# A Cross Sectional Study on the Association Between Traffic Related Air Pollutants and Biomarkers of Metabolic Dysfunction

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## ABSTRACT

It is well known that poor diet, lack of exercise, and genetics are all factors that contribute to metabolic diseases. New research reveals associations between long-term air pollution exposure and subclinical inflammation, including metabolic syndrome in adults; however, these associations are less explored in children. Communities of lower socioeconomic standing, especially those of minority populations, are most vulnerable to sources of air pollution. In many of these areas, both houses and schools are located close to areas of concentrated sources of air pollution, such as highways or factories. In this study, we conduct cross-sectional regression analyses in a predominantly low income sample of children (n=218) to explore associations between several traffic-related air pollutants: $NO_2$ , $NO_x$, elemental carbon (EC) and polycyclic aromatic hydrocarbons (PAHs) and blood biomarkers for risk of metabolic disease: high-density lipoprotein (HDL, the "good" type of cholesterol) and C-reactive protein (CRP, a marker of systemic inflammation. Overall, the results demonstrated a pattern of decreasing HDL with increases in $NO_2$, with the strongest for short term exposure (1-day) and an estimate of -15.4 mg/dL decrease per IQR increase in NO2 (95% CI = -27.4, -3.4). Bringing to light possible associations between air pollution and adverse health outcomes could promote policy change and community-wide programs to alleviate issues of environmental justice stemming from unequal burden of environmental stressors.

## KEYWORDS

airway inflammation, epidemiology, pediatric studies, C-reactive protein, HDL cholesterol

## INTRODUCTION

Particulate air pollution, especially that of small particles such as $PM_{2.5}$, has been of great health concern since the incidence of the "London Fog" in the early 1950s, where a sharp increase of mortality resulted from air pollution produced during coal burning (Corton et al. 2015, Schwartz et al. 1990). Emerging evidence showed that prolonged increases in air pollution led to impaired resistance to illnesses, resulting in increased cardiopulmonary deaths observed during air pollution episodes that may be due to a compromised respiratory system (Peters et al. 1997). There were also indicators of increases in morbidity, such as higher incidences of pneumonia, insurance claims and hospital admissions from November 1952 to January 1953 compared to previous years (Bell and Davis 2001). Four years later, the Clean Air Act of 1956 was enacted by the United Kingdom in direct response to the Great Smog of London in 1952 (Corton et al. 2015). Later on, the United States followed suit when the newly established Environmental Protection Agency (EPA) began using the Clean Air Act of 1970 to enforce air quality standards to control air pollution. From 1990-2015, annual concentrations of $PM_{10}$ have significantly decreased by 39% in the US (Park 2017, EPA 2016). However, despite effective public policy improving ambient air quality, air pollution remains a leading contributor to the global burden of disease (Park 2017, Cohen et al. 2017) and growing evidence suggests an association with metabolic dysfunction and $PM_{2.5}$ exposure at a population level (Bowe et al 2016).

Metabolic syndrome, a cluster of conditions that increase the risk of heart disease and diabetes (Mayo Clinic), has recently been associated with exposure to air pollution in mouse model studies, adult and pediatric epidemiological studies. The contributing components of metabolic syndrome, such as insulin resistance, central adiposity, elevated blood pressure, and dyslipidemia have also been shown to have a positive association with air pollution exposure in literature (Haberzettl et al. 2016, Sun et al. 2009, Alderte et al. 2017, Toledo-Corral et al. 2018, Park 2017). Currently, the hypothesized biological mechanism behind this association is that pollutants entering the bloodstream induce upregulated inflammatory responses in tissues of distant organs (liver, pancreas), leading to clinically harmful effects such as glucose intolerance from insulin resistance (Bowe et al 2016, Alderte et al. 2017, Miller et al. 2012). Studies using mouse models have investigated the biological mechanism by which particulate matter might contribute to metabolic diseases. Mice fed either a normal or high fat diet were exposed either to HEPA filtered

air or air with concentrated $PM_{2.5}$ and were followed for changes in systemic and organ-specific insulin sensitivity and inflammation (Haberzettl et al. 2016). In these mice, short term exposure to $PM_{2.5}$ inhibited insulin signaling, resulting in vascular insulin resistance and inflammation caused by pulmonary oxidative stress (Haberzettl et al. 2016). Exposure to $PM_{2.5}$ was also observed to be associated with deviations of insulin sensitivity and amplified adipose inflammation in mouse models of diet induced obesity (Sun et al. 2009). In another study, $PM_{10}$ exposure induced in vivo expression of MetS related genes in mice, specifically genes related to inflammation, lipid and cholesterol metabolism and atherosclerosis (Brocato et al. 2014). Based on this evidence from air pollution exposure in mouse models, insulin resistance, dyslipidemia and central adiposity may be related to particulate matter exposure via inflammatory pathways.

Several epidemiological studies have observed an association between air pollution exposure and metabolic biomarkers of inflammation in large population studies. A cross sectional study of adults exposed to household air pollution showed that fine particulate matter on epithelial cells lining the airways can activate inflammatory signaling cascade events, triggering adverse respiratory health effects such as asthma (Miller et al. 2012, Haberzettl et al. 2016, Villarreal et al. 2008). Moreover, in a random sample of 3256 adults during the winter of 1984–85, those with more air pollution exposure had increased plasma viscosity, thought to result from peripheral airways inflammation. The study results discuss that increased blood coagulability could result in increased cardiovascular events following urban pollution episodes (Peters et al. 1997). Several of these studies suggest that direct airway inflammatory effects from air pollutants such as ozone, secondhand tobacco smoke, and wood smoke, can lead to indirect effects on the circulatory system via the inflammatory response (Villarreal et al. 2008, Nightingale et al. 2018). Other epidemiological studies analyzed the correlation between air pollution exposure and markers of metabolic dysregulation. For example, the MESA Air study (The Multi-Ethnic Study of Atherosclerosis and Air Pollution) looked at the development of arteriosclerosis and atherosclerosis biomarkers in a population of 7,551 older adults that did not have cardiovascular disease at the start of the study. There were positive associations between longer-term exposure to TRAPs and markers of inflammation and coagulation, as well as increased rate of development of atherosclerosis measured by coronary artery calcium (CAC) (Kaufman et al. 2016). The Swiss Cohort Study on Air Pollution and Lung and Heart Diseases in Adults (3769 participants) demonstrated a positive association between traffic-related air pollutants ($NO_2$ and $PM_{10}$) with

impaired fasting glycemia (Eze et al. 2015). A recent study found an association between exposure to higher levels of traffic-related air pollution and significantly lower levels of high-density lipoprotein (HDL) cholesterol and apoA-I in a healthy study population with no cardiovascular complications (Li et al. 2019). In an analysis of US counties, there was a positive correlation between the ambient level of $PM_{2.5}$ and the prevalence of type II diabetes, but not obesity, suggesting that the exposure to air pollution to type II diabetes might be obesity independent (Mazidi et al. 2017).

Although there is abundant research in adults on the relationship between air pollution exposure and effects on metabolic dysregulation, there are still gaps in the literature concerning the effect in children. A cohort study on school children, 158 asthmatic and 50 non-asthmatic, demonstrated a positive association of air pollution exposure and inflammatory biomarkers such as interleukin-8 (IL-8) in nasal lavage and Fractional exhaled Nitric Oxide (FeNO), suggesting significant increases in airway inflammation (Villarreal et al. 2008). Additionally, another cross-sectional study cohort studied populations of overweight and obese African American and Latino children in urban Los Angeles suggests that TRAP exposure is positively associated with lower insulin sensitivity, higher fasting insulin and higher fasting glucose (Toledo-Corral et al. 2018). Similarly, in a longitudinal cohort study of overweight and obese Latino children (ranging from 8-15 years, n=314) between 2001 and 2012 in Los Angeles, CA, there was a positive association between $NO_2$ and $PM_{2.5}$ exposure and a faster decline in insulin sensitivity (Alderete et al. 2017). More studies in this area of interest can further provide evidence of ambient air pollution associated with atherosclerosis, as low levels of HDL could be a risk factor for atherosclerosis, heart failure, and future cardiovascular death (Li et al. 2019). Thus, early work suggests that the inflammatory biomarkers of airway inflammation triggered by air pollution are related to diabetes and that early life exposure to risk factors of obesity in children can contribute to metabolic dysregulation later on in life.

The Children's Health and Air Pollution Study (CHAPS) is a research project looking at the adverse health outcomes of air pollution in early childhood. This thesis investigates the exposure to TRAPs and blood biomarkers of metabolic disease in a population of low-SES, mostly Latinx children, in the San Joaquin Valley of California. The aim is to build upon an early CHAPS project which analyzed TRAPs association with other markers signaling risk of metabolic syndrome, which found a significant association between 1-year average NO2 exposure and waist-

height ratio, a measurement for obesity, and an associations between ambient PAH exposure levels and increased HbA1c and 8-isoprostane (Mann et al., *in press*). The purpose of this continuation is to provide further evidence of how early air pollution exposure can lead to adverse health effects, with regards to inflammation and metabolic disorders in children. This cohort has exceptionally detailed exposure data relative to other cohorts of children. If we find the expected association between TRAPs and markers of metabolic status in children, this could be a reason to push for efforts to increase awareness of environmental health effects in a clinical setting. We hypothesize that oxidative stress induced by exposure to ambient air pollutants such as PM 2.5 and ozone in highly polluted areas in San Joaquin Valley leads to systemic inflammation and metabolic dysfunction. This can be gauged by an increase in biomarker levels measured in the blood of these school children. Evidence of a link between traffic related air pollution and these biomarkers would suggest that air pollution plays a contributing role to abnormal fat and glucose metabolism, which could then lead to increased risk of obesity and diabetes.

## METHODS

Our hypothesis is that oxidative stress induced by exposure to traffic air pollutants in highly polluted areas of San Joaquin Valley leads to systemic inflammation and metabolic dysfunction. This can be gauged by an increase in CRP levels, an indicator of inflammation, and decrease in HDL cholesterol, a key regulator in normal metabolism, measured in the study cohort. Evidence of a link between traffic-related air pollution and these biomarkers would suggest that air pollution plays a contributing role to abnormal fat and glucose metabolism, which may then lead to increased risk of obesity and diabetes.

### Study population

The data for this analysis comes from the Children's Health and Air Pollution Study (CHAPS), which is assessing the impact of air pollution on the health of children living in the San Joaquin Valley. This study population originally came from a CHAPS recruited study consisting of children aged 6 to 8 years enrolled in Fresno elementary schools in the Fresno Unified School District (FUSD) and asked to follow up with the same population two years later, which resulted

in the 8 to 10 year old study population for this project. Details of the recruitment of the cohort can be found in prior publications from the group (Mann, *in press*). For this analysis, there were 218 subjects.

**Outcome variables**

HDL cholesterol was measured in mg/dL and C-reactive protein was measured in mg/L using standard clinical laboratory techniques. In order to minimize participant burden and maximize study participation, the study's selection of biomarkers did not require children to fast before visit and blood draws. Initially, we planned to also assess urinary 8 isoprostane, a marker for oxidative stress; however, due to COVID-19, analyses of the samples were suspended and the isoprostane data set is limited to only 55 subjects, so this thesis will only include analysis of outcome variables HDL cholesterol and C-reactive protein. C-reactive protein was plotted on a histogram and found to be a skewed distribution. To normalize the distribution, CRP levels were transformed to logarithmic values.

**Exposure variables**

With the goal of measuring individual exposure, ambient air pollution exposure levels were used as a proxy to gauge each household's exposure. The project used an Aethalometer, specifically model AE42 from Magee Scientific company, in order to measure levels of elemental carbon (EC). Ambient PAHs (polycyclic aromatic hydrocarbons) are measured by a valid data technique. $NO_2$ and $NO_x$ were sent to Richmond Field Station testing facilities with special air quality monitors to measure air quality exposure. Exposure was matched to households when participants reported their residential street address, city, and state. Each address was then geocoded using ESRI software or Google Earth, which links to a lifetime, residential history of each participant. Pollutants were measured at different time periods: 1 day, which is average pollutant exposure level the day of study date or office visit when participants had their biomarker level measured; mean week, which is average pollutant exposure the week before study date; 1, 3, 6 month averages, which are the average exposures these number of months before study date; and 1 year average, which is average exposure the year before study date.

**Linear regression models**

To quantify a relationship between biomarker levels and exposure level of air pollutants, regression models were conducted in the statistical programming language R version 3.6.3, using the packages ggplot2, gridExtra, lubridate, mgcv, ResourceSelection, tableone, tidyverse, tinytex. In the analysis, several parameters need to be considered as potential confounding variables: ethnic category, annual family income (as a proxy for socioeconomic status in this population), whether the child lives with a smoker, physical activity, and seasonality changes (Figure 1). This data was collected when each participant was interviewed with a detailed, structured health and general history questionnaire upon arriving at the study center. For the outcome variable HDL cholesterol, a generalized additive model was used, which is essentially a form of a linear regression in which the seasonality variable was related to the outcome via a smooth function, rather than a line. The strength of the associations between measured covariates (race/ethnicity, annual family income, household smoking, physical activity) were also included in the generalized additive models as linear terms. For outcome variable CRP, rather than utilizing the numerical lab value of CRP level, we converted the CRP into a binary variable of either above limit of detection or below limit of detection, because it is normal for children to be below the limit of detection, without seasonality effects expected. The binary CRP level was analyzed in a linear regression model and the estimate is an odds ratio of getting a CRP above limit of detection reading. A Hosmer-Lemeshow test, a goodness of fit test for logistic regression, was conducted to test how well data satisfies model assumptions. If the Goodness of Fit test has a p-value less than 0.05 it means the model does not fit well, indicating that the assumptions of the model would be violated. Almost all logistic regression models for outcome variables have p-values above 0.05, which suggests that the model does fit well. The other covariates as mentioned above (race/ethnicity, annual family income, household smoking, physical activity) were included in the regression model.

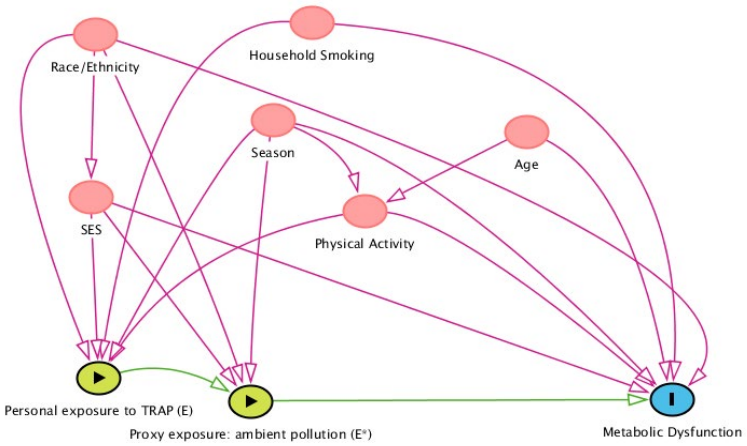


**Figure 1. DAG (directed acyclic graph).** This is the DAG for our regression model. Covariates variables to include in the regression model were determined using a directed acyclic graph.

## RESULTS

### Study population

The study cohort consists of 218 children. 46.8% of the sample is female, and 81.7% is Latinx. This is a sample with low socio-economic status overall; nearly 76% of the study population consists of participants from a family of <$15,000 annual household income, and 70% of the study population do not own a home (Table 1).

**Table 1. Socio-demographic characteristics of 7-year old cohort.** Characteristics include the children's activity level, BMI, and their mother's highest level of education

| Characteristic | # |
|---|---|
| Study population | \218 |
| Girl | 102 |
| Hispanic | 178 |
| Does Not Own Home | 153 |
| Lives with Smoker | 42 |
| CRP below limit of detection | 76 |
| **Physical Activity** | |
| Less Active | 20 |
| About As Active | 135 |
| More Active | 63 |
| **BMI** | |
| Underweight | 2 |
| Normal weight | 110 |
| Overweight | 42 |
| Obese | 64 |
| **Maternal Education** | |
| 8th grade | 0 |
| Some High School | 37 |
| Some High School or GED | 52 |
| Some College | 54 |

## Outcome variables

HDL cholesterol levels in our sample were normally distributed with a median of 50 mg/dL (normal value for children is >45 mg/dL) and C-reactive protein levels were generally low but with a long right tail in the distribution (Quest Diagnostics). Because the normal value for CRP in kids is below the limit of detection (0.3 mg/L) (Quest Diagnostics), CRP lab values were used as below the limit of detection (n=76) and above the limit of detection (n=108) (Table 1).

## Exposure variables

**Table 2. Summary characteristics of air pollution exposure data.** Median, 25th percentile, 75th percentile for pollutant exposures ($NO_2$, $NO_X$, PAH245, EC)

| Pollutant | 1 day average | 1 week average | 1 month average | 3 mo average | 6 mo average | 1 year average |
|---|---|---|---|---|---|---|
| $NO_2$ (ppb) Median | 3.74 | 4.53 | 3.95 | 3.71 | 5.28 | 6.73 |
| 25th %le | 5.37 | 4.62 | 4.52 | 4.04 | 4.75 | 6.88 |
| 75th %le | 3.38 | 3.58 | 4.28 | 3.77 | 4.34 | 6.38 |
| $NO_X$ (ppb) Median | 9.75 | 10.47 | 9.86 | 9.84 | 13.01 | 15.59 |
| 25th %le | 10.06 | 10.35 | 10.54 | 10.23 | 11.78 | 15.8 |
| 75th %le | 7.57 | 8.23 | 9.24 | 8.63 | 9.91 | 13.64 |
| PAH456 ($\frac{ng}{m^3}$) Median | 5.83 | 5.76 | 4.44 | 4.96 | 7.64 | 9.27 |
| 25th %le | 5.31 | 5.45 | 5.28 | 4.28 | 6.33 | 8.51 |
| 75th %le | 5.29 | 5.52 | 5.24 | 4.29 | 5.65 | 8.25 |
| EC ($\frac{\mu g}{m^3}$) Median | 0.39 | 0.59 | 0.48 | 0.4 | 0.39 | 0.53 |
| 25th %le | 0.55 | 0.43 | 0.5 | 0.39 | 0.36 | 0.47 |
| 75th %le | 0.56 | 0.77 | 0.61 | 0.46 | 0.4 | 0.45 |

The highest median exposure was 15.59 ppb of $NO_x$ from the 1-year average estimate (Table 2). Amongst the pollutants, there is most variability in the $NO_x$ exposures with a standard deviation of 11.35. In general, there is less variability with longer exposure windows (Figure 2).

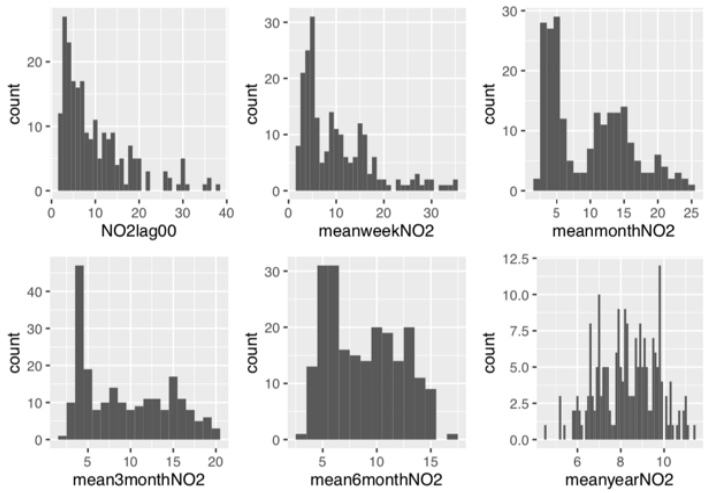


**Figure 2. Distribution of air pollution exposure data.** As a representative example of exposure data, $NO_2$ exposure (ppb) was plotted on histograms. Each exposure pollutant was plotted with 6 representative lag times: 1 day average, 1 week average, 1 month average, 3 month average, 6 month average, and 1 year average.

## Linear regression models

In this cohort, exposure to $NO_2$ was consistently associated with decreases in measured HDL, with the largest effect seen in 3-month average exposure (median = -15.4 mg/dL decrease per IQR increase in $NO_2$, 95% confidence interval -27.4, -3.4) (Table 3). Though not all exposure windows reached statistical significance, there was a consistent pattern of decreased HDL with increased $NO_2$ exposure, $NO_X$ exposure, PAH456 3-month, 6-month, and 1-year exposure, and EC 3-month, 6-month, and 1-year exposure. The estimate of change (the absolute difference) in the HDL cholesterol outcome per interquartile range increase of the pollutant were adjusted for whether or not the child lives with a smoker, whether or not the child is Latinx, physical activity, household income and a smoothed term for the day of the study (Table 3).

**Table 3. HDL cholesterol generalized additive model regression analysis output.** Result estimates are the change of HDL cholesterol values (in units of mg/dL) per intra-quartile range of the pollutant. The threshold for significance is p <0.01. P values below this threshold have been italicized.

| Pollutant | 1 day average | 1 week average | 1 month average | 3 mo average | 6 mo average | 1 year average |
|---|---|---|---|---|---|---|
| $NO_2$ IQRs | 9.4 | 10.1 | 9.7 | 9.3 | 6 | 2.2 |
| $NO_2$ (ppb) | | | | | | |
| Results | | | | | | |
| Estimate | -5.1 | -4.6 | -0.1 | -15.4 | -7.9 | -3.1 |
| 95% CI | (-8.4,-1.8) | (-8.7,-0.6) | (-3.2,3) | (-27.4,-3.4) | (-15.4,-0.4) | (-6.3,0.1) |
| Pvalue | *0.003* | 0.026 | 0.95 | *0.013* | 0.041 | 0.057 |
| % Deviance | 24.4 | 22.4 | 9.9 | 22.6 | 22.6 | 12 |
| $NO_X$ IQRs | 13.1 | 13.4 | 14.7 | 12.6 | 8.7 | 3.5 |
| $NO_X$ (ppb) | | | | | | |
| Results | | | | | | |
| Estimate | -1.2 | -4.2 | -6.8 | -6.7 | -3.6 | -2.9 |
| 95% CI | (-4.1,1.7) | (-8,-0.5) | (-13.9,0.2) | (-13,-0.3) | (-6.6,-0.5) | (-5.4,-0.5) |
| Pvalue | 0.404 | 0.027 | 0.06 | 0.042 | 0.022 | 0.018 |
| % Deviance | 14.7 | 21.8 | 21 | 16.8 | 12.8 | 13 |
| PAH IQRs | 7.7 | 7.9 | 8.4 | 7.9 | 5.2 | 0.8 |
| PAH ($\frac{ng}{m^3}$) | | | | | | |
| Results | | | | | | |
| Estimate | 0.2 | 0.1 | -0.4 | -4.9 | -2.7 | -1.4 |
| 95% CI | (-2.6,3.1) | (-2.9,3.1) | (-3.9,3) | (-17.5,7.7) | (-6.5,1.1) | (-4.3,1.4) |
| Pvalue | 0.869 | 0.957 | 0.81 | 0.448 | 0.163 | 0.327 |
| % Deviance | 9.9 | 9.9 | 9.9 | 14.6 | 11.1 | 10.6 |
| EC IQRs | 0.5 | 0.4 | 0.4 | 0.3 | 0.2 | 0.1 |
| EC ($\frac{\mu g}{m^3}$) | | | | | | |
| Results | | | | | | |
| Estimate | -1.4 | -1.3 | 0.8 | -6 | -3.2 | -1.7 |
| 95% CI | (-4.4,1.5) | (-4,1.5) | (-1.8,3.5) | (-12,0.1) | (-6.2,-0.3) | (-4.2,0.7) |
| Pvalue | 0.33 | 0.366 | 0.536 | 0.054 | 0.034 | 0.172 |
| % Deviance | 14.8 | 14.8 | 10.3 | 16.5 | 12.7 | 11.1 |

For C-reactive protein, no significant association was found with exposure pollutants, but a consistent pattern of increased odds ratio of detectable CRP with increased $NO_2$ 1-month, 3-month, 6-month, and 1-year exposure, $NO_X$ 1-day, 1-month, 3-month, and 6-month exposure, PAH456 exposure for all time windows, and EC 6-month exposure (Table 4). The odds ratio for having a detectable CRP value comparing children whose pollutant values are one interquartile range higher were adjusted for whether or not the child lives with a smoker, whether or not the child is Latinx, physical activity, and household income (Table 4).

**Table 4. C-reactive protein linear regression analysis output.** Results are the estimate of change in the outcome (the odds ratio of getting a CRP above limit of detection reading) per intra-quartile range of the pollutant. The threshold for significance is $p < 0.01$.

| Pollutant | 1 day average | 1 week average | 1 month average | 3 mo average | 6 mo average | 1 year average |
|---|---|---|---|---|---|---|
| $NO_2$ (ppb) IQRs | 9.4 | 10.1 | 9.7 | 9.3 | 6 | 2.2 |
| $NO_2$ (ppb) Results | | | | | | |
| Estimate | 1.2 | 1.4 | 1.1 | 0.6 | 0.6 | 1 |
| 95% CI | (-2.2,2.6) | (-2.1,2.8) | (-2.8,3) | (-3.8,2.8) | (-3.2,2.2) | (-2.7,2.8) |
| Pvalue | 0.286 | 0.129 | 0.748 | 0.347 | 0.129 | 0.892 |
| % Deviance | 20.5 | 22.5 | 20.2 | 20.1 | 20.1 | 21.9 |
| $NO_X$ ($\frac{ng}{m^3}$) IQRs | 13.1 | 13.4 | 14.7 | 12.6 | 8.7 | 3.5 |
| $NO_X$ ($\frac{ng}{m^3}$) Results | | | | | | |
| Estimate | 1.2 | 1.4 | 1 | 0.7 | 0.7 | 1 |
| 95% CI | (-2.3,2.6) | (-2.2,2.9) | (-3.2,3.3) | (-3.2,2.5) | (-2.9,2.2) | (-2.4,2.4) |
| Pvalue | 0.395 | 0.231 | 0.981 | 0.351 | 0.188 | 0.943 |
| % Deviance | 20.2 | 21 | 20.2 | 20 | 22.4 | 22 |
| PAH ($\frac{ng}{m^3}$) IQRs | 7.7 | 7.9 | 8.4 | 7.9 | 5.2 | 0.8 |
| PAH ($\frac{ng}{m^3}$) Results | | | | | | |
| Estimate | 1 | 0.9 | 0.6 | 0.5 | 0.6 | 1.1 |
| 95% CI | (-2.7,2.8) | (-2.9,2.8) | (-3.8,2.6) | (-3.9,2.4) | (-3.3,2.2) | (-2.5,2.7) |
| Pvalue | 0.98 | 0.838 | 0.263 | 0.14 | 0.112 | 0.683 |
| % Deviance | 20.2 | 20.7 | 22.4 | 22.3 | 23.7 | 22.3 |
| EC ($\frac{\mu g}{m^3}$) IQRs | 0.5 | 0.4 | 0.4 | 0.3 | 0.2 | 0.1 |
| EC ($\frac{\mu g}{m^3}$) Results | | | | | | |
| Estimate | 1.2 | 1.3 | 1 | 1 | 0.7 | 1.2 |
| 95% CI | (-2.2,2.6) | (-2.1,2.6) | (-3.7,3.6) | (-2.9,2.9) | (-3.2,2.5) | (-2.4,2.7) |
| Pvalue | 0.439 | 0.142 | 0.964 | 0.999 | 0.384 | 0.601 |
| % Deviance | 19.6 | 22.3 | 59.6 | 20.5 | 22.8 | 21.6 |

To compare the effect of estimates across pollutant time exposure, changes in HDL cholesterol were presented as estimates of 95% confidence interval for an interquartile change in each pollutant (Figure 3). Estimates of CRP are presented as odds ratio of having a CRP above the detection level (Figure 3). In general, the variability in confidence intervals is larger in exposure

time frames that are longer. For instance, in HDL cholesterol outcomes, the pattern of exposure of $NO_2$ has less variability in smaller time exposure windows (1-day and 1-week).

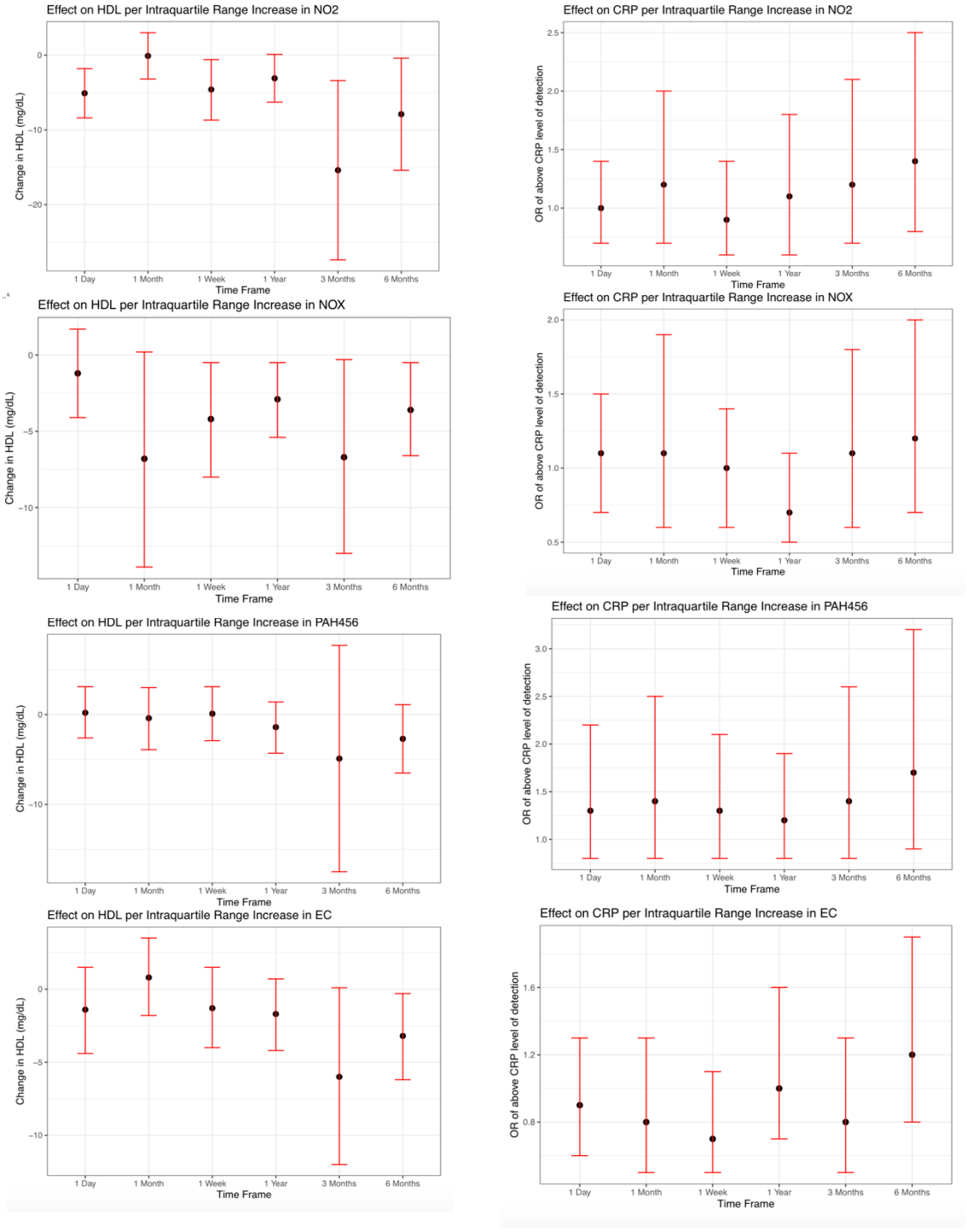


**Figure 3. Estimates of HDL cholesterol change (left) and odds ratio of getting detectable CRP level (right).** Red error bars indicates 95% confidence intervals and estimates are plotted across 6 time frames. * = indicates significant p-value

## DISCUSSION

In this well-characterized 9-year old child cohort, traffic-related air pollution (TRAP) was associated with HDL cholesterol, a biomarker of metabolic dysregulation. There is a significant association between HDL cholesterol and $NO_2$ exposure in the prior day, and a pattern of results that suggest that $NO_2$ may be associated with HDL cholesterol levels across longer time frames as well. Of the pollutants assessed, $NO_2$ had the largest effect on HDL levels. The estimated changes in HDL levels with $NO_x$ exposures were also negative, though with confidence intervals that crossed the null. No trend of decreased HDL cholesterol was observed for PAH456 exposure. For EC exposure, at longer time frames (3-month average and longer)), there is a nonsignificant trend of decreased HDL. These results suggest that there is some association between traffic-related air pollution and decreased HDL.

For C-reactive protein, none of the exposure pollutants showed significant association. However, results across all pollutants suggest that those with higher pollutant exposure may have slightly increased odds of detectable CRP levels. These findings suggest that TRAPs may be associated with lower HDL levels and a higher odd of a detectable CRP. This set of findings is consistent with previous literature that suggests air pollution can lead to damage of airways lung cells that triggers a local inflammatory response, resulting in cytokine release that spills over to the circulatory system (Haberzettl et al. 2016, Miller et al. 2012). In turn, these sites of heightened vascular inflammation are the source of several chronic disorders, including metabolic dysfunction that can contribute to metabolic syndrome or type II diabetes (Hussain et al. 2016). Additionally, lack of protective measures to act against excess reactive oxygen species (ROS) produced by oxidative stress and inflammation can lead to damage of important cellular molecules, such as lipids including HDL cholesterol (Hussain et al. 2016).

### Limitations and Future Direction

There are several strengths of this research study, including the comprehensive and high-quality exposure data, a careful outcome assessment of biomarkers, and a highly exposed study population of vulnerable children of color. On the other hand, some weaknesses include the study being a cross sectional analysis, which makes it difficult to assess causality, and the study

population is a relatively small sample size. It is possible that our participants are from a small area, the part of Fresno served by FUSD, and for this reason seasonal and spatial variability may not be enough to detect association. Additionally, using ambient exposure matched to household and street address as a proxy for individual exposure may contain some estimation error.

Some inconsistencies in data are present in $NO_2$ 1-month exposure analyses with outcome variable HDL cholesterol. The $NO_2$ 1-month exposure estimate for change in HDL cholesterol is close to the null and the confidence interval spans the null. This could be due to lack of variability in exposure or due to physiological factors that influence the outcome, since a 1-month period is neither acute nor chronic exposure. Another explanation would be if $NO_2$ 1-month exposure was not variable enough, but looking at the histogram of exposure, the data is fairly variable (Figure 2), so it is most likely due to degree of exposure.

Future directions include possibly conducting a prospective cohort study of following the data from the 7-year-old children to the 9-year old data set, looking at the same biomarkers of metabolic dysregulation, HDL and CRP. This could help provide a longitudinal view of air pollution exposure in a two-year time frame to investigate the effect of even longer exposure on the study cohort. We also plan to include urinary 8-isoprostane as a marker of oxidative stress in our final analyses when writing the manuscript for a paper that the group plans on submitting to a publication journal, and adding more exposure pollutants, including $PM_{2.5}$, $PM_{10}$, CO and $O_3$.

Overall, our results support the hypothesis that acute exposure to TRAPs primarily impacts inflammation that can affect metabolic dysfunction, including in young children. Low-grade systemic inflammation is associated with metabolic syndrome and is an important factor in instigating premature atherosclerosis (Marsland et al. 2010). For this reason, it is crucial to consider these obesity and metabolic indicators in children, as early life exposure to ambient air pollution has observed associations with later-life cardiometabolic disease (Fleisch et al. 2017, Park 2016).

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## REFERENCES

Eze, Ikenna C et al. "Long-Term Exposure to Ambient Air Pollution and Metabolic Syndrome in Adults." PloS one vol. 10,6 e0130337. 23 Jun. 2015, doi:10.1371/journal.pone.0130337

Bowe B, Xie Y, Li T, Yan Y, Xian H, Al-Aly Z. The 2016 global and national burden of diabetes mellitus attributable to PM2·5 air pollution. Lancet Planet Health 2018

Pearson JF, Bachireddy C, Shyamprasad S, Goldfine AB, Brownstein JS. Association between fine particulate matter and diabetes prevalence in the U.S. Diabetes Care. 2010;33(10):2196-201.

Li W, Dorans KS, Wilker EH, et al. Ambient air pollution, adipokines, and glucose homeostasis: The Framingham Heart Study. Environ Int. 2018;111:14-22.

Hamanaka, Robert B, and Gökhan M Mutlu. "Particulate Matter Air Pollution: Effects on the Cardiovascular System." Frontiers in endocrinology vol. 9 680. 16 Nov. 2018, doi:10.3389/fendo.2018.00680

Qiu H, Schooling CM, Sun S, et al. Long-term exposure to fine particulate matter air pollution and type 2 diabetes mellitus in elderly: A cohort study in Hong Kong. Environ Int. 2018;113:350-356.

Kaufman JD, Spalt EW, Curl CL, et al. Advances in Understanding Air Pollution and CVD. Glob Heart. 2016;11(3):343-352.

Li J, Zhou C, Xu H, et al. Ambient Air Pollution Is Associated With HDL (High-Density Lipoprotein) Dysfunction in Healthy Adults. Arterioscler Thromb Vasc Biol. 2019;39(3):513-522.

Mao S, Li S, Wang C, et al. Is long-term PM exposure associated with blood lipids and dyslipidemias in a Chinese rural population?. Environ Int. 2020;138:105637.

Qiu H, Schooling CM, Sun S, et al. Long-term exposure to fine particulate matter air pollution and type 2 diabetes mellitus in elderly: A cohort study in Hong Kong. Environ Int. 2018;113:350-356.

Park SK, Auchincloss AH, O'neill MS, et al. Particulate air pollution, metabolic syndrome, and heart rate variability: the multi-ethnic study of atherosclerosis (MESA). Environ Health Perspect. 2010;118(10):1406-11.

Peters, A, Döring A, Wichmann HE, Koenig W. Increased plasma viscosity during an air pollution episode: a link to mortality? The Lancet, 1997, Vol. 349 (9065), pp. 1582–87

Yin F, Lawal A, Ricks J, et al. Diesel exhaust induces systemic lipid peroxidation and development of dysfunctional pro-oxidant and pro-inflammatory high-density lipoprotein. Arterioscler Thromb Vasc Biol. 2013;33(6):1153-61.

Nel AE, Diaz-sanchez D, Ng D, Hiura T, Saxon A. Enhancement of allergic inflammation by the interaction between diesel exhaust particles and the immune system. J Allergy Clin Immunol. 1998;102(4 Pt 1):539-54.

Nightingale R, Lesosky M, Flitz G, et al. Noncommunicable Respiratory Disease and Air Pollution Exposure in Malawi (CAPS). A Cross-Sectional Study. Am J Respir Crit Care Med. 2019;199(5):613-621.

Mann JK, Lutzker L, Margolis H et al. *In press*. Traffic-related Air Pollution is Associated with Glucose Dysregulation, Blood Pressure, and Oxidative Stress in Children. 2020.

Fleisch AF, Luttmann-Gibson H, Perng W, Rifas-Shiman SL, Coull BA, Kloog I, Koutrakis P, Schwartz JD, Zanobetti A, Mantzoros CS, Gillman MW, Gold DR, Oken E. Prenatal and early life exposure to traffic pollution and cardiometabolic health in childhood. Pediatr Obes. 2017 Feb; 12(1):48-57.

Christine L. Corton. 2015. London Fog—The Biography. The Belknap Press of Harvard University Press. Cambridge, MA.

Schwartz J, Marcus A. Mortality and Air Pollution J London: A Time Series Analysis. American Journal of Epidemiology. Volume 131, Issue 1. January 1990, Pages 185–194

Miller MR, Shaw CA, Langrish JP. From particles to patients: oxidative stress and the cardiovascular effects of air pollution. Future Cardiol. 2012;8(4):577-602.

Haberzettl, Petra et al. "Exposure to Fine Particulate Air Pollution Causes Vascular Insulin Resistance by Inducing Pulmonary Oxidative Stress" Environmental health perspectives vol. 124,12 (2016): 1830-1839.

Hussain T, Tan B, Yin Y, Blachier F, Tossou MC, Rahu N. Oxidative Stress and Inflammation: What Polyphenols Can Do for Us?. Oxid Med Cell Longev. 2016;2016:7432797.

Barraza-villarreal A, Sunyer J, Hernandez-cadena L, et al. Air pollution, airway inflammation, and lung function in a cohort study of Mexico City schoolchildren. Environ Health Perspect. 2008;116(6):832-8.

Kravitz-wirtz N, Teixeira S, Hajat A, Woo B, Crowder K, Takeuchi D. Early-Life Air Pollution Exposure, Neighborhood Poverty, and Childhood Asthma in the United States, 1990–2014. Int J Environ Res Public Health. 2018;15(6)

Toledo-corral CM, Alderete TL, Habre R, et al. Effects of air pollution exposure on glucose metabolism in Los Angeles minority children. Pediatr Obes. 2018;13(1):54-62.

Park, Sung Kyun. "Ambient Air Pollution and Type 2 Diabetes: Do the Metabolic Effects of Air Pollution Start Early in Life?." Diabetes vol. 66,7 (2017): 1755-1757. doi:10.2337/dbi17-0012

Babatola, Samuel Soledayo. "Global burden of diseases attributable to air pollution." Journal of public health in Africa vol. 9,3 813. 21 Dec. 2018, doi:10.4081/jphia.2018.813

Cohen AJ, Brauer M, Burnett R, et al. Estimates and 25-year trends of the global burden of disease attributable to ambient air pollution: an analysis of data from the Global Burden of Diseases Study 2015. Lancet. 2017;389(10082):1907-1918.

Miller MR, Shaw CA, Langrish JP. From particles to patients: oxidative stress and the cardiovascular effects of air pollution. Future Cardiol. 2012;8(4):577-602.

Du Y, Xu X, Chu M, Guo Y, Wang J. Air particulate matter and cardiovascular disease: the epidemiological, biomedical and clinical evidence. J Thorac Dis. 2016;8(1):E8-E19.

EPA [Environmental Protection Agency]. 2016. Particulate matter (PM10) trends. A Report of the EPA Science Advisory Board (EPA-SAB-11-013). EPA, Washington, D.C., USA.

Brook RD, Rajagopalan S, Pope CA 3rd, Brook JR, Bhatnagar A, Diez-Roux AV, Holguin F, Hong Y, Luepker RV, Mittleman MA, Peters A, Siscovick D, Smith SC Jr, Whitsel L, Kaufman JD; Particulate matter air pollution and cardiovascular disease: An update to the

scientific statement from the American Heart Association. Circulation. 2010 Jun 1;121(21):2331-78.

Sun Q, Yue P, Deiuliis JA, et al. Ambient air pollution exaggerates adipose inflammation and insulin resistance in a mouse model of diet-induced obesity. Circulation. 2009;119(4):538-46.

Rylance S, Nightingale R, Naunje A, Mbalume F, Jewell C, Balmes JR, Grigg J, Mortimer K. Lung health and exposure to air pollution in Malawian children (CAPS): a cross-sectional study. Thorax. 2019 Aug 29.

Marsland AL, Mccaffery JM, Muldoon MF, Manuck SB. Systemic inflammation and the metabolic syndrome among middle-aged community volunteers. Metab Clin Exp. 2010;59(12):1801-8.

Brocato J, Sun H, Shamy M, et al. Particulate matter from Saudi Arabia induces genes involved in inflammation, metabolic syndrome and atherosclerosis. J Toxicol Environ Health Part A. 2014;77(13):751-66.

Haberzettl, Petra et al. "Exposure to Fine Particulate Air Pollution Causes Vascular Insulin Resistance by Inducing Pulmonary Oxidative Stress" Environmental health perspectives vol. 124,12 (2016): 1830-1839.

WHO [World Health Organization]. 2005. Air quality guidelines—global update. WHO, Washington, D.C., USA.

Babatola, SS. "Global burden of diseases attributable to air pollution." Journal of public health in Africa vol. 9,3 813. 21 Dec. 2018, doi:10.4081/jphia.2018.813

Cohen AJ, Brauer M, Burnett R, et al. Estimates and 25-year trends of the global burden of disease attributable to ambient air pollution: an analysis of data from the Global Burden of Diseases Study 2015. Lancet. 2017;389(10082):1907-1918.

Fossati S, Valvi D, Martinez D, et al. Prenatal air pollution exposure and growth and cardio-metabolic risk in preschoolers. Environ Int. 2020;138:105619.

Park SK, Auchincloss AH, O'neill MS, et al. Particulate air pollution, metabolic syndrome, and heart rate variability: the multi-ethnic study of atherosclerosis (MESA). Environ Health Perspect. 2010;118(10):1406-11.

Mazidi M, Speakman JR. Ambient particulate air pollution (PM2.5) is associated with the ratio of type 2 diabetes to obesity. Sci Rep. 2017;7(1):9144.