Prenatal nitrate air pollution exposure and reduced child lung function: timing and fetal sex effects

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Abstract

BACKGROUND: Prenatal particulate air pollution exposure may alter lung growth and development in utero in a time-sensitive and sex-specific manner, resulting in reduced lung function in childhood. Such relationships have not been examined for nitrate (NO₃⁻).

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Take home message:
Airborne nitrate exposure during weeks 6-12 of pregnancy is associated with decreased child lung function in boys
METHODS: We implemented Bayesian distributed lag interaction models (BDLIMs) to identify sensitive prenatal windows for the influence of NO$_3^-$ on lung function at age 7 years, assessing effect modification by fetal sex. Analyses included 191 mother-child dyads. Daily ambient NO$_3^-$ exposure over pregnancy was estimated using a hybrid chemical transport (Geos-Chem)/land-use regression model. Spirometry was performed at mean (SD) age of 6.99 (0.89) years, with forced expiratory volume in one second (FEV$_1$) and forced vital capacity (FVC) z-scores accounting for child age, sex, height and race/ethnicity.

RESULTS: Most mothers were Hispanic (65%) or Black (22%), had ≤ high school education (67%), and never smoked (71%); 17% children had asthma. BDLIMs adjusted for maternal age and education and child’s asthma identified an early sensitive window of 6-12 weeks gestation, during which increased NO$_3^-$ was significantly associated with reduced FEV$_1$ z-scores specifically among boys. BDLIM analyses demonstrated similar sex-specific patterns for FVC.

CONCLUSION: Early gestational NO$_3^-$ exposure is associated with reduced child lung function, especially in boys.

Keywords
nitrate; lung function; prenatal; air pollution; child

INTRODUCTION
Lung development begins in utero and involves carefully timed sequences of biologic events throughout the gestational period that are vulnerable to disruption by environmental toxic exposures. Alterations in the programming of critical processes necessary for optimal structural and functional development of the respiratory system during pregnancy contribute to postnatal pulmonary abnormalities. Identifying prenatal determinants of childhood lung function is critical, as early deficits may increase the risk of future respiratory disease during childhood and into adulthood.

Growing evidence links prenatal exposures to ambient air pollution with adverse respiratory outcomes in children, including reported associations between fine particulate matter (PM) exposure during pregnancy and the development of childhood wheeze and asthma. Studies examining the influence of prenatal exposures upon pulmonary function in children have been limited and have focused on select air pollutants. For example, one study assessing the impact of prenatal ambient air pollution upon newborn lung function found that increased exposure to higher levels of PM less than or equal to 10 micrometers in aerodynamic diameter (PM$_{10}$), both averaged over pregnancy and within each trimester, was associated with significantly higher minute ventilation at 5 weeks of age, with strongest effects observed in the third trimester. More lasting effects of prenatal pollutant exposure were demonstrated by Jedrychowski et al. in a cohort of 176 nonsmoking mothers asked to wear personal monitoring samplers, showing that the highest quartile of personal exposure to PM less than or equal to 2.5 micrometers in aerodynamic diameter (PM$_{2.5}$) assessed over 48 hours in the second trimester of pregnancy was associated with significant deficits in forced vital capacity (FVC) and forced expiratory volume in the first second (FEV$_1$) in children at 5 years of age. Another study of 1295 Spanish mother-child pairs
demonstrated that increased exposures to both ambient benzene and NO\textsubscript{2}, estimated from geographically distributed passive samplers and averaged over the second trimester, were associated with reduced FEV\textsubscript{1} among preschool children.\textsuperscript{13}

These few studies lend support to the hypothesis that prenatal exposure to air pollution may alter \textit{in utero} development of the respiratory system in offspring, resulting in reduced early childhood lung function. However, particulate pollutant exposures considered thus far have been defined by size (e.g., PM\textsubscript{2.5}) and are therefore heterogeneous in composition, and less is known about the specific health effects of components, such as nitrates, within these toxic mixtures. Ambient nitrates (NO\textsubscript{3}\textsuperscript{−}) represent one of the major components of PM\textsubscript{2.5}, originating from the atmospheric oxidation of gaseous nitrogen oxides (NO\textsubscript{x}) emitted from mobile and stationary combustive sources and travelling extended distances to pollute large geographic areas.\textsuperscript{14} Contributing to a significant burden of particulate pollution, ambient nitrate has been specifically associated with health risks including restricted prenatal growth\textsuperscript{16} and other negative birth outcomes;\textsuperscript{17} however, respiratory health effects in children exposed to NO\textsubscript{3}\textsuperscript{−} prenatally have remained largely unexamined. We have recently published the significant association between NO\textsubscript{3}− exposure during gestation with increased risk of asthma in children,\textsuperscript{18} suggesting that this specific PM component may be an important driver of such respiratory health outcomes, and that a greater understanding of the adverse effects of individual pollutants may better inform pollution reduction strategies. However, whether time-sensitive effects of \textit{in utero} nitrate (NO\textsubscript{3}−) air pollution exposure culminate in reduced lung function in childhood is unknown.

Furthermore, studies to date are limited by their arbitrary classification of exposure timing (i.e., averaged over pregnancy or within clinically defined trimesters) rather than coinciding exposure with embryologic stages of respiratory growth and development, which can result in missed or biased findings,\textsuperscript{19} underscoring the need to consider these relationships with a higher degree of temporal specificity. For example, using a data-driven approach to more objectively identify sensitive windows, our group recently demonstrated that prenatal nitrate exposure during two distinct prenatal periods—one early in gestation at 7-19 weeks and the other late in pregnancy at 33-49 weeks—was associated with higher odds of developing asthma in children.\textsuperscript{18} While some of the timed biologic events involved in the programing of asthma during these sensitive windows may overlap with those processes responsible for the attainment of optimal lung function, others may be distinct; therefore, the timing effects of pollutant exposures upon other respiratory outcomes warrant further investigation.

In addition, our prior work has consistently revealed the sex-specific nature of the adverse respiratory effects of prenatal pollution exposure,\textsuperscript{9,18,20} suggesting increased vulnerability in the male fetus. As lung development is well known to progress differently in males compared to females beginning \textit{in utero}, with male fetuses lagging behind females of similar gestational age,\textsuperscript{21} investigations into pollutant-mediated effects upon lung growth and development must consider effects in boys separately from girls, and may reveal exposure-outcome associations within sex strata which are not readily evident in the overall population. Accordingly, in order to examine the impact of prenatal NO\textsubscript{3}− exposure on children’s lung function, we implemented Bayesian distributed lag interaction models (BDLIMs) to estimate windows of vulnerability between prenatal NO\textsubscript{3}− exposure and
childhood pulmonary function measured at age 7 years, accounting for the effect modification of sex.

METHODS

Participants

Pregnant women were participants in the Asthma Coalition on Community, Environment and Social Stress (ACCESS) study, a pregnancy cohort designed to assess the effects of perinatal toxic exposures upon childhood respiratory health. Cohort recruitment procedures have been previously detailed elsewhere\textsuperscript{22} and are summarized briefly here. English- or Spanish-speaking women ≥18 years of age receiving prenatal care at Brigham and Women’s Hospital (BWH), Boston Medical Center (BMC), and affiliated community health centers were enrolled by research staff between August 2002 and July 2009 (mean [standard deviation (SD)] of 28.4 [7.9] weeks’ gestation); 78% of those approached who were eligible agreed to enroll. Of these, 955 gave birth to a singleton live-born infant and continued follow-up. Supplemental funding was obtained to assess lung function in children aged 5 to 8 years, of which 230 children naive to spirometry with a mean [SD] age of 6.99 [0.89] years completed spirometry testing between March 2012 and September 2014 before funding was truncated. Those with acceptable spirometry and complete environmental data were included in the analytic sample (n=191). Those included in the analytic sample did not differ significantly based on maternal education or age, or child sex compared to those in the larger cohort; they were more likely to be Hispanic (Table S1).

The human studies committees at BWH and BMC approved the study, and participants gave written consent in their primary language.

Ambient nitrate exposure

Daily ambient nitrate particle exposure was estimated as described previously.\textsuperscript{18,23} In brief, we used a validated hybrid model of the global 3-D chemical transport model (GEOS-Chem),\textsuperscript{24} which simulates the formation, transportation and deposition of total PM\textsubscript{2.5} and its major components (including airborne nitrates) in the atmosphere, based on emission inventory and meteorological assimilation data. Our hybrid model took the simulation outcome from this chemical transport model, incorporated land-use regression terms and meteorological variables, and calibrated it with ground monitoring data. After model training, the trained model predicted daily exposure at 1 km x 1 km grid cells for each pregnant participant, based on the residence at enrollment (and updated if moved). This hybrid approach allows for both a finer spatial and temporal characterization of patterns of exposure to PM\textsubscript{2.5} components within an urban environment. In order to reduce potential noise caused by day-to-day variation, weekly averages of NO\textsubscript{3}\textsuperscript{−} were used in the analyses.

Lung function outcome

Children underwent spirometric testing in their homes by trained research assistants as described in detail in our prior publication.\textsuperscript{25} To summarize, spirometry was performed at 7 years of age, at which point cooperation with voluntary breathing maneuvers required for reliable testing could be obtained, excluding those participants who reported acute...
respiratory symptoms within the last 3 weeks. A stadiometer measured height to the nearest 0.1 cm, and an electronic scale measured weight to the nearest 0.1 kg. Spirometry was performed according to American Thoracic Society (ATS) guidelines,26,27 with techniques modified for children ≤8 years of age,28,29 using a portable MedGraphics™ laptop-based spirometer which displays real-time flow-volume plots to facilitate testing. Short-acting B-agonists, atropinics, and theophylline preparations were withheld for 4 hours and long-acting B-agonists withheld for 12 hours prior to testing. FVC (liters) and FEV₁ (liters) were recorded from a minimum of 3 (no more than 8) maneuvers. All results from testing were over read to ensure quality control. Spirometry measures, height and weight were all approximately normally distributed. Raw values were adjusted for age, sex, height, and race/ethnicity using multivariable regression and then converted to z-scores (mean= 0, standard deviation of 1), which unlike percent predicted values, reflect each child’s lung function parameter in relation to that of other children in the distribution of our ethnically diverse population.30

Other Covariates

Mother’s age at enrollment and educational status, and child’s race/ethnicity and sex, were self-reported. Child’s gestational age, date of birth, and birth weight were collected from the medical record. Prenatal and postnatal tobacco smoke exposure was assessed based on mother’s response to questions regarding whether the mother or others in the household smoked, either at enrollment or the third trimester, and postpartum, respectively. Mothers reported a child’s diagnosis of asthma during repeated face-to-face interviews at approximately 3-month intervals from birth to the first 24 months, and annually thereafter, in response to the question: “Has a doctor or nurse ever said that your child had asthma?” Of those children with asthma, the majority (78.6%) were given a diagnosis after 3 years of age.31 Infant’s gestational age at birth was calculated using reported last menstrual period, or obstetric estimates based on a second-trimester ultrasonogram if dates differed by more than 10 days on medical record review. Sex-specific birth weight for gestational age (BWGA) z-scores were calculated based on US reference data.32

Statistical analysis

Analysis included 191 mothers and their singleton children who had available daily prenatal NO₃⁻ levels and pulmonary function data, respectively. We restricted our sample to infants born ≥37 weeks gestation to minimize over-adjustment, since prematurity was considered to be a pathway variable based on associations between prenatal air pollution and prematurity,33 as well as between prematurity and low lung function.34 Exposure estimates of the remaining days of a 40-week gestation period for those infants (n=63) born between 37 and 40 weeks gestation were based on post-natal nitrate estimates corresponding to this time.

In order to identify sensitive windows during the gestational period where prenatal NO₃⁻ is associated with reduced lung function parameters, we estimated the time-varying association of each mother’s average weekly exposures throughout pregnancy. Bayesian distributed lag interaction models (BDLIM) were employed to first determine the overall association between prenatal NO₃⁻ and lung function, and subsequently examined effect modification by fetal sex. BDLIM is equivalent to a traditional DLM when estimating the overall
association, but extends the DLM framework to allow for modification of either the timing of the window, within-window effect, or both. By partitioning the distributed lag function into two components—weights that identify sensitive windows of susceptibility and coefficients that estimate the magnitude of within-window effects—BDLIM allows for situations where subgroups have the same sensitive windows (weights) but different within-window effects or vice versa, unlike the standard DLM framework. If the weights are constant throughout time, the BDLIM is equivalent to a model with mean pollutant exposure interacted with child sex. When the weights vary with time, the model identifies time period with greater weight (i.e. sensitive windows) that graphically appear as a bump where nitrate is associated with reduced lung function. In addition to determining sensitive windows, we employed BDLIM to estimate the cumulative effect of NO$_3^-$ exposure over the entire pregnancy for each sex-specific subgroup, accounting for identified group-specific sensitive windows and within-window effects, as previously described.$^{35}$

All models included the covariates already accounted for in deriving the spirometry measure z-scores (child’s age, sex, height, race/ethnicity), and additionally adjusted for potential confounders linked to nitrate exposure and lung function including maternal age and education level and child’s asthma diagnosis. Finally, we conducted sensitivity analyses additionally adjusted for BWGA z-score, a potential pathway variable. All analyses were implemented in R statistical software (v3.3.1, Vienna, Austria).

**RESULTS**

The majority of mothers were Hispanic (65%) or Black (22%), had ≤12 years of education (67%), and reported no pre- or post-natal smoking (71%) - characteristics which were evenly distributed over infant sex (Table 1). Girls had similar birth weight for gestational age z-scores compared to boys (Wilcoxon rank sum test p>0.4). Thirty-three children (17%) had physician-diagnosed asthma, with more boys than girls having asthma (27% vs. 7%, p<0.01). Average (median [interquartile range (IQR)]) ambient prenatal NO$_3^-$ concentration was 1.33 [1.15-1.49] μg/m$^3$ and was similar for both sexes.

**Overall association between prenatal NO$_3^-$ and childhood lung function**

Adjusted BDLIMs were first used to estimate the overall associations between weekly NO$_3^-$ during gestation and lung function. While no statistically significant sensitive window was identified in the overall population, the shape of the associations across gestation suggests an early window in pregnancy during which higher exposure to nitrates was associated with reduced FEV$_1$ (Figure 1a). Similar relationships were identified for individual associations between NO$_3^-$ and FVC (Figure 2a), FEF$_{25-75}$ (Figure S1a), and FEV$_1$/FVC (Figure S2a). The estimated cumulative effect over the entire gestational period, accounting for both sensitive windows and within-window effects, demonstrated a reduced FEV$_1$ (mean change [95% CI] = −0.10 [−0.66, 0.44] liters (L) and FVC (mean change [95% CI]= −0.27 [−0.73, 0.17] L), associated with a 1 μg/m$^3$ increase in NO$_3^-$, though this did not reach statistical significance.
Effect modification by fetal sex

Figure 1b demonstrates results from a BDLIM analysis of the association of prenatal ambient nitrate exposure and FEV$_1$, accounting for effect modification by child sex. We found that the sensitive window suggested in the overall population was more pronounced in boys, with a statistically significant window identified at 6-12 weeks of gestation. Similar contours of the associations during gestation were observed for FVC, FEF$_{25-75}$, and FEV$_1$/FVC in boys (Figures 2b, S1b, S2b, respectively). No sensitive window was found between prenatal nitrate exposure and decreased lung function among girls, including for either FEV$_1$ or FVC. The BDLIM analysis suggested that effect modification by sex was attributable to difference in the magnitude of the within-window association, while the window was not different between the two sexes (the normalized posterior density was 0.72 for FEV$_1$ model, interpreted as the probability that this was the best fitting pattern of effect modification). Lastly, sensitivity analyses for models that included adjustment for BWGA z-score did not yield substantive changes in our findings (data not shown).

DISCUSSION

This is the first prospective study to combine advanced statistical modeling with highly temporally resolved ambient NO$_3^-$ exposure estimates to link susceptible windows of prenatal NO$_3^-$ exposure with early childhood lung function. Our results not only add to a growing literature linking prenatal ambient air pollution exposures with childhood respiratory outcomes, but represent a key advancement in more objectively determining sensitive time windows within pregnancy that are most susceptible to specific environmental insults such as nitrate pollution. By leveraging weekly-averaged nitrate exposure data across pregnancy and applying advanced statistical modeling based on the data collected rather than pre-defined time periods, estimates of time-varying associations with childhood lung function coincide with specific embryologic stages of respiratory system development. Even within our relatively small sample size, our analyses revealed that increased nitrate exposure over 6-12 weeks of gestation was significantly associated with reduced FEV$_1$ in young school-aged children, particularly among boys. Similar reductions in FVC consistently trended within the same gestational window, highlighting that nitrate exposure during this early period of rapid development may alter critical processes involved in achieving the full potential of lung growth by early childhood. Our prior findings demonstrating an association between prenatal nitrate exposure and increased child asthma risk also corresponded to an early stage of lung development inclusive of this critical window.\textsuperscript{18} Such overlap of these sensitive windows during pregnancy may suggest pathophysiologic processes common to the development of asthma and lung function deficits, reinforcing the capability of early nitrate pollutant exposures to influence respiratory programming across the life course. Overlaying identified windows of exposure upon our current knowledge about the progression of developmental events \textit{in utero} may provide a more definitive understanding of the effects of air pollutants on offspring outcomes and can inform future studies investigating the underlying cellular and physiologic mechanisms being perturbed in these settings.
Several mechanisms have been proposed to explain the effects of *in utero* environmental exposures upon lung development, and highlight the importance of timing in altering the course of fetal growth trajectories. The early window of vulnerability identified in our results coincides with the pseudo-glandular stage of lung development, which is responsible for the formation of supporting structures of the airway, including smooth muscle, mucosal glands, and vasculature responsible for exchange of oxygen and nutrients. We hypothesize that disruptions by airborne toxins experienced specifically during this embryologic time point may restrict subsequent gestational lung growth by compromising the formation of these supporting structures and nutritional availability early on, thereby failing to sustain adequate growth for the rest of the developmental period *in utero* and hindering the achievement of maximal lung function. For example, pollutant-mediated restriction of pulmonary angiogenesis during the pseudo-glandular period may in turn result in fetal hypoxia, one proposed mechanism underlying compromised lung development. In addition, one can consider the possibility that interruption of early airway smooth muscle development occurring during this lung stage may contribute to altered downstream lung compliance. Indeed, Tang et al. demonstrated in animal studies that one month-old rats exposed to PM$_{2.5}$ *in utero* had post-natal lungs with significant decreases in alveolar size and inspiratory lung volumes, as well as changes in elastic recoil resulting in decreased lung compliance. These changes corresponded to histologic evidence of interstitial proliferation and up-regulation of epithelial-mesenchymal transition (EMT) pathways necessary for repair of lung injury. EMT may therefore play a role in pathologic changes that occur in response to oxidative stress insults, leading to progression of pulmonary dysfunction in the young developing fetus. While to our knowledge there are no such animal studies examining the effects of prenatal nitrate exposure, similar disruption of morphologic patterns affecting lung function may occur during exposure to airborne nitrates as well, and further studies are needed to explore the mechanisms behind these developmental events.

A key contribution of our work is our finding that the association of prenatal nitrate exposure to lung function deficits is sex-specific, with males being most vulnerable. Determining the subgroup that was most vulnerable to NO$_3^-$ was achieved through BDLIM, which allows for the evaluation of interactions by fetal sex, while taking account of the sensitive window identified. This sex-specific effect is consistent with numerous studies in the literature that demonstrate increased susceptibility to the adverse effects of prenatal air pollution exposure, including PM$_{2.5}$ and nitrate pollution, though the reasons for these differences are unclear. Human studies have detected sex differences in lung development starting in early gestation, with the male fetus lagging behind in the structural, histologic, and functional maturity of the respiratory system, as well as having a relative lack of protective mechanisms such as surfactant production, compared to gestational age-matched females. While the biologic mechanisms behind male vulnerability to air pollutant exposures remain to be elucidated further, animal studies have demonstrated sex-specific developmental changes in experimental models of other perinatal toxic insults, for example greater structural deficits in alveolarization and lung angiogenesis in newborn male pups exposed to hyperoxia compared to their female counterparts. Therefore, our results add further human data to the growing evidence of sexual dimorphism in the alteration of lung development occurring under the influence of environmental exposures.
This study has several novel strengths. Prenatal ambient nitrate levels for each mother were assessed daily over the 40-week span of gestation using a novel hybrid chemical transport/LUR model calibrated at a 1 x 1 km grid based on mother’s residence during pregnancy, resulting in both high temporal and spatial resolution of the exposure. We then leveraged these pollutant estimates to apply data-driven, advanced statistical methods to objectively identify critical windows of susceptibility within pregnancy in an ethnically mixed urban population considered vulnerable to pollutant effects. Finally, to our knowledge, this is the first report to investigate sex-specific effects of a key component of particulate air pollution on childhood lung function. Moreover, the identification of a sensitive window that is significantly associated with reduced lung function lends credence to the idea that there is a critical period during gestation during which these toxic exposures can have a lasting effect on lung growth. Whether these nitrate-associated effects are irrevocable changes that persist beyond early childhood is unclear, and further longitudinal studies of NO₃⁻ exposure with repeat lung function measures can shed light to the permanence of these consequences.

There are also notable limitations to our study. In contrast to observations regarding the sensitive window identified among boys, the association between cumulative nitrate exposure over gestation and lung function was not significant in the sample as a whole. This likely reflects the need to account for child sex differences in vulnerability although it also may reflect inadequate power to detect an effect given the moderate sample size. Thus, these associations should be explored further in larger cohorts. We do not adjust for indoor air pollution, which is a significant exposure, particularly in inner-city homes. Studies also find that variations in indoor source particles are largely uncorrelated with variations in outdoor source particles.

Thus, although particles of indoor origin are an important predictor of respiratory health in and of themselves, they are unlikely to confound associations between ambient particulate matter and asthma. Of note, in sensitivity analyses adjusting for tobacco smoke exposure (a key contributor to urban indoor air pollution), our results were unchanged. Due to the sample size, our models did not allow statistical consideration of multi-pollutant effects, such as interaction with other types of particulate air pollutants. In addition, there may be other unaccounted environmental influences or toxins that co-vary with nitrate pollution to result in the link with reduced lung function. Future work in larger samples should consider multi-pollutant models to account for potential synergistic effects with other particulate sources of ambient pollution. Finally, our participants reflect a high-risk demographic of mothers and children, and results may not be generalizable to other populations within the US or the world.

In conclusion, boys born to mothers with higher prenatal NO₃⁻ exposure in the first trimester, specifically between 6 to 12 weeks gestation, were more likely to have reductions in both FEV₁ and FVC at 7 years. These findings not only have significant implications for health of children, but also for lung health across the lifespan, as early life lung function has been demonstrated to predict adult respiratory disease. In light of the accumulating evidence linking prenatal air pollution and early respiratory morbidity, public policies aimed at reducing environmental exposures among pregnant women may have meaningful...
influence upon promoting optimal lung development and preventing respiratory illness in later life. Targeted interventions to minimize toxic exposures in utero may have even greater impact on lung function than those that have been previously demonstrated for improvements in ambient pollution exposure achieved during childhood. 50

**Supplementary Material**

Refer to Web version on PubMed Central for supplementary material.

**ACKNOWLEDGMENTS**

Funding sources: Supported by National Institute of Health (NIH) grants R01 ES010932, U01 HL072494, and R01 HL080674 (R.J.W., principal investigator [PI]) for the Asthma Coalition on Community, Environment, and Social Stress project; the CLEAN Air Center dedicated to air pollution estimates was funded by U.S. Environmental Protection Agency grant RD 83479801, and phenotyping and biostatistical support was funded by NIH P30 ES023515 (R.O.W., PI), P30 ES000002 (B.A.C., PI of Biostatistics Core), and National Institute of Environmental Health Sciences grant T32ES007142 (B.A.C. [PI]). Funding sources had no role in the writing of the manuscript or decision to submit for publication. The corresponding author had full access to the data in the study and had final responsibility for the decision to submit for publication.

All research was approved by the Partner’s Health Care Human Research Committee.

**References**


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<td>- Prenatal nitrate exposure is associated with reduced lung function at age 7 years</td>
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<td>- Link between prenatal nitrate exposure and lung function deficits is male-specific</td>
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<td>- Significant effects occur during early sensitive window of 6-12 weeks gestation</td>
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Figure 1. Association between weekly ambient nitrate (NO$_3^-$) over gestation and childhood lung function: FEV$_1$ z-score.
Bayesian distributed lag interaction models (BDLIM) were used to estimate week-specific effects for (a) overall sample and (b) interaction by sex. Models were adjusted for maternal age and education level and child’s asthma diagnosis, and z-scores were adjusted for child’s age at spirometry, sex, height, and race/ethnicity. The x-axis demarcates the gestational age in weeks. The y-axis represents the change in FEV$_1$ z-scores corresponding to each $\mu$g/m$^3$ increase in NO$_3^-$. Solid lines represent the predicted time-varying change in lung function and gray areas indicates the 95% confidence intervals (CI). A sensitive window is identified when the estimated point-wise 95% CI does not include zero.
Figure 2. Association between weekly ambient nitrate (NO$_3^-$) over gestation and childhood lung function: FVC z-score.
Bayesian distributed lag interaction models (BDLIM) were used to estimate week-specific effects for (a) overall sample and (b) interaction by sex. Models were adjusted for maternal age and education level and child’s asthma diagnosis, and z-scores were adjusted for child’s age at spirometry, sex, height, and race/ethnicity. The x-axis demarcates the gestational age in weeks. The y-axis represents the change in FVC z-scores corresponding to each μg/m$^3$ increase in NO$_3^-$ . Solid lines represent the time-varying predicted change in lung function and gray areas indicates the 95% confidence intervals (CI). A sensitive window is identified when the estimated point-wise 95% CI does not include zero.
## ACCESS participant characteristics (n=191)

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</tr>
<tr>
<td>z-score of FEV₁ (mean, SD) ‡</td>
<td>0.02 0.98</td>
</tr>
<tr>
<td>z-score of FVC (mean, SD) ‡</td>
<td>−0.003 1.01</td>
</tr>
<tr>
<td>z-score of FEV₁/FVC ratio (mean, SD) ‡</td>
<td>−0.002 1.01</td>
</tr>
<tr>
<td>z-score of FEF₂⁵⁻⁷⁵ (mean, SD) ‡</td>
<td>0.04 0.96</td>
</tr>
</tbody>
</table>

* Averaged over entire pregnancy
† Combination of prenatal maternal smoking and postnatal household smoking status
Adjusted for age, sex, height, race