 1996;**25**(3):609—16.
18. Donohue KM, Al-alem U, Perzanowski MS, Chew GL, Johnson A, Divjan A, et al. Anti-cockroach and anti-mouse IgE are associated with early wheeze and atopy in an inner-city birth cohort. *J Allergy Clin Immunol* 2008;**122**(5):914—20.
19. Kanoh T, Suzuki T, Ishimori M, Ikeda S, Ohasawa M, Ohkuni H, et al. Adjuvant activities of pyrene, anthracene, fluoranthene and benzo(a)pyrene in production of anti-IgE antibody to Japanese cedar pollen allergen in mice. *J Clin Lab Immunol* 1996;**48**(4):133—47.
20. Lannero E, Wickman M, van Hage M, Bergstrom A, Pershagen G, Nordvall L. Exposure to environmental tobacco smoke and sensitisation in children. *Thorax* 2008;**63**(2):172—6.
21. Wu ZX, Hunter DD, Kish VL, Benders KM, Batchelor TP, Dey RD. Prenatal and early, but not late, postnatal exposure of mice to sidestream tobacco smoke increases airway hyperresponsiveness later in life. *Environ Health Perspect* 2009; **117**(9):1434—40.
22. Auten RL, Potts EN, Mason SN, Fischer B, Huang Y, Foster WM. Maternal exposure to particulate matter increases postnatal ozone-induced airway hyperreactivity in juvenile mice. *Am J Respir Crit Care Med* 2009;**180**(12):1218—26.
23. Tielsch JM, Katz J, Thulasiraj RD, Coles CL, Sheeladevi S, Yanik EL, et al. Exposure to indoor biomass fuel and tobacco smoke and risk of adverse reproductive outcomes, mortality, respiratory morbidity and growth among newborn infants in south India. *Int J Epidemiol* 2009;**38**(5):1351—63.
24. Mortimer K, Neugebauer R, Lurmann F, Alcorn S, Balmes J, Tager I. Early-lifetime exposure to air pollution and allergic sensitization in children with asthma. *J Asthma* 2008;**45**(10): 874—81.
25. Wang C, Salam MT, Islam T, Wenten M, Gauderman WJ, Gilliland FD. Effects of in utero and childhood tobacco smoke exposure and beta2-adrenergic receptor genotype on childhood asthma and wheezing. *Pediatrics* 2008;**122**(1):e107—114.
26. Slama R, Morgenstern V, Cyrys J, Zutavern A, Herbarth O, Wichmann HE, et al. Traffic-related atmospheric pollutants levels during pregnancy and offspring's term birth weight: a study relying on a land-use regression exposure model. *Environ Health Perspect* 2007;**115**(9):1283—92.
27. Gergen PJ, Fowler JA, Maurer KR, Davis WW, Overpeck MD. The burden of environmental tobacco smoke exposure on the respiratory health of children 2 months through 5 years of age in the United States: third National Health and Nutrition Examination Survey, 1988 to 1994. *Pediatrics* 1998;**101**(2). E8.
28. Fedulov AV, Leme A, Yang Z, Dahl M, Lim R, Mariani TJ, et al. Pulmonary exposure to particles during pregnancy causes increased neonatal asthma susceptibility. *Am J Respir Cell Mol Biol* 2008;**38**(1):57—67.
29. Mortimer K, Neugebauer R, Lurmann F, Alcorn S, Balmes J, Tager I. Air pollution and pulmonary function in asthmatic children: effects of prenatal and lifetime exposures. *Epidemiology* 2008;**19**(4):550—7. discussion 61—2.
30. van der Toorn M, Rezayat D, Kauffman HF, Bakker SJ, Gans RO, Koeter GH, et al. Lipid-soluble components in cigarette smoke induce mitochondrial production of reactive oxygen species in lung epithelial cells. *Am J Physiol Lung Cell Mol Physiol* 2009; **297**(1):L109—14.
31. Edirisinghe I, Yang SR, Yao H, Rajendrasozhan S, Caito S, Adenuga D, et al. VEGFR-2 inhibition augments cigarette smoke-induced oxidative stress and inflammatory responses leading to endothelial dysfunction. *FASEB J* 2008;**22**(7): 2297—310.
32. Park CS, Kim TB, Lee KY, Moon KA, Bae YJ, Jang MK, et al. Increased oxidative stress in the airway and development of allergic inflammation in a mouse model of asthma. *Ann Allergy Asthma Immunol* 2009;**103**(3):238—47.
33. Burgess JK, Boustany S, Moir LM, Weckmann M, Lau JY, Grafton K, et al. Reduction of tumstatin in asthmatic airways contributes to angiogenesis, inflammation, and hyperresponsiveness. Am J Respir Crit Care Med. **181**(2): 106—115.
34. Spannhake EW, Reddy SP, Jacoby DB, Yu XY, Saatian B, Tian J. Synergism between rhinovirus infection and oxidant pollutant

exposure enhances airway epithelial cell cytokine production. *Environ Health Perspect* 2002;**110**(7):665—70.
35. Chit Ming Wong LY, Thuan Quoc Thach, et al. Modification by influenza on health effects of air pollution in Hong Kong. *Environ Health Perspect* 2009;**117**(2):248—53.
36. Shankardass K, McConnell R, Jerrett M, Milam J, Richardson J, Berhane K. Parental stress increases the effect of traffic-related air pollution on childhood asthma incidence. *Proc Natl Acad Sci U S A* 2009;**106**(30):12406—11.
37. McConnell R, Berhane K, Molitor J, Gilliland F, Kunzli N, Thorne PS, et al. Dog ownership enhances symptomatic responses to air pollution in children with asthma. *Environ Health Perspect* 2006;**114**(12):1910—5.
38. Jedrychowski W. Intrauterine exposure to polycyclic aromatic hydrocarbons, fine particulate matter and early wheeze. Pospective birth cohort study in 4-year olds. *Pediatr Allergy Immunol* 2010;**21**(4):e723—732.
39. Herr G, Skokanova V, et al. Exposure to air pollution in critical prenatal time windows and IgE levels in newborns. *Pediatr Allergy Immunol*; 2010:1—11.
40. Lutz CT, Browne G, Petzold CR. Methylcholanthrene causes increased thymocyte apoptosis. *Toxicology* 1998;**128**(2): 151—67.
41. Davila DR, Romero DL, Burchiel SW. Human T cells are highly sensitive to suppression of mitogenesis by polycyclic aromatic hydrocarbons and this effect is differentially reversed by alpha-naphthoflavone. *Toxicol Appl Pharmacol* 1996;**139**(2): 333—41.
42. Allan LL, Schlezinger JJ, Shansab M, Sherr DH. CYP1A1 in polycyclic aromatic hydrocarbon-induced B lymphocyte growth suppression. *Biochem Biophys Res Commun* 2006;**342**(1): 227—35.
43. Mann KK, Doerre S, Schlezinger JJ, Sherr DH, Quadri S. The role of NF-kappaB as a survival factor in environmental chemical-induced pre-B cell apoptosis. *Mol Pharmacol* 2001;**59**(2): 302—9.
44. Breton CV, Vora H, Salam MT, Islam T, Wenten M, Gauderman WJ, et al. Variation in the GST mu locus and tobacco smoke exposure as determinants of childhood lung function. *Am J Respir Crit Care Med* 2009;**179**(7):601—7.
45. Kabesch M, Hoefler C, Carr D, Leupold W, Weiland SK, von Mutius E. Glutathione S transferase deficiency and passive smoking increase childhood asthma. *Thorax* 2004;**59**(7): 569—73.
46. Rogers AJ, Brasch-Andersen C, Ionita-Laza I, Murphy A, Sharma S, Klanderman BJ, et al. The interaction of glutathione S-transferase M1-null variants with tobacco smoke exposure and the development of childhood asthma. *Clin Exp Allergy* 2009;**39**(11):1721—9.
47. Takano H, Yoshikawa T, Ichinose T, Miyabara Y, Imaoka K, Sagai M. Diesel exhaust particles enhance antigen-induced airway inflammation and local cytokine expression in mice. *Am J Respir Crit Care Med* 1997;**156**(1):36—42.
48. Bommel H, Haake M, Luft P, Horejs-Hoeck J, Hein H, Bartels J, et al. The diesel exhaust component pyrene induces expression of IL-8 but not of eotaxin. *Int Immunopharmacol* 2003; **3**(10—11):1371—9.
49. Bommel H, Li-Weber M, Serfling E, Duschl A. The environmental pollutant pyrene induces the production of IL-4. *J Allergy Clin Immunol* 2000;**105**(4):796—802.
50. Salvi SS, Nordenhall C, Blomberg A, Rudell B, Pourazar J, Kelly FJ, et al. Acute exposure to diesel exhaust increases IL-8 and GRO-alpha production in healthy human airways. *Am J Respir Crit Care Med* 2000;**161**(2 Pt 1):550—7.
51. Schober W, Lubitz S, Belloni B, Gebauer G, Lintelmann J, Matuschek G, et al. Environmental polycyclic aromatic hydrocarbons (PAHs) enhance allergic inflammation by acting on human basophils. *Inhal Toxicol* 2007;**19**(Suppl. 1):151—6.
52. Babu KS, Arshad SH. The role of allergy in the development of airway inflammation in children. *Paediatr Respir Rev* 2003; **4**(1):40—6.
53. Castellazzi AM, Maccario R, Moretta A, De Amici M, Gasparoni A, Chirico G, et al. Effect of active and passive smoking during pregnancy on natural killer-cell activity in infants. *J Allergy Clin Immunol* 1999;**103**(1 Pt 1):172—3.
54. Lin L, Zhu H, Quan C, Grunig G, Ballaney M, Jin X, et al. Prenatal allergen and diesel exhaust exposure and their effects on allergy in adult offspring mice. *Allergy Asthma Clin Immunol* 2010;**6**(1). 7.
55. Brauer M. Air pollution and development of asthma, allergy and infections in a birth cohort. *Eur Respir J* 2007;**29**(5): 879—88.
56. Rauh VA, Whyatt RM, Garfinkel R, Andrews H, Hoepner L, Reyes A, et al. Developmental effects of exposure to environmental tobacco smoke and material hardship among inner-city children. *Neurotoxicol Teratol* 2004;**26**(3):373—85.
57. DeLorenze GN, Kharrazi M, Kaufman FL, Eskenazi B, Bernert JT. Exposure to environmental tobacco smoke in pregnant women: the association between self-report and serum cotinine. *Environ Res* 2002;**90**(1):21—32.
58. Wagenknecht LE, Manolio TA, Sidney S, Burke GL, Haley NJ. Environmental tobacco smoke exposure as determined by cotinine in black and white young adults: the CARDIA Study. *Environ Res* 1993;**63**(1):39—46.