University of Cincinnati

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I, Nicholas C Newman, hereby submit this original work as part of the requirements for the degree of Master of Science in Clinical and Translational Research.

It is entitled:
Traffic Related Air Pollution Exposure in the First Year of Life and Hyperactivity at Age Seven in a High Risk Atopic Birth Cohort

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Traffic Related Air Pollution Exposure in the First Year of Life and Hyperactivity at Age Seven in a High
Risk Atopic Birth Cohort

A thesis submitted to the
Graduate School
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Master of Science

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the Division of Epidemiology and Biostatistics
Department of Environmental health
of the College of Medicine

by

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D.O. Kirksville College of Osteopathic Medicine, June 1997
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Abstract

Objective: The purpose of this study is to explore the association between attention deficit hyperactivity disorder (ADHD) symptoms and exposure to traffic related air pollution (TRAP) in children at risk for atopic disease.

Methods: A cross sectional analysis assessing exposure to TRAP and ADHD symptoms was conducted utilizing seven-year-old children who were participants in a longitudinal birth cohort. They were recruited from a mid-sized metropolitan area with a mix of rural, suburban, and urban areas. Enrollment in the Cincinnati Childhood Allergy and Air Pollution Study (CCAAPS) cohort required at least one parent with atopy and birth residence either <400m or >1500m from a major highway or bus route. Exposure to TRAP was modeled using data from 27 air monitoring stations over the first year of life using land-use regression modeling. ADHD symptoms were measured using the Hyperactivity, Inattention, Aggression, Conduct Problems, and Atypicality T scores from the Behavioral Assessment System for Children 2nd Edition, Parent Rating Scale (BASC-2).

Results: At age seven, 18.4% of children were reported to have hyperactive behaviors in the “at risk” range (T score > 59), 19.3% had inattentive behaviors, 15.7% had aggressive behaviors, 14.1% had conduct problems, and 14.3% had atypical behaviors. In analysis adjusting for cigarette exposure, maternal education, rhinitis, and habitual snoring, the children with the highest tertile of TRAP exposure during their first year of life had significantly more hyperactive behaviors than children exposed to lower amounts of TRAP (aOR=1.68 [1.02, 2.74]). After stratifying by race, black children who had high TRAP exposure had significantly higher aggressive behaviors than children exposed to lower amounts of TRAP (aOR=4.19 [1.08, 21.58]).
Conclusions: Higher exposure to TRAP is associated with hyperactive behaviors in children at risk for atopy. Black race modifies the association between TRAP exposure and aggressive behaviors.
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**Introduction**

Attention Deficit/Hyperactivity Disorder (ADHD) is a common psychiatric diagnosis in children characterized by symptoms of inattentiveness and/or hyperactivity that are present in more than one setting, that are present before age seven, and not explained by another cause. ADHD affects between seven and 9.5% of children in the United States between 4 and 15 years old, or approximately 4.0 million children. Epidemiological studies suggest that both genetic and environmental factors are associated with the development of ADHD. Analysis of The National Health and Nutrition Examination Survey (NHANES) 1999-2002 data demonstrates an association between environmental tobacco smoke, lead and ADHD. Although there is a strong familial tendency towards the development of ADHD with heritability estimated to be as high as 75%; there is no single gene that explains the heritability of ADHD.

Traffic-related air pollution exposure (TRAP), consisting partly of gaseous pollutants (oxides of nitrogen and sulfur), and particles, including ultrafine particles less than 100nm in diameter. It is associated with adverse effects on the cardiopulmonary system in children. In particular, smaller particles, including ultrafine particles (UFP) are implicated in the pathophysiology of air pollution related disease. Despite improvements in technology and stricter environmental regulation, as of 2006 there were 211 counties in the United States that were in nonattainment of their PM2.5 levels.

Research in animals and humans demonstrates that the brain is another organ that can be affected by ultrafine particle exposure. Using a rat model, Oberdorster et al. demonstrated translocation of inhaled ultrafine particles from the respiratory tract to extrapulmonary sites including the olfactory bulb. Work done by Calderon et al. demonstrated an association between exposure to high levels of ambient air pollution and histological changes in the brain consistent with neuroinflammation. This autopsy study of otherwise normal children and young adults who died from accidents also demonstrated the
presence of particulate matter in the perivascular tissues of the capillaries in the frontal cortex.

Additional work by this same group demonstrated brain parenchymal changes on MRI as well as significant deficits in cognition, memory, and executive function in children exposed to high levels of ambient air pollution as compared to matched controls. Both Suglia et al. and Wang et al. have demonstrated a significant association between air pollution and negative neurocognitive outcomes in children. Exposure to black carbon (a marker of TRAP and diesel exhaust) is associated with decreased cognitive scores for verbal, nonverbal intelligence and memory in a childhood population.

In this same cohort, they noted an association between black carbon exposure, decreased lung function, and decreased cognitive scores. Negative but non-statistically significant associations were found between NO2 exposure (surrogate for TRAP) and cognitive development in children in a European birth cohort.

There are also a number of studies that suggest ADHD may be related to an allergic/hypersensitivity disorder. Although the controversial hypothesis of a connection between food additives and hyperkinetic symptoms was proposed more than thirty years ago, ongoing work has strengthened the argument that there may be a connection between ingested or inhaled compounds and ADHD. Two more recent double-blinded randomized controlled trials demonstrate an association between the consumption of food additives (sodium benzoate and artificial colorings) with ADHD symptoms in a community setting. Observational data demonstrates a high prevalence of allergic rhinitis in children with ADHD. A recent study has shown a non-statistically significant trend towards ADHD in children with asthma. Additional recent work also suggests that neurocognitive deficits at age four may predict atopic symptoms at age six.

The association between early childhood exposures to environmental chemicals and subsequent neurodevelopmental disorders has been reviewed elsewhere. Knowledge about the deleterious
The effects of lead, polychlorinated biphenyls, and mercury on children’s developing brains suggests that early life exposures can have substantial consequences for adverse neurobehavioral outcomes later in life. The objective of this analysis is to examine the effect of TRAP during early neurodevelopment on ADHD symptoms in a group of children at risk for atopy.

**Materials and Methods:**

**Study Population**
The sample for the analysis was drawn from participants in the Cincinnati Childhood Allergy and Air Pollution Study (CCAAPS). CCAAPS is a prospective birth cohort that was selected based on their residence at time of birth being either near (<400m) or far (>1500m) from a major highway or bus route. Only children with atopic parents, as documented by skin prick testing, were included in the study. The study design has been described previously. Briefly, using birth records, newborns were identified from the Cincinnati metropolitan area from October 2001 to July 2003. Eligibility was verified after geocoding residential addresses from the birth certificates. Parents were screened using an allergy symptom questionnaire (ASQ). Any affirmative response on the ASQ qualified the parent to undergo skin prick testing (SPT). A positive SPT to one aeroallergen was required to enroll an infant in the study. Children attended clinic visits at age one, two, three, four and seven years when a history and physical exam were obtained and SPT was performed. Parents also reported the locations that their child spent their time during the previous year. At age seven, parents also completed the Behavioral Assessment System for Children, Parent Rating Scale 2nd Edition (BASC-2). The CCAAPS study was approved by the University of Cincinnati Institutional Review Board, parents provided informed consent prior to their children enrolling in the study.
Traffic-Related Air Pollution
Exposure to TRAP was estimated using a land-use regression (LUR) model adjusted for land use, traffic, and other geographic covariates. Between 2001 and 2005 there were 24 air monitors maintained by the CCAAPS study and three additional detectors maintained by the Hamilton County Department of Environmental Services (HCDOES). Air samples were obtained on a rotating basis from the CCAAPS-maintained detectors. PM2.5 samples were collected on 37-mm Teflon membrane filters and 37-mm quartz filters using Harvard-type impactors. The average concentration of elemental carbon attributable to traffic (ECAT) were calculated as described elsewhere. ECAT was used as a surrogate marker of traffic exposure and utilized to estimate each individual child’s TRAP exposure. A time-weighted average daily exposure to ECAT was determined for each year of the child’s life based upon the LUR model and the reported number of hours per week that the child spent in different parts of the Cincinnati metropolitan area.

Attention Deficit/Hyperactivity Disorder Symptoms
At the seven year clinic visit, the child’s parent completed the Behavioral Assessment System for Children 2nd Edition, Parent Rating Scale (BASC-2). The BASC-2 is a psychological assessment questionnaire consisting of 160 questions that are answered “Never, Sometimes, Often, and Always” that is completed by a parent. It is designed to assess a child’s adaptive and problematic behaviors in both the community and home settings. The BASC-2 was validated for use on a United States based school age population. After the parent completed the BASC-2, it was scored electronically (BASC-2 PRQ ASSIST™). BASC-2 scores consist of composite and subscale T scores with a mean of 50 and standard deviation of 10. For this analysis, we examined the hyperactivity, attention problems, aggression, conduct problems, and atypicality subscales, as these symptoms can co-exist with ADHD. To prevent the results from being compromised, the BASC-2 has three internal validity scores. The F (“Fake bad”) index assesses whether the parent rated their child in an overly negative fashion. A high score on this
scale suggests that the parent rated the child’s behavior more severely than expected. The Response Pattern (R) Index is a count of the number of times an individual item’s response differs from the previous item’s response. A high R index value suggests that parent was not attentive to content of the questions. The Consistency Index identifies situations where the parent provided different answers to questions that are usually answered similarly. The scoring protocol for the BASC-2 allows for no more than two missing items per each rating scale.

All surveys with a large number of missing items were excluded (n=5) as were all surveys with F-Index scores greater than six (n=3). Surveys with a consistency score greater than 17 were also excluded (n=5). Surveys with response pattern scores suggesting inattention to the questionnaire were also excluded (n=9). Some surveys had multiple consistency problems and were counted twice. Ultimately 22 surveys were excluded from the analysis. The final number of surveys available for analysis was 576.

The T scores for individual participants were dichotomized into those in the “at risk” range or higher versus those below the “at risk” cut-off T score of 59. The “at risk” designation is used clinically to indicate those children who have significant but not yet clinically important behavioral problems in a given subscale.

**Housing age**

Housing age was used as a surrogate marker for lead exposure. Using the child’s address from age 6 months, housing age was determined by looking up the year built from the appropriate auditor’s website (Ohio addresses and some Kentucky addresses) or by manually reviewing county auditor’s records (some Kentucky addresses). If this information was not available (the home was demolished or the age was not listed), the age was imputed using the mean age of the homes surrounding the address in question. The age of the home was transformed to a categorical variable based on the age of the home. If the home was built before 1950, the value is 1, and if it was built after 1950, the value is 0.
**Rhinitis**
Rhinitis was defined as an affirmative answer to the question “In the past 12 months, has your child ever had a problem with sneezing, or a runny, or a blocked nose when he/she DID NOT have a cold or flu?” This question was asked of parents at each study clinic visit. Responses to this question from years one to four were available for analysis. To reduce the chance for misclassification, a child was scored positive for rhinitis only if a parent reported a positive answer at two or more different visits.

**Habitual Snoring**
Habitual snoring was defined as a parent report of snoring three to four times per week during the first year of life. This was assessed at the year one visit.

**Tobacco smoke exposure**
Environmental tobacco smoke (ETS) exposure during pregnancy was estimated using a questionnaire that asked about number of packs of cigarettes smoked during each trimester of the pregnancy. The mean of this number of cigarettes/day for each trimester was averaged over the pregnancy to determine the average number of cigarettes per day during the pregnancy. Postnatal ETS exposure during the 1st year of life was also obtained from a questionnaire then dichotomized into exposed and non-exposed based on parental report of any cigarettes smoked in the home. Hair cotinine, a biomarker for ETS exposure was also obtained. Hair samples were collected by cutting approximately 20 strands of hair from the occipital region of the scalp at each annual visit. After adjusting for weight, these samples were analyzed by radioimmunoassay at the Hospital for Sick Children in Toronto, Canada. The limit of detection for cotinine was 0.02ng/mg.\(^36\) Hair cotinine levels below the limit of detection were imputing based on the method of Hornung and Reed, using the equation: \[\text{Imputed value} = \frac{\text{LCD}}{\sqrt{2}}.\]\(^37\) Hair cotinine levels were treated as continuous variables and were log transformed because the distribution was skewed to low exposure.
**Demographic variables**
Maternal education was dichotomized into completion of high school/ GED or higher, compared to non-completion of high school. Race was dichotomized into those of African American descent versus non-African American descent based on the race of either parent. Race was determined through parental report.

**Breast feeding**
Mothers reported their duration of breastfeeding at the annual study visits from years one through three. A dichotomous variable was created representing five or more months of breastfeeding. The five month breakpoint was selected because breastfeeding for greater than 20 weeks has been associated with significant improvements in both ADHD symptoms and executive functioning.38

**Eczema clinical symptoms**
We defined the presence of eczema based on parental report (at the 4 year visit) of the following symptoms: scratching, redness, “raised bumps,” or dry/scaling skin over 6 of the past 12 months.
Determination of atopic eczema was based on combining the above skin symptoms with positive SPT results.39

**Skin Prick Tests**
Skin prick testing (SPT) was done at each study clinic visit. Because there were children who were not consistently positive for skin prick testing, only those with persistently positive SPT for aeroallergens were counted as being SPT positive. Persistently positive aeroallergen SPT was defined as any two or more years with a positive aeroallergen SPT during the first four years of life.

**Daycare**
Parents were asked where their child spent his/her time in the months prior to the six month and twelve month clinic visits. They were asked whether one of the locations was a babysitter, daycare, or a relative. This was defined as “out of home daycare.” This address location was also checked to see if it
was a licensed daycare center. Based on this information, we determined whether the child was in “out of home daycare.” If the child was determined to be in out of home daycare at either the 6 month or 12 month visit, they were labeled as daycare. Daycare was dichotomized for the first year into yes or no.

**Statistical Analysis**

Descriptive statistics were calculated for population characteristics, environmental exposures, objective and subjective medical findings, and behavioral scores. Linear regression was carried out to examine the univariate relationship between dependent variables (BASC-2 T scores) and the independent variable (first year TRAP exposure). To better understand the relationship between TRAP exposure and hyperactivity scores in the “at risk” range, a contingency analysis was done comparing tertiles of TRAP of exposure to hyperactivity T scores in the “at risk” range. The relationship between TRAP and hyperactivity changed at the highest tertile as compared to the lower two tertiles. Therefore, TRAP exposure was dichotomized at the highest tertile versus the lower two tertiles.

Prior to constructing multivariate logistic regression models with the BASC-2 hyperactivity T score as an outcome, a panel of possible covariates were tested including: gender, race, maternal education, paternal education, household income, Medicaid status, duration of breastfeeding, pre- and postnatal cigarette exposure, hair cotinine levels, rhinitis status, skin prick testing status, age of home, daycare attendance, atopic dermatitis status, and habitual snoring. Since there were a large number of covariates identified, several methods were used to reduce the number of covariates. Associations between binary outcomes and predictor variables were evaluated using Pearson Chi-Square for categorical predictors and logistic regression for continuous predictors. Equality of the distributions of continuous outcomes across grouped continuous and categorical predictor variables was tested by one-way ANOVA (normally distributed) or Wilcoxon/Kruskal-Willis testing (non-normally distributed). To screen for potential confounding all covariates were compared individually with the primary exposure of
interest, TRAP exposure during the first year of life. Those covariates that were statistically significant at the $\alpha=0.05$ level were included in the final model.

Those covariates that were associated with T scores for hyperactivity and attention at the 0.05 level were included in the final models. For parsimony only maternal education was chosen to represent socio-economic status. The final logistic regression models included adjustment for only environmental exposures alone, environmental exposures plus demographic factors, and finally environmental, demographic, and medical risk factors.

We dichotomized TRAP exposure into highest tertile versus lower two tertiles and dichotomized BASC-2 T scores into those in the at-risk range or worse versus those not in the at risk range.

In order to evaluate the separate effects of race and TRAP, indicator variables which combined the levels of each variable were created and compared.

Statistical analysis was carried out using JMP 9.40

**Results**

A total of 599 children had BASC-2 questionnaires available for analysis, of which 576 passed the BASC-2 internal validity measures criteria. Of the 576 children who had valid BASC-2 evaluations at age 7 and a complete 1 year assessment, 55 percent were male and 20.3 percent were African American. Maternal education was used as a surrogate for socio-economic status as it was significantly associated with income and Medicaid status. Maternal education was at the level of high school/GED or less for 21.3 percent of mothers. Eleven percent reported tobacco use during pregnancy whereas 21.6 percent reported tobacco exposure during the first year of life. The mean ± standard deviation (SD) for age was 6.91 ± 0.31 years. Of the 576 children with valid BASC-2 questionnaires, the mean ± SD T scores for
selected subscales are as follows: Hyperactivity, 50.8 ± 10.3; Aggression, 50.3 ± 9.6; Conduct Problems, 50.8 ± 10.6; Atypicality, 49.4 ± 9.5; and Attention Problems, 50.9 ± 10.2. The mean ± SD predicted time-weighted TRAP exposure estimate (expressed in µg/m³ elemental carbon attributable to traffic) was 0.40 ± 0.14. As compared to the 186 children who did not have valid data for our analysis, there were significant differences in maternal education, family income, year two hair cotinine, and prenatal cigarette exposure. Children who had data for analysis were more likely to have mothers with higher education, higher household income and less cigarette exposure. The percentage of children of black ancestry was not significantly different between the groups and the mean TRAP exposure was not significantly different. See table 1.

Prenatal and post natal tobacco smoke exposure based on questionnaire was strongly associated with year 2 hair cotinine levels (p<0.001). This association was present for both children of black ancestry (P<0.001) and non-black ancestry (p<0.001).

Black ancestry was significantly associated with TRAP exposure (p<0.001). Therefore, due to the strong association between black race and the primary exposure variable, the analyses were conducted both stratifying by race and non-stratified. TRAP was also associated with the cofactors of home age (p<0.001), lower maternal education (p<0.001), lower family income (p<0.001), breast feeding for less than 5 months (p<0.001), and higher hair cotinine (p<0.001). Black race was not significantly associated with either rhinitis in the first year of life or persistent rhinitis. Black race was not significantly associated with risk for a hyperactivity T score in the at risk range.

Report of rhinitis in the 1st year of life was significantly associated with a higher Somatization T score in the year 7 BASC-2 (p=0.03). It was not significantly associated with a Somatization T score in the at-risk range or higher (p=0.07). Persistent rhinitis was associated with higher Somatization T scores (p=0.02) and a higher odds of being in the at-risk or clinical range (p=0.01). Report of persistent rhinitis was
significantly associated with use of second-generation anti-histamines \((p<0.01)\), but not with the use of first-generation anti-histamines \((p=0.84)\). Use of either first or second-generation anti-histamines was not associated with BASC-2 T scores in the clinical range for the hyperactivity, attention, aggression, conduct problems or atypicality subscales.

Linear regression analysis using continuous values for TRAP \((\mu g/m^3)\) versus BASC-2 T scores was carried out for the five subscales of interest (table 2). Only Atypicality was significantly associated with increases in TRAP exposure.

Bivariate analysis of TRAP exposure and BASC-2 T scores demonstrated associations between the Externalizing \((p=0.01)\), and Behavioral Symptom Index \((p=0.01)\) composite scales as well as the Hyperactivity \((p<0.01)\), Conduct Problems \((p<0.01)\), Depression \((p=0.02)\), Atypicality \((p<0.01)\), Adaptability \((p=0.01)\), Leadership \((p<0.01)\), and Activities of Daily Living \((p=0.01)\) subscales (table 3). In keeping with our initial hypothesis that TRAP exposure may be related to ADHD symptoms, our analysis focused only on those ADHD-related subscales.

**Logistic regression analysis**

Logistic regression models (table 4) were developed to demonstrate the association between high TRAP exposure and the ADHD related BASC-2 T scores (hyperactivity, inattention, aggression, conduct problems, and atypicality). In logistic regression analyses, an increase in TRAP from the lower two tertiles to the highest tertile predicted an odds ratio for a hyperactivity score in the at risk range of 1.87 \((95\% CI: 1.22, 2.88)\), an odds ratio for inattention of 1.41 \((CI: 0.92, 2.15)\), an odds ratio for aggression of 1.49 \((CI: 0.94, 2.36)\), an odds ratio of conduct problems 2.08 \((CI: 1.29, 3.34)\), and the odds ratio for Atypicality 2.02 \((CI: 1.26, 3.25)\). Next, logistic multivariate models adjusting for gender, cigarette exposure during the first year of life, and maternal education were constructed. In this analysis the odds ratio for an increased hyperactivity score was 1.69 \((CI: 1.04, 2.73)\) and odds ratios for
attention, aggression, conduct problems, and atypically were not statistically significant. Further adjustment for reported rhinitis in two or more years and for habitual snoring during the first year of life did not reduce the effect of TRAP on hyperactivity. Stratification for race nullified the effect of TRAP for some BASC-2 T scores, but emphasized the effect of TRAP on aggression T scores in those of black ancestry (table 4).

Discussion
In this prospective birth cohort, early life exposure to TRAP was associated with increased odds of significant hyperactivity symptoms at school age. Based on this analysis, it is not possible to imply causation between TRAP exposure and ADHD symptoms. Although not statistically significant, we did find that other BASC-2 subscales related to ADHD were also negatively impacted by exposure to TRAP. Since multiple aspects of behavior are negatively affected by TRAP exposure, this suggests that our finding is not purely due to chance. In addition, for the aggression BASC-2 subscale, children of black ancestry were significantly more likely to be in the at risk range when exposed to TRAP as compared to their non-black counterparts. Finally, there was also a strong relationship between habitual snoring and rhinitis in early life with later symptoms of hyperactivity.

Several biological mechanisms could explain the association between hyperactive behaviors and exposure to TRAP. Children chronically exposed to environments with high PM2.5 experience significantly increased levels of inflammatory mediators and vasoconstrictors.41 Neuroinflammatory changes were observed in a group of children who lived in a high PM2.5 environment as evidenced by upregulation of cyclooxygenase-2, interleukin-1beta and CD14 in the frontal cortex.13 These same investigators also found particles <100nm diameter in intraluminal erythrocytes within the frontal cortex.13 Dysfunction of frontal-cortical circuits is associated with ADHD.5 In vitro studies show that diesel exhaust particles (DEP), a component of TRAP, are selectively toxic to dopaminergic neurons in a
rat tissue culture system.\textsuperscript{42} Although the exact biological mechanism for ADHD has not been identified, there is evidence implicating the dopaminergic system.\textsuperscript{5,43-45} The combination of evidence of deposition of TRAP into the prefrontal areas and its toxicity to dopaminergic neurons suggests a biological mechanism for the pathogenesis of TRAP in regards to ADHD.

Inhalation of TRAP also affects both the upper and lower airways. Children with high TRAP exposure have both irritation of the nasal airways as well as increased wheezing and night time cough.\textsuperscript{31,46,47} A study of children with ADHD found that they had a higher prevalence of sleep disorders as measured by polysomnography as compared to controls.\textsuperscript{48} This included children with sleep related disorder breathing. We found that habitual snoring in early life was associated with higher hyperactivity T scores on the BASC-2 and that this relationship existed in the context of high TRAP exposure. Rhinitis for more than one year was also significantly associated with hyperactivity. Irritation of either upper or lower airways from TRAP exposure may be sufficient to impact sleep efficiency and impact symptoms of ADHD.

In blacks with high TRAP exposure, there were higher scores for aggression. A similar association has been found between lead exposure in early childhood and aggressive behaviors later in life.\textsuperscript{49} This suggests a similar mechanism or else a common risk factor. Since exposure to TRAP was also strongly associated with an older home, it is possible that there is an effect of lead exposure in this group. The age of the home however, was not associated with increased odds for a high hyperactivity T score. Therefore the association between TRAP and aggressive behaviors is independent of the risk for lead poisoning due to the family living in an older home.

We hypothesized that children with allergies would be more susceptible to TRAP exposure because of the chronic inflammation of their airways, thus allowing easier passage of UFP into the body. Although we found an association between rhinitis and hyperactivity, there was no significant association
between positive skin prick testing and hyperactivity. This suggests that there is an association between manifestation of symptoms commonly associated with allergy and hyperactivity, but not with atopy per se.

**Strengths and Limitations**
Among the strengths of this study is that it is a longitudinal birth cohort and the measurements of TRAP over the course of the study are well characterized. The BASC-2 is a carefully validated instrument that provides scores that are applicable to a clinical setting. The cohort is racially heterogeneous thus allowing us to look at the impact of race on both the exposures and the outcome. Finally, since the population of the Cincinnati metropolitan area tends to be stable, we have maintained a high percentage of the original cohort in the study (78%). Children who completed the study did not differ in their exposure to TRAP during the first year of life as compared to those who did not complete the study. However, the children who completed the study had mothers who were significantly better educated, had higher family income and less prenatal tobacco smoke exposure than those who did not finish the study.

There was a trend towards children with higher early life TRAP exposure being prescribed any ADHD medications. Although the number of children on ADHD medications was small (n=17), the percentage of children in our study on ADHD medications (3%) matches what is found in national samples. The percentage of children with either hyperactivity (6%) or inattention (5%) scores in the clinical range suggests that this group is not different from the general population in terms of ADHD prevalence, given the age of the children in the study.

We found an association between high TRAP exposure and atypical behaviors (behaviors that can be associated with autism spectrum disorders), but this association diminished when incorporated into a multivariate model. Recently published work from the Childhood Autism Risks from Genetics and the
Environment (CHARGE) Study, found a strong relationship between distance to highway in autism in a case-control study after correcting for covariates. The reason for this difference is likely due to lack of power in our study to detect this association. In a cohort of children from Menorca, Spain with high exposure to oxides of nitrogen from indoor cooking, there was an association between NOx exposure and inattention behaviors but not hyperactivity. We found no association between TRAP exposure and inattention, but a strong association between TRAP and hyperactivity. This can be explained because there is a complex relationship between NOx levels and fine particulate matter (PM) generated during cooking. Since indoor PM production varies indoors depending on the technique of cooking (i.e. frying) regardless of the baseline level of air pollution created by the burning of the flame to cook, measurement of NOx levels may not be a good surrogate for PM production.

One limitation of the study is that the population was selected on the basis of being at high risk for atopy and therefore may not be generalizable. We found however, that the percentage of children who would meet clinical criteria for ADHD based on their BASC-2 scores alone (not the DSM-IV criteria for ADHD) is close to the national estimates for ADHD prevalence. The Third National Health and Nutritional Examination Survey (NHANES III) found that 54.3% of the U.S. population was skin prick test positive to at least one of ten common allergens. Thus, this at-risk cohort represents over 50% of the U.S. population.

Unfortunately, we do not have any information on the behavioral health history of the families in the study. However, there is no evidence to suggest that families with a history of ADHD would be more likely to live, play, and attend school in an area with high TRAP exposure. Childhood lead exposure is associated with ADHD but children in this study did not provide a blood lead level. As a surrogate, we looked at the age of the home as the majority of lead poisoning is children is caused by exposure to
deteriorating paint in older buildings. We found no association between the age of the home and a child’s hyperactivity T score.

In the future, it will be important to understand whether it is early childhood exposure to TRAP or TRAP exposures closer to the time of behavioral testing that is more important in influencing the ADHD phenotype. This information may inform research regarding the biological mechanism as well as approaches to ameliorate the exposure. Gene x environment interactions have been observed with the GSTP1 variant Ile105Val, indoor air pollution from NOx and behavior and cognition.52 This association has not been demonstrated with TRAP exposure and understanding this may describe populations that are at higher risk for the ill effects of TRAP exposure. Given the large numbers of people exposed to TRAP, this understanding may have implications for public health.
### Tables

#### Table 1a: Characteristics and environmental exposures for children either included or excluded from study analysis (n=576).

<table>
<thead>
<tr>
<th>Demographic Characteristics</th>
<th>Included n=576</th>
<th></th>
<th>Excluded n=186</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>%</td>
<td>Mean(SD)</td>
<td>No.</td>
</tr>
<tr>
<td>Age when BASC-2 administered</td>
<td>576</td>
<td>6.9(0.3)</td>
<td>NA</td>
<td>49</td>
</tr>
<tr>
<td>Black ancestry</td>
<td>117</td>
<td>20.3</td>
<td>NA</td>
<td>49</td>
</tr>
<tr>
<td>Male</td>
<td>317</td>
<td>55</td>
<td>NA</td>
<td>98</td>
</tr>
<tr>
<td>Mother’s education level (HS/GED or less)*</td>
<td>119</td>
<td>21.3</td>
<td>NA</td>
<td>66</td>
</tr>
<tr>
<td>Family income &lt;$30K*</td>
<td>131</td>
<td>23.5</td>
<td>NA</td>
<td>71</td>
</tr>
<tr>
<td><strong>Environmental Exposures</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TRAP Exposure Year 1</td>
<td>576</td>
<td>0.4(0.1)</td>
<td></td>
<td>186</td>
</tr>
<tr>
<td>Hair cotinine Year 2*</td>
<td>451</td>
<td>0.2(0.3)</td>
<td></td>
<td>123</td>
</tr>
<tr>
<td>Prenatal cigarette exposure*</td>
<td>57</td>
<td>10.5</td>
<td></td>
<td>33</td>
</tr>
<tr>
<td>Cigarette exposure Year 1</td>
<td>110</td>
<td>21.6</td>
<td></td>
<td>47</td>
</tr>
<tr>
<td>Home built before 1950</td>
<td>134</td>
<td>26.9</td>
<td></td>
<td>38</td>
</tr>
</tbody>
</table>

* p < 0.05 comparing children included and excluded from analysis

#### Table 1b: Additional characteristics of children who completed the study through age 7

<table>
<thead>
<tr>
<th>Past Medical History</th>
<th>No.</th>
<th>%</th>
<th>Mean(SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Any positive SPT in 2 or more years</td>
<td>260</td>
<td>45.1</td>
<td></td>
</tr>
<tr>
<td>Rhinitis reported in 2 or more years</td>
<td>228</td>
<td>39.6</td>
<td></td>
</tr>
<tr>
<td>Atopic dermatitis in year 3</td>
<td>68</td>
<td>14.6</td>
<td></td>
</tr>
<tr>
<td>Habitual snoring in year 1</td>
<td>85</td>
<td>15.8</td>
<td></td>
</tr>
<tr>
<td>Any ADHD medications Year 7</td>
<td>17</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td><strong>BASC-2 T scores, year 7 (continuous)</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hyperactivity</td>
<td>50.8(10.3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inattention</td>
<td>50.9(10.2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aggression</td>
<td>50.3(9.6)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Conduct Problems</td>
<td>50.8(10.6)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Atypicality</td>
<td>49.4(9.5)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>BASC-2 T scores “at-risk” range</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hyperactivity</td>
<td>106</td>
<td>18.4</td>
<td></td>
</tr>
<tr>
<td>Inattention</td>
<td>111</td>
<td>19.3</td>
<td></td>
</tr>
<tr>
<td>Aggression</td>
<td>90</td>
<td>15.7</td>
<td></td>
</tr>
<tr>
<td>Conduct Problems</td>
<td>81</td>
<td>14.1</td>
<td></td>
</tr>
<tr>
<td>Atypicality</td>
<td>82</td>
<td>14.3</td>
<td></td>
</tr>
</tbody>
</table>
Table 2: Parameter estimates ($\beta$) with 95% Confidence Intervals (CI) obtained from linear regression of selected BASC-2 T scores on TRAP exposure during the first year of life ($n = 576$), Cincinnati Childhood Allergy and Air Pollution Study*

<table>
<thead>
<tr>
<th>Hyperactivity</th>
<th>Attention</th>
<th>Aggression***</th>
<th>Conduct Problems</th>
<th>Atypicality</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\beta$</td>
<td>$\beta$</td>
<td>$\beta$</td>
<td>$\beta$</td>
<td>$\beta$</td>
</tr>
<tr>
<td>0.7</td>
<td>0.6</td>
<td>0.001</td>
<td>0.8</td>
<td>0.9</td>
</tr>
<tr>
<td>-0.1, 1.5</td>
<td>-0.2, 1.3</td>
<td>-0.7, 0.8</td>
<td>-0.03, 1.6</td>
<td>0.1, 1.6**</td>
</tr>
</tbody>
</table>

* Change in subscale score per interquartile-range (0.13 µg/m$^2$) increase in TRAP level

** $p < 0.05$

*** Due to missing data for the aggression subscale, $n = 575$

Table 3: Univariate analysis of BASC-2 T scores in at risk range by highest tertile of TRAP Odds Ratios (OR) with 95% Confidence Interval (CI) ($n = 576$)

<table>
<thead>
<tr>
<th>BASC-2 Scale</th>
<th>OR</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Externalizing***</td>
<td>1.8</td>
<td>1.2, 2.8**</td>
</tr>
<tr>
<td>Hyperactivity</td>
<td>1.9</td>
<td>1.2, 2.9**</td>
</tr>
<tr>
<td>Aggression***</td>
<td>1.5</td>
<td>0.9, 2.4*</td>
</tr>
<tr>
<td>Conduct Problems</td>
<td>2.1</td>
<td>1.3, 3.3**</td>
</tr>
<tr>
<td>Internalizing</td>
<td>1.1</td>
<td>0.7, 1.8</td>
</tr>
<tr>
<td>Anxiety</td>
<td>1.2</td>
<td>0.8, 1.9</td>
</tr>
<tr>
<td>Depression</td>
<td>1.8</td>
<td>1.1, 3.1*</td>
</tr>
<tr>
<td>Somatization</td>
<td>1.3</td>
<td>0.8, 2.1</td>
</tr>
<tr>
<td>Behavioral Symptom Index</td>
<td>1.9</td>
<td>1.2, 3.1**</td>
</tr>
<tr>
<td>Atypicality</td>
<td>2.0</td>
<td>1.3, 3.3**</td>
</tr>
<tr>
<td>Withdrawal</td>
<td>1.3</td>
<td>0.8, 2.0</td>
</tr>
<tr>
<td>Attention</td>
<td>1.4</td>
<td>0.9, 2.2</td>
</tr>
<tr>
<td>Adaptive Skills***</td>
<td>1.6</td>
<td>1.0, 2.5</td>
</tr>
<tr>
<td>Adaptability</td>
<td>1.8</td>
<td>1.1, 2.8*</td>
</tr>
<tr>
<td>Social Skills</td>
<td>1.2</td>
<td>0.8, 1.9</td>
</tr>
<tr>
<td>Leadership</td>
<td>2.7</td>
<td>1.4, 5.3**</td>
</tr>
<tr>
<td>Activities of Daily Living</td>
<td>1.6</td>
<td>1.0, 2.6*</td>
</tr>
<tr>
<td>Functional Communication***</td>
<td>1.4</td>
<td>0.9, 2.2</td>
</tr>
</tbody>
</table>

* $p < 0.05$

** $p < 0.01$

*** Due to missing data $n = 575$
Table 4. Odds ratio (OR) with 95% confidence interval (CI) from logistic regression of ADHD-related BASC-2 subscales by highest tertile of first year TRAP estimates; the first model is with TRAP alone, the others with adjustment (n = 576), Cincinnati Childhood Allergy and Air Pollution Study

<table>
<thead>
<tr>
<th>TRAP Model</th>
<th>Hyperactivity</th>
<th>Attention Problems**</th>
<th>Aggression</th>
<th>Conduct Problems</th>
<th>Atypicality</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.9 1.2, 2.9*</td>
<td>1.4 0.9, 2.2</td>
<td>1.5 0.9, 2.4</td>
<td>2.1 1.3, 3.3</td>
<td>2.0 1.3, 3.3</td>
</tr>
<tr>
<td>Adjusted for gender, year 1 cigarette exposure, and maternal education</td>
<td>1.7 1.0, 2.7*</td>
<td>1.1 0.6, 1.7</td>
<td>1.2 0.7, 2.0</td>
<td>1.5 0.9, 2.6</td>
<td>1.5 0.9, 2.6</td>
</tr>
<tr>
<td>Adjusted for above factors + two or more years of reported rhinitis, habitual snoring in year 1</td>
<td>1.7 1.0, 2.7*</td>
<td>1.0 0.6, 1.7</td>
<td>1.8 0.7, 2.0</td>
<td>1.6 0.9, 2.7</td>
<td>1.5 0.8, 2.6</td>
</tr>
</tbody>
</table>

* p < 0.05  
** Due to missing data n = 575
Table 5. Odds ratio (OR) with 95% confidence interval (CI) from logistic regression of ADHD-related BASC-2 subscales by highest tertile of first year TRAP estimates; the first model is with TRAP alone, the others with adjustment (n = 576) all stratified by race, Cincinnati Childhood Allergy and Air Pollution Study

<table>
<thead>
<tr>
<th>TRAP Model</th>
<th>Hyperactivity</th>
<th>Attention Problems</th>
<th>Aggression</th>
<th>Conduct Problems</th>
<th>Atypicality</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
<td>OR 95% CI</td>
</tr>
<tr>
<td><strong>Non-black race</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.8 1.1, 3.0</td>
<td>1.1 0.6, 1.8</td>
<td>1.5 0.6, 2.0</td>
<td>1.5 0.8, 2.7</td>
<td>1.7 0.9, 3.0</td>
</tr>
<tr>
<td>Adjusted for gender, year 1 cigarette exposure, and maternal education</td>
<td>1.7 1.0, 3.0</td>
<td>0.9 0.5, 1.6</td>
<td>0.9 0.5, 1.7</td>
<td>1.2 0.6, 2.3</td>
<td>1.5 0.8, 2.8</td>
</tr>
<tr>
<td>Adjusted for above factors + two or more years of reported rhinitis, habitual snoring in year 1</td>
<td>1.7 1.0, 3.0</td>
<td>0.9 0.5, 1.6</td>
<td>0.9 0.5, 1.7</td>
<td>1.2 0.6, 2.4</td>
<td>1.6 0.8, 3.0</td>
</tr>
<tr>
<td><strong>Black race</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.9 0.8, 5.4</td>
<td>1.5 0.6, 3.5</td>
<td>4.3 1.3, 19.4*</td>
<td>3.9 1.4, 14.4*</td>
<td>2.7 1.0, 8.6</td>
</tr>
<tr>
<td>Adjusted for gender, year 1 cigarette exposure, and maternal education</td>
<td>2.0 0.7, 6.3</td>
<td>1.1 0.4, 3.0</td>
<td>4.5 1.21, 22.8*</td>
<td>2.9 0.9, 11.4</td>
<td>1.40 0.4, 5.0</td>
</tr>
<tr>
<td>Adjusted for above factors + two or more years of reported rhinitis, habitual snoring in year 1</td>
<td>1.5 0.5, 5.3</td>
<td>0.8 0.3, 2.4</td>
<td>4.2 1.1, 21.6*</td>
<td>2.6 0.7, 10.8</td>
<td>1.27 0.4, 4.9</td>
</tr>
</tbody>
</table>

* p < 0.05
References

Appendix

Role in the study

Study design and hypothesis formation:
   The hypothesis and aims of this research were conceived and developed by Dr. Nicholas Newman with guidance from Dr. Kim Dietrich and Dr. Grace LeMasters. The Cincinnati Childhood Allergy and Air Pollution Study (CCAAPS) was designed and implemented by Dr. Grace LeMasters.

Data:
   The data used in this research was obtained from the CCAAPS cohort. Mr. Jeff Burkle gave assistance with accessing the CCAAPS database.

Literature review:
   Literature reviews were carried out by Dr. Nicholas Newman.

Statistical Analysis:
   Statistical analysis methods were proposed and conducted by Dr. Nicholas Newman with guidance from Dr. Linda Levin, Dr. Kim Dietrich, and Dr. Grace LeMasters.

Writing:
   Text was written by Dr. Nicholas Newman. Dr. Kim Dietrich assisted with editing of the text.

Funding

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Institutional Review Board

This study protocol was approved by the University of Cincinnati Institutional Review Board, protocol number 01-07-16-02.