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Effects of In Utero and Early Life $PM_{2.5}$ Exposure on Disease Risk Among Infants and Young Children

DISSERTATION

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by

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DEDICATION

To

Soraya and Samir Girguis
My parents, my foundation, my inspiration.
Pishoy Haroun
My partner, my support, my encouragement.

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ABSTRACT OF THE DISSERTATION

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By

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Doctor of Philosophy in Public Health

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Particulate matter with a diameter of 2.5 microns or less ($PM_{2.5}$), a surrogate estimate of traffic-related air pollution, is widespread and a risk to public health. In-utero exposures to $PM_{2.5}$ may increase risk of birth defects, the leading cause of infant death during the first year of life in the United States, while early life exposures may increase risk of morbidities, such as bronchiolitis, the leading cause of infant hospitalizations, or otitis media (OM), the most common childhood infection. Infants may be more susceptible to negative $PM_{2.5}$ effects because they are more likely to be active, breathe more air per pound of body mass, and are still developing. The effects of in utero and early life $PM_{2.5}$ exposure on the risk of birth defects and infant bronchiolitis and OM are assessed among all births from 2001-2009 using the Pregnancy Early Life Longitudinal Data System (PELL), a Massachusetts birth cohort that has been linked to all subsequent records of clinical encounter. $PM_{2.5}$ exposure models were based on data from satellite remote sensing, which provide extensive spatial coverage throughout Massachusetts. Findings suggest in utero $PM_{2.5}$ exposure during specific critical windows of exposure may be associated with risk of specific cardiac defects. Acute early life $PM_{2.5}$ exposure was associated with risk of infant

bronchiolitis and OM clinical encounters, especially among preterm infants. Investigations of chronic $PM_{2.5}$ exposure indicate null associations with infant bronchiolitis and OM clinical encounters, even after rigorous control for confounding.

INTRODUCTION

This work investigates the relationships between in-utero and early life exposure to ambient particulate matter with a diameter of 2.5 μm or less ($PM_{2.5}$) and risk of birth defects and infant morbidity using a population based cohort. It combines satellite remote sensing data for modeling of $PM_{2.5}$ and advanced statistical methods for assessing spatial and temporal variability within the geocoded Pregnancy to Early Life Longitudinal (PELL) cohort. PELL is a population-based integrative data system consisting of multiple linked databases that can be used for longitudinal analyses of infant health outcomes in Massachusetts (MA). This analysis includes MA live births exposed from 2001 through 2009, including 200,000 linked sequential sibling groups. The first chapter examines the associations between in-utero $PM_{2.5}$ exposure, based on maternal residence at time of birth, and the risk of cardiac, neural tube, and oral facial birth defects are assessed. The effects of chronic and acute early life $PM_{2.5}$ exposures on infant bronchiolitis, the leading cause of infant hospitalization, and otitis media (ear infection), the most common childhood infection are assessed in the second chapter. A case-cross over epidemiological design is used to investigate the association between short term $PM_{2.5}$ exposures and clinical encounters of infant bronchiolitis and otitis media. Such a study design controls for confounding by temporal variables. Short term lags are included to determine the influence of $PM_{2.5}$ on exacerbation of existing symptoms and extended lags will inform whether $PM_{2.5}$ modifies susceptibility to infection. In the third chapter, to understand the influence of chronic early life $PM_{2.5}$ exposure, a nested case-control design is utilized. Separate analyses are conducted with two different control groups: matched controls by date of birth and gestational age and sibling controls to aggressively control for individual level confounding. For an

exposure as widespread as ambient $PM_{2.5}$, this study has the potential to make an important contribution to the field of environmental epidemiology with a significant public health impact.

OVERVIEW OF THE DISSERTATION

The following three chapters demonstrate research on risk of morbidity among infants and young children and traffic-related air pollution exposure during various time periods of the early life course. The dissertation is divided into in-utero and early life exposure. In-utero exposure was examined in relation to risk of birth defects while early life exposure was examined in relation to risk of infant bronchiolitis and otitis media. Early life exposures are further divided into acute exposure and chronic exposure.

The aim of chapter 1 is to determine the association between in-utero $PM_{2.5}$ exposures and risk of birth defects. Case-control studies are used to investigate the association between critical developmental windows of exposure to $PM_{2.5}$ and risk of cardiac, oral facial, and neural tube birth defects. Secondary case-control studies are used to investigate the association between crude measures of traffic-related air pollution, such as distance to major roadways and road density, and risk of cardiac, oral facial, and neural tube birth defects. Lastly, generalized additive models (GAMs) are used to examine spatial-temporal patterns of cardiac, oral facial, and neural tube birth defects risk.

Chapter 2 aims to investigate the effect of acute early life $PM_{2.5}$ exposures on the risk of infant bronchiolitis and otitis media. The associations between short-term $PM_{2.5}$ exposure and infant bronchiolitis and otitis media using case-crossover studies with short and long term lags for exacerbation of symptoms and susceptibility of infection, respectively are determined. Further, effect modification is assessed by season, gestational age, birth weight, hospitalization

frequency, insurance payer code, infant sex, age of infant at time of clinical encounter, insurance, income, maternal race, and breastfeeding status to identify susceptible subgroups in secondary analyses.

Chapter 3 aims to determine the effect of chronic early life $PM_{2.5}$ exposures on the risk of infant bronchiolitis and otitis media. Associations between long-term $PM_{2.5}$ exposure and risk of infant bronchiolitis and otitis media are estimated using time and health matched case-control studies. Additionally, sibling matched case-control studies are used to account for unmeasured confounding by family. A secondary analysis is conducted of only siblings with the same birth location.

Exposure of Interest

Motor vehicle exhaust is a widespread major source of urban air pollution. As the mixture of traffic-related air pollutants are difficult to measure, residential proximity to major roadways and cumulative traffic density within a buffer area have traditionally been used as indirect estimates of traffic-related exposure (1-2). Although imprecise, they account for traffic volume and distance which influence air pollution gradients near roads. Traffic emissions diminish to near background levels within 150-300 meters of the primary source (3). Many studies have found that living close to major roads or in areas of high traffic density is associated with adverse respiratory (4-7), cardiac (8-10), and pregnancy outcomes (11-12).

A more direct estimate of traffic-related air pollution is through the measurement of $PM_{2.5,}$ a complex mixture of small particles and liquid droplets from vehicle exhaust emissions, industrial activities, and coal and wood burning. Depending on its source, $PM_{2.5}$ is composed of a combination of acids, such as sulfates and nitrates, metals, organic matter, dust and dirt. The U.S.

Environmental Protection Agency (EPA) calculated the public health burden attributable to $PM_{2.5}$ in the United States, finding that the percentage of ischemic heart disease mortality associated with $PM_{2.5}$ exposure was as high as 11% in some counties (in New York) (13). Globally in 2013, over 2.1 million $PM_{2.5}$-related deaths occurred (14). In addition, exposure to $PM_{2.5}$ has been associated with increases in hospital admissions, doctor and ER visits, medication use, and school and work absences due to heart and lung disease (14-16). These findings demonstrate that despite recent improvements in air quality through reduced coal burning and vehicle emissions, current levels of $PM_{2.5}$ still pose a risk to public health. It is suspected that $PM_{2.5}$ is also responsible for lung cancer deaths, infant mortality, and adverse developmental outcomes among infants including low birth weight (16-19). To date, there have been very few studies focusing on the effects of $PM_{2.5}$ on the fetus, infant, or young child. This study aims to investigate the effects of in-utero and early life $PM_{2.5}$ exposure on risk of birth defects and infant morbidity among Massachusetts (MA) births exposed from 2001-2009.

Epidemiology and Etiology of Birth Defects

Birth defects occur in 3% of all US born infants (20-22). Cardiac, oral facial cleft and neural tube defects are among the most common anomalies observed (21). In this research, which utilizes a MA detailed data linking system, PELL, birth defects occur in 2% of all live and stillbirths. Although rare, birth defects are the leading cause of infant death within the first year of life (20). Cardiovascular defects are the most common defect type both in MA and nationally. Infants born with birth defects have increased risk of negative health outcomes later in life and often need surgery and further medical treatment. Hospitalizations for birth defects cost the U.S. over $2.6 billion annually (22). This cost is higher when including the financial and emotional

impact of living with birth defects. The average cost of care for an infant with a cardiac anomaly is estimated to be $18,600 per hospital stay (20)**.** To date, the cause of many birth defects is unknown. Early studies determined that exposure to tobacco smoke among pregnant women significantly increases risk of various birth defects (23-33). Multiple meta-analyses have been conducted to confirm these findings (34-37). Male sex, moms greater than 35 years of age, and alcohol consumption have also been associated with birth defects (34).

Cross sectional studies in Eastern Europe were the first to indicate an association between exposures to traffic-related air pollution and risk of birth defects (38-39). Subsequent observational studies that have examined the association between ambient air pollution and risk of birth defects (40-54) have not yielded clear results, in part due to differences in methodology, background $PM_{2.5}$ levels, and populations (55). Among birth defect studies that included $PM_{2.5}$ in their analysis (49,51-54), some have found null associations between $PM_{2.5}$ exposure and birth defects (48,52,54), although a few studies have found significant increased associations (54) while other have found protective (53-54) associations.

Epidemiology and Etiology of Infant Bronchiolitis

Bronchiolitis, a lower respiratory tract infection, is the leading cause of hospitalizations among children during the first year of life (56). Most bronchiolitis cases are caused by respiratory syncytial virus (RSV) infection but there are other viruses which can cause bronchiolitis. Other viruses that can cause bronchiolitis include: Adenovirus, Influenza, and Parainfluenza Virus. Although most infants are RSV seropositive, there exists a large disparity in symptoms and severity exhibited. Bronchiolitis begins with a mild upper respiratory infection and subsequent symptoms are experienced. These symptoms include lack of oxygen (cyanosis),

breathing difficulty, cough, fatigue, intercostals retraction, and tachypnea. Some infants experience mild symptoms while others are hospitalized (57).

In MA, the annual hospitalization rate for bronchiolitis among infants less than 1 year of age is 2.1 per 1,000 and the annual rate of emergency department (ED) visits is 4.6 per 1,000. Exposures such as indoor wood burning and environmental tobacco smoke have been associated with increased risk of hospitalization for bronchiolitis (58-59). Other risk factors include premature birth, small for gestational age, male sex, low socioeconomic status (SES), genetics, and sibling crowding (50). Children who suffer from bronchiolitis during infancy are at increased risk for asthma later in life (60), but it is currently unclear whether bronchiolitis infection is a causal factor in the development of asthma or if the illness shares a common underlying cause. Although the literature on infant bronchiolitis and $PM_{2.5}$ is limited, it is suggestive of a possible association (61-62). Analysis in areas of relatively high $PM_{2.5}$ background levels, such as Los Angeles, have found positive associations with risk of bronchiolitis and increased lifetime $PM_{2.5}$ exposure (63). Studies in areas with low background $PM_{2.5}$ concentrations found no association between $PM_{2.5}$ exposure and bronchiolitis risk (64-65). Bronchiolitis hospitalization rates have been shown to peak in winter (61) corresponding to $PM_{2.5}$ cycles. Approximately 90% of bronchiolitis cases occur during cold months. Premature infants have been identified as a susceptible subgroup (62). A positive correlation has been found between bronchiolitis and acute $PM_{2.5}$ exposure in a retrospective study in Italy (66).

Epidemiology and Etiology of Otitis Media

Otitis media (OM), or inflammation of the middle ear, is one of the most frequent childhood infections among children less than 3 years of age (67), the most common cause for

medical care besides a healthy child visit, and a major cause for antibiotic use within the first few years of life (68). Sixty percent of infants will have experienced at least one episode of OM by one year of life (69). The estimated annual cost related to OM in the U.S. is $3.8 billion (70). In MA, the annual hospitalization rate for OM among children less than 3 years of age is 1.1 per 1,000 and the annual rate of ED visits is 13.8 per 1,000. Risk factors include male sex, young infants, short breast feeding durations, eustachian tube dysfunction and cleft palate, low SES, genetics, and sibling crowding (68). Seasonal variations have also been reported with OM incidence, with increases in winter and fall (70) that correspond to $PM_{2.5}$ variations. OM can lead to hearing loss which can impact speech and delay cognitive development. Much like bronchiolitis, OM is typically caused by a viral infection and environmental exposures such as tobacco smoke and indoor wood burning have been implicated in the etiology of disease (70-73). Currently, there is little literature on the association between OM and $PM_{2.5}$. One study found that there was an association between lifetime exposure to $PM_{2.5}$ and OM (74). Even in geographic locations of relatively low $PM_{2.5}$ levels, increased exposure to $PM_{2.5}$ two months prior to the physician visit was associated with risk of OM (75). Studies investigating the association between other traffic-related air pollutants, such as nitrogen oxide and benzene, also have found positive associations (76-77).

Significance of Research

This research addresses many of the weaknesses of earlier $PM_{2.5}$ studies. To improve on previous studies, exposure is more accurately assessed. Instead of linking infant zip codes to data from the closest $PM_{2.5}$ ground monitor, this study will use $PM_{2.5}$ concentrations at the infant's geocoded birth addresses, measured by satellite, which will reduce exposure misclassification

(78-79). The use of satellite-based $PM_{2.5}$ measures allows for defect-specific critical windows of exposure and lag periods to assess the relationship between in-utero exposures and risk of birth defects and infant morbidity. Additionally, the PELL cohort includes nine years of data that links all MA births to hospitalizations and emergency department visits, providing a large study population for case ascertainment and allowing for a case-crossover analysis of short-term exposures and sibling and matched controls for case-controls studies of long-term exposures.

There is currently limited knowledge of risk factors associated with birth defects. Using spatio-temporal epidemiologic analyses, the influences of residential location, in-utero $PM_{2.5}$ exposure, and spatial confounding associated with birth defect risk is assessed in MA. By accounting for space and time variables, spatially varying risk factors are visually assessed by comparing geographic patterns with and without specific risk factors. Additionally, existing studies have not examined the effects of both chronic and acute postnatal $PM_{2.5}$ exposures and infant bronchiolitis and OM among infants. Children who are brought into the emergency department include the most severe cases of bronchiolitis and OM and represent a large financial burden in terms of healthcare costs. By utilizing various epidemiological methods to assess the relationship between in-utero $PM_{2.5}$ exposures among the most severe cases, this study is able to successfully characterize various aspects of risk including symptom exacerbation (or severity), and susceptible subgroups.

The significance of this is supported by growing toxicologic and epidemiologic evidence that ambient $PM_{2.5}$ exposure increases potentiation of the disease process, through inflammatory pathways in the body. Since ambient $PM_{2.5}$ is a common exposure, associations determined from this research can have a significant public health impact. Birth defects, infant bronchiolitis, and

OM all have serious long-term consequences and any reductions in these adverse outcomes would have important public health implications.

Innovation

This research seeks to understand the effects of $PM_{2.5}$ exposure during various time periods of the early life course. Critical windows of exposure are assessed during the prenatal period that corresponds to specific birth defect risk. Additionally, critical windows of exposure are assessed according to date of clinical encounter to evaluate the influence of $PM_{2.5}$ on severity of infant bronchiolitis and otitis media. Both chronic (lifetime) and acute early life exposures are investigated in respect to otitis media and infant bronchiolitis risk.

An innovative aspect of this research includes using PELL to study these outcomes. In addition, the use of remote sensing data to characterize $PM_{2.5}$ exposures is a novel component of this work. All addresses have been geocoded to the street level, which is an improvement to the existing studies that currently assign zip code-level exposures. Typically, such investigations of traffic-related air pollution have utilized ground monitors and land use regression to measure $PM_{2.5}$ exposure. Instead, this study uses remote sensing data with novel spatial statistics to model exposure. Satellite driven statistical models are advantageous (79) as they provide continuous spatiotemporal coverage over a large geographic region for multiple years. This research uses satellite based aerosol optical depth (AOD), a direct measurement of fine particle abundance in the atmospheric column, at a resolution of 4km. $PM_{2.5}$ exposure models are obtained from the US EPA Clean Air Center at Emory University.

Investigators have found that when studying birth defects, associations are biased due to broad birth defect groupings which are composed of defects with differing etiologies (80).

International Classification of Diseases, Ninth Revision, Clinical Modification (ICD-9-CM) codes are used to investigate anatomical specific birth defect groupings. In addition to anatomical groupings, the 5 most common birth defects are also assessed to determine if there is an association between in-utero $PM_{2.5}$ and the most frequently diagnosed birth defects. Furthermore, targeted critical windows of exposure based on what is known of the birth defect etiology are used in addition to traditional trimester-long exposure periods, which have previously been used when investigating birth defects.

Registry based analyses are often prone to residual confounding due to limited information available from birth records (81). This research accounts for extensive confounders utilizing extensive information available through PELL. Such confounders include demographics, prenatal care, medical history, and geographic information. The case crossover study adjusts for all factors which do not vary over the short term including other air pollutants, tobacco smoke, and home crowding. The sibling control study adjusts for any family level confounders, such as the proclivity to use the fireplace during the winter or second hand smoke tobacco exposure. In addition, novel spatial analysis methods identify geographic patterns that may indicate the presence of residual confounding.

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CHAPTER 1
Maternal Exposure to Traffic-Related Air Pollution and Birth Defects in Massachusetts

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ABSTRACT

Exposures to particulate matter with diameter of 2.5 μm or less ($PM_{2.5}$) may influence risk of birth defects. We examined associations between maternal exposure to prenatal traffic-related air pollution and risk of cardiac, orofacial, and neural tube defects among Massachusetts births conceived 2001 through 2008. Our analyses included 2,729 cardiac, 255 neural tube, and 729 orofacial defects. We used satellite remote sensing, meteorological and land use data to assess $PM_{2.5}$ and traffic-related exposures at geocoded birth addresses. We calculated adjusted odds ratios (OR) and confidence intervals (CI) using logistic regression models. Generalized additive models were used to assess spatial patterns of defect risk. There were positive associations for a 10μg/m^3 increase in $PM_{2.5}$ and perimembranous ventricular septal defects (OR = 1.34, 95% CI: 0.98, 1.83), patent foramen ovale (OR = 1.19, 95% CI: 0.92, 1.54) and patent ductus arteriosus (OR = 1.20, 95% CI: 0.95, 1.62). Only ostium secundum atrial septal defects displayed significant spatial variation after accounting for known risk factors. $PM_{2.5}$ and traffic-related exposure results were protective for cleft lip but inconsistent for neural tube defects. Further studies are needed that examine both $PM_{2.5}$ exposure and local traffic-related air pollution.

INTRODUCTION

Birth defects are prevalent in 3% of US live births (1), with cardiac, orofacial, and neural tube defects among the most common defects observed (2). Exposure to air pollution during pregnancy has been suggested to increase risk of birth defects (3-8) in some studies. The time between conception and birth is a sensitive and critical time for fetal development due to rapid cell proliferation and rapid development of various organ systems, thus understanding the influence of ambient exposures on fetal development may elucidate the mechanisms behind abnormal fetal development. Studies of fetal exposure to traffic-related air pollution including particulate matter with a diameter of 2.5 μm or less ($PM_{2.5}$) have been associated with adverse birth outcomes such as intrauterine growth retardation and preterm births (9-10), but investigations of the association of $PM_{2.5}$ on birth defect risk have been inconclusive (11-18).

Exposure estimates for earlier studies were constrained to individuals living near air monitoring stations without daily assessments, limiting both spatial and temporal resolution of the exposure assessment resulting in exposure misclassification (19, 20). Earlier studies were unable to adjust for important confounders such as individual-level socioeconomic status (SES) and may have been limited by case ascertainment over a short study period (19, 20). Only one other study (17) to our knowledge has accounted for road density and residential distance to roadways, local measures of traffic-related air pollution, in addition to $PM_{2.5}$ estimates, when assessing risk of birth defects and exposure to ambient air pollution. Satellite-based $PM_{2.5}$ prediction models can provide additional spatial and temporal information for exposure assessment. Models have evolved from using one single predictor (21) to multiple predictors (22-24) and from one-stage models (25) to multi-stage non-linear models (26-29).

The objective of this study is to examine the relationship between cardiac, orofacial, and neural tube defects and traffic-related air pollution using satellite-based $PM_{2.5}$ exposure estimates and eight years of birth defects data for Massachusetts. To further assess the influence of $PM_{2.5}$ on birth defect risk, our study includes an analysis of geographic patterns of birth defects across Massachusetts.

METHODS

Study population

We obtained all live and still births from the Massachusetts state birth registry with an estimated conception date from January 1, 2001 through December 31, 2008. All births in the Massachusetts Birth Defects Registry having cardiac, orofacial, and neural tube defects (*International Classification of Diseases*, Ninth Revision, Clinical Modification (ICD-9-CM) codes 740.0-743.0, 745.0-748.0 and 749.0-749.3) were identified as cases. The Massachusetts Birth Defects Monitoring Program conducts an active surveillance system to collect diagnoses made before age one. We randomly selected 1,000 infants conceived each year to serve as a common control group among all live births without the defects of interest. We excluded birth addresses that could not be successfully geocoded to x and y coordinates (2%) and we excluded syndromic births (5%) that were associated with the outcomes of interest (Table 1.1) among cases and controls. The Institutional Review Boards of the University of California at Irvine and the Massachusetts Department of Public Health approved this research.

Due to differing etiologies, we divided cardiac defects into anatomical groupings based on ICD-9-CM groups. We only included groups with more than 70 cases to ensure sufficient

numbers for model convergence. We also assessed the five most common single ICD-9-CM code cardiac defect diagnoses as their own outcome group. In total, there were 17 outcome groups for cardiac anomalies: transposition of great vessels, tetralogy of fallot, ventricular septal defect, ostium secundum atrial septal defect, endocardial cushion defect, pulmonary valve atresia and stenosis, aortic valve stenosis, hypoplastic left heart syndrome, patent ductus arteriosus, coarctation of aorta, pulmonary artery anomalies, insufficiency of the aortic valve, atrial septal defect- not otherwise specified (a subset of ostium secundum atrial septal defect), perimembranious ventricular septal defect (a subset of ventricular septal defect), muscular ventricular septal defect (a subset of ventricular septal defect), single common atrium (a subset of endocardial cushion defects), and patent foramen ovale (a subset of ostium secundum atrial septal defect). Cases of patent ductus arteriosus and patent foramen ovale were excluded if the infant was preterm (<36 weeks, 4% of patent ductus arteriosus and patent foramen ovale cases). Infants with more than one birth defect were categorized into multiple defect groups unless diagnoses were similar (Table 1.2). Because the majority of cases had multiple birth defect diagnosis (74%), to assess if there was a difference between infants with multiple defects and isolated defects, these two groups were analyzed separately.

For neural tube defects, spina bifida was the most common defect and was analyzed separately; all other neural tube defects were analyzed together due to small numbers. Anencephaly was excluded (13% of neural tube defects) as those included in the registry may not be representative of all anencephaly cases due to early termination. Orofacial defects were divided into two ICD-9-CM categories: cleft lip with or without palate and cleft palate only.

Exposure assessment

Our primary analysis examined the relationship between birth defects and $PM_{2.5}$ exposures modeled using satellite remote sensing, meteorological and land use data. Aerosol optical depth (AOD) is the integral of particle light extinction coefficients from the surface to the top of the atmosphere. It is a measure of the degree to which aerosols prevent light from penetrating the atmosphere and retrieved using wavelengths most sensitive to particles with sizes from 0.1 to 2 μm (30). Thus, AOD is related to the loadings of fine particles in the atmosphere and is a strong predictor of ground-level $PM_{2.5}$ concentrations as most fine particles are emitted and confined in the boundary layer. The number of stationary ambient monitors is limited and the distribution is sparse, while AOD-estimated $PM_{2.5}$ concentrations have the potential to expand the spatiotemporal coverage of ground networks and improve the accuracy of estimates of personal exposure to $PM_{2.5}$ (31). The Geostationary Operational Environmental Satellite (GOES) is the major weather satellite operated by the National Oceanic and Atmospheric Administration (NOAA). GOES provides an aerosol and smoke product (GASP) with AOD retrievals every 30 minutes from sunrise to sunset at 4 km nominal spatial resolution. We obtained GASP AOD data from the NOAA National Environmental Satellite, Data, and Information Service. In this study, AOD measurements (available from 9 am to 3 pm local time) were averaged to generate daily AOD estimates (24).

The 24-hour average $PM_{2.5}$ concentrations from 2001 to 2008 collected from 35 U.S. Environmental Protection Agency (EPA) Federal Reference Monitors (FRM) were downloaded from the EPA's Air Quality System Technology Transfer Network (http://www.epa.gov/ttn/airs/airsaqs/). Meteorological fields, including temperature and wind speed, were provided by the North American Land Data Assimilation System (NLDAS) Phase 2 and downloaded from the NLDAS website (http://ldas.gsfc.nasa.gov/nldas/). Elevation data were

obtained from the National Elevation Dataset (NED) (http://ned.usgs.gov). Major roads were extracted from ESRI StreetMap USA (Environmental Systems Research Institute, Inc., Redland, CA). Forest cover data were derived from 2001 and 2006 land cover maps downloaded from the National Land Cover Database (NLCD) (http://www.mrlc.gov). Primary $PM_{2.5}$ emissions were obtained from the 2002, 2005, and 2008 EPA National Emission Inventory (NEI) facility emissions reports. We developed a linear mixed effects model with 24-hour average EPA $PM_{2.5}$ measurements from 2001 to 2008 as the dependent variable and AOD, meteorological fields and land use variables as predictors. The model incorporates day-specific random intercepts and slopes for AOD, temperature, and wind speed to account for the temporally varying relationship between $PM_{2.5}$ (based on fixed ground monitors) and AOD. (32). This model was run annually for a 4 km modeling grid covering the spatial extent of Massachusetts to estimate daily $PM_{2.5}$ concentrations from 2001 to 2008. The model structure can be expressed as

$$PM_{2.5,st} = (\beta_0 + \beta_{0,t}) + (\beta_1 + \beta_{1,t})AOD_{st} + (\beta_2 + \beta_{2,t})Temperature_{st} + (\beta_3 + \beta_{3,t})Wind\ Speed_{st} + \beta_3 Elevation_s + \beta_4 Major\ Roads_s + \beta_5 Forest\ Cover_s + \beta_6 Point\ Emissions_s + \varepsilon_{st}$$
$$(\beta_{0,t}\beta_{1,t}\beta_{2,t}\beta_{3,t}) \sim N[(0,0,0,0), \Psi]$$

where $PM_{2.5,st}$ is the measured ground level $PM_{2.5}$ concentration ($\mu g/m^3$) at site *s* in day *t*; β_0 and $\beta_{0,t}$ (day-specific) are the fixed and random intercept, respectively; AOD_{st} is the GASP AOD value (unitless) at site *s* in day *t* ; β_1 and $\beta_{1,t}$ (day-specific) are the fixed and random slopes for AOD, respectively; $Temperature_{st}$ is the air temperature (K) at site *s* in day *t*; β_2 and $\beta_{2,t}$ (day-specific) are the fixed and random slopes for temperature, respectively; $Wind\ Speed_{st}$ is the 2 meters above ground wind speed (m/sec) at site *s* in day *t*; β_3 and $\beta_{3,t}$ (day-specific) are the fixed and random slopes for wind speed, respectively; $Elevation_s$ is elevation values (m) at site *s*; $Major\ Roads_s$ is road length (m) at site *s*; $Forest\ Cover_s$ is percentage of forest cover (unitless) at site *s*; $Point\ Emissions_s$ is point emissions (tons per year) at site *s*; and Ψ is an unstructured

variance-covariance matrix for the random effects. A ten-fold cross validation (CV) was conducted to evaluate the performance of the model, and statistical indicators including the coefficient of determination (R^2) and square root of the mean squared prediction errors (RMSPE) were calculated between predicted $PM_{2.5}$ concentrations and observations. The results show that CV RMSPE ranges from 2.42 to 3.50μg/m^3, and CV R^2 ranges from 0.78 to 0.88 for years 2001 through 2008, indicating a good performance of the model.

The clinically estimated gestational age of infants was subtracted from date of birth to calculate conception date. The exposure assessment was performed for varying gestational weeks depending on the outcome of interest. For cardiac, neural tube, and orofacial defects, the windows of exposure are weeks 3–7, 1-4, and 6-12 of pregnancy, respectively, as these periods are considered to be the most critical exposure window for the development of the specified birth defects (33-35). Daily $PM_{2.5}$ estimates were averaged for each exposure assessment interval. Infants were assigned a $PM_{2.5}$ exposure measure if there was at least a single daily $PM_{2.5}$ estimate for each week of the relevant gestational period. Satellite measures provide extensive spatial coverage throughout Massachusetts allowing us to assign exposure estimates to 95% of Massachusetts births included in our study. As a sensitivity analysis, we also assessed the effects of average daily $PM_{2.5}$ over the first trimester of pregnancy.

To understand the influence of local traffic-related pollution on a near-roadway spatial scale, we considered the role of distance to major roadways and traffic density near birth residence (36-39). Using geographic information system software (ArcGIS, version 10.0; ESRI), we calculated the shortest distance (m) between each maternal address at birth and the nearest Class 1 (limited access highways) or 2 (multilane highways without limited access) road segment, and traffic density was calculated by summing the annual average daily traffic (AADT)

within a 200 meter grid of Class 1 and 2 road segments (38), as air pollution from traffic reaches background levels around 200 meters (37). Sensitivity analysis was conducted using Class 3 road segments (other numbered routes) (Figure 1.1). To estimate density for road segments with missing AADT counts, the AADT from the segments nearest to the missing segment of the same class with available data was used.

Statistical analysis

We used logistic regression models to calculate crude and adjusted odds ratios (ORs) and 95% confidence intervals (CI) for each birth defect outcome group and $PM_{2.5}$ exposure. Exposure-response relations were also investigated using cubic splines. We considered adjustment for the following covariates obtained from Massachusetts birth records: plurality, maternal race/ethnicity, maternal education, maternal language preference, delivery payment source, smoking during pregnancy, alcohol consumption during pregnancy, adequacy of prenatal care (measured by the Adequacy of Prenatal Care Utilization Index), marital status, and maternal age. Infant covariates included birth year, parity, and season of conception. We used geocoded addresses to determine the median household income and median home value of census block groups. Analyses that included local traffic measures (traffic density and distance to roadways) were modeled continuously using cubic spline models. For each defect group, we used the change of estimate criterion (10%) to determine which covariates would be included in the model (40). We also assessed effect modification of $PM_{2.5}$ exposure by maternal education. All analyses were conducted using R (R Software Version 3.0.3). The R packages mgcv was used for the cubic spline models.

We examined spatial patterns of birth defects using generalized additive models (GAMs) for each outcome group (41, 42) to determine the residual influence of geographic location after accounting for $PM_{2.5}$ as a confounder and potential mediator. The optimal amount of smoothing was determined by minimizing the Akaike Information Criterion and permutations were used to test for the significance of the smooth term for location. The underlying spatial pattern of the defect was first assessed using an unadjusted model with only the smooth term for location. Two adjusted GAMS were also modeled. The first was adjusted for potential confounders including maternal education, age, race, adequacy of prenatal care, and season of conception. The second included all the indicated covariates from the first model with the addition of satellite-based $PM_{2.5}$ measures to assess residual spatial patterns after accounting for the contribution of $PM_{2.5}$ and other risk factors. Although the percent of missing information was less than 5% for each variable of interest, we applied multivariate imputation for variables with missing values using the predictive mean matching, logistic regression, and polytomous logistic regression imputation method for continuous, binary, and categorical variables, respectively, to evaluate the influence of missing data on our risk estimates. The R package MapGAM and mice was used to run the spatial GAMs to create the maps and multiple imputation, respectively.

RESULTS

We obtained records for 2,729 cardiac, 726 orofacial, 255 neural tube defect cases, and 7,816 controls geocoded, non-syndromic births from the 611,854 births in Massachusetts conceived from January 1, 2001 to December 31, 2008. The percentage of cases with mothers 30-34 years old ranged from 27-31%, depending on the defect. Between 61-72% of mothers were of non-Hispanic White race/ethnicity, and between 56-74% of mothers reported education levels greater

than high school (Table 1.3). Eight percent of control mothers reported smoking during pregnancy, while up to 10% of mothers with orofacial cleft reported smoking. The proportion of mothers that reported drinking during pregnancy ranged from 0.5-2% among the defect groups. Approximately 75% of all cases reported "adequate" prenatal care.

$PM_{2.5}$ and traffic-related exposures

Our $PM_{2.5}$ analysis included 2,610 (95.6%) cardiac, 692 (95.3%) orofacial, and 247 (96.8%) neural tube defects with a measure of in-utero $PM_{2.5}$ exposure for the relevant gestational period. Of the 7,816 controls, 278 (4%), 352 (5%), and 254 (3%) were excluded from the cardiac, orofacial, and neural tube analyses, respectively, due to missing $PM_{2.5}$ data during their respective critical window of exposure. Traffic density and residential distance to nearest major roadway were successfully calculated for all cases and controls.

Cubic splines relating continuous satellite-based $PM_{2.5}$ exposure to log odds of birth defects were approximately linear (see example, Figure 1.2). As such, we report results from logistic regression models of the association between birth defects and $PM_{2.5}$ exposure modeled continuously for a 10 $\mu g/m^3$ increase of $PM_{2.5}$ (Table 1.2). The adjusted ORs for perimembranous ventricular septal defects (OR = 1.34, 95% CI: 0.98, 1.83), patent foramen ovale (OR = 1.18, 95% CI: 0.91, 1.53) and patent ductus arteriosus (OR = 1.24, 95% CI: 0.94, 1.62) were elevated and approaching significance. Odds ratios for the remaining cardiac defects were generally null with wide confidence intervals. Results for the general neural tube defects group suggested an inverse association (OR = 0.70, 95% CI: 0.46, 1.05) whereas for spina bifida the OR was slightly elevated (OR = 1.22, 95% CI: 0.61, 2.30), although neither was statistically significant. Non-significant inverse associations were also observed for orofacial defects (cleft

lip with or without palate: OR = 0.76, 95% CI: 0.50, 1.10; cleft palate: OR = 0.89, 95% CI: 0.54, 1.46). We found that crude estimates were similar to adjusted estimates for all defects except for endocardial cushion and all orofacial defects (Table 1.4). When assessing a wider exposure window of the first trimester of pregnancy, we found similar results for most defects (Table 1.5). Compared to estimates using narrow exposure windows, estimates obtained using first trimester exposure windows were closer to the null for patent ductus arteriosus and atrial septal defect and further away from the null for tetralogy of fallot and endocardial cushion defect. We found evidence for effect modification of $PM_{2.5}$ exposure by maternal education level for endocardial cushion defects ($P = 0.017$), perimembranous VSD ($P = 0.030$), and single common atrium ($P = 0.020$) (Table 1.6).

For comparison, we also examined the relationship between birth defects and local traffic measures. Results for residential proximity to major roadways, modeled continuously with cubic splines are shown in Figure 1.3 for defects that yielded significant associations. Residential distance to major roads was significantly associated with risk of ostium secundum atrial septal defects ($P = 0.039$), endocardial cushion defects ($P = 0.014$), patent ductus arteriosus ($P = 0.047$), atrial septal defects, NOS ($P = 0.018$), insufficiency of the aortic valve ($P = 0.019$), and cleft lip with or without palate ($P = 0.017$). The association between odds of patent foramen ovale and residential distance to major roadway was borderline significant ($P = 0.077$). Traffic-related pollution reaches near background levels between 200-300 meters away from roadways (39). With the exception of atrial septal defects NOS, all defects displayed decreasing risk as distance to major roadways increased for the first 300 meters . The overall trend for atrial septal defects, NOS and cleft lip with or without palate was positive, indicating that odds of defect increased as distance increased. We believe that associations with distances greater than 300

meters may be describing other measures such as access to care or SES (36). General neural tube defects, spina bifida, and cleft palate defects were not significantly associated with residential distance to major roadways (P = 0.317 , 0.747, and 0.803, respectively).

Cubic splines indicated that traffic density and risk of defects was approximately linear (Figure 1.2), therefore odds ratios are presented for an interquartile range increase of AADT (Table 1.7). Results show similar associations with traffic density as many of the same defects associated with $PM_{2.5}$ and distance to major roadways. Ostium secundum atrial septal defects (OR = 1.03, 95% CI: 1.00, 1.06), patent foramen ovale (OR =1.05, 95% CI: 1.01, 1.08), and insufficiency of the aortic valve (OR = 1.07, 95% CI: 1.01, 1.12) were positively associated with traffic density. Patent ductus arteriosus was positively associated with traffic density and approaching significance (OR = 1.03, 95% CI: 0.99, 1.07). Cleft lip with or without palate was negatively associated with traffic density (OR = 0.92, 95% CI: 0.85, 0.98). Results were similar when including class 3 roads in the analysis.

Geographic location

Spatial analyses using GAMs showed significant associations between geographic location and certain birth defects (Table 1.8). Birth location remained statistically significant (P = 0.004) for ostium secundum atrial septal defects after adjusting for demographic, socioeconomic, behavioral risk factors, and $PM_{2.5}$ exposure (Figure 1.4). The relationship between birth location and hypoplastic left heart syndrome was borderline statistically significant (P = 0.067) and was fully explained only after adding $PM_{2.5}$ to the model (P = 0.144, Figure 1.4). To determine the influence of missing data on outcomes in the spatial analysis, five imputed data sets were generated to run the GAMs and permutation tests for hypoplastic left heart syndrome and ostium

secundum atrial septal defect. Results were similar to those generated using the original dataset (Figure 1.5).

Although unadjusted models indicated significant spatial variation across Massachusetts for other defects, adjusted models suggested that the patterns were due to socioeconomic, demographic, and behavioral risk factors, and including $PM_{2.5}$ did not alter the geographic pattern.

DISCUSSION

We examined the spatial relationship between $PM_{2.5}$ and other traffic-related measures using anatomical groupings of cardiac, neural tube and orofacial birth defects. There is evidence to support the hypothesis that exposure to $PM_{2.5}$ and traffic-related air pollution increases risk of patent foramen ovale and patent ductus arteriosus, as these defects displayed a positive but non-significant association with $PM_{2.5.}$ Patent foramen ovale displayed positive significant associations with both distance to major roadways and traffic density.

We assessed traffic-related air pollution at various spatial scales, as our $PM_{2.5}$ measures represent larger scale pollution whereas traffic density and distance to major roads represent more local measures of pollution. Perimembranous ventricular septal defects demonstrated a positive but non-significant association with $PM_{2.5}$ exposure but not with local measures of pollution. Ostium secundum atrial septal defects and insufficiency of the aortic valve defects were significantly associated with local measures of air pollution, but not $PM_{2.5}$, indicating that there may be a specific local pollutant affecting the development of these defects.

There was a noticeable rise in risk for ostium secundum type atrial septal defect, patent ductus arteriosus, patent foramen ovale, insufficiency of the aortic valve, and cleft lip with or

without palate among infants with birth addresses around 1,000-1,500 meters away from a major roadway (Figure 1.3). It is not clear that the rise and decline in risk around 1,000-1,500 meters is statistically significant as it is contained with the CI at shorter distances for most outcomes. It is therefore difficult to tell if the higher risk at that distance is meaningful or an effect of random variation for most birth defects. However, the CIs are narrower for insufficiency of the aortic valve, suggesting that the pattern is meaningful for that outcome. Moreover, the shared pattern of increased risk estimates around 1,000-1,500 meters suggests that some unmeasured risk factor for birth defects may be more common in that range. We examined the confounders of infants whose birth address was between 1,000-1,500 meters away from the nearest major roadway and found they were similar to births residing at other distances. Further investigation is needed to better understand the shared pattern of increased risk estimates around 1,000-1,500 meters. Residential location remained significantly associated with ostium secundum atrial septal defects in the fully adjusted spatial model, suggesting that the persistent areas of increased risk are not fully explained by individual risk factors included in the model. These spatial patterns may be due to unidentified environmental or social determinants.

Interestingly, we observed inverse associations between $PM_{2.5}$ and traffic density and cleft lip with or without palate, although the association with $PM_{2.5}$ was not significant. Other studies have found no association between cleft lip with or without palate and $PM_{2.5}$ (16, 18), yielding null inverse associations. One study also found a consistent, but non-significant inverse association with $PM_{2.5}$ and cleft lip with or without palate across all $PM_{2.5}$ quartiles (17).

In our secondary analysis of effect modification, we found that women with the lowest education level (less than high school) had the highest effect estimate compared to women with at least high school or college education for a majority of birth defects. Although there is some

evidence for effect modification, this stratified analysis contains small numbers of cases, especially in the less than high school category, and therefore the effect estimates have wide confidence intervals.

The mapped results of our crude GAMs indicated there was a statistically significant association between geographic location and certain birth defects. Adjusting for $PM_{2.5}$ exposure influenced the spatial patterns for hypoplastic left heart syndrome only, suggesting that spatial patterns of increased risk for all other defects are not strongly associated with $PM_{2.5}$. Furthermore, despite analyzing eight years of births for an entire state, rare birth defects are prone to small case numbers, limiting the power to detect significant spatial associations.

We investigated three groups of atrial septal defects; ostium secundum atrial septal defects, atrial septal defects, NOS, and patent foramen ovale. To our knowledge no other studies have examined the influence of $PM_{2.5}$ exposure on risk of ostium secundum atrial septal defects. We did not find any studies that investigated the role of patent foramen ovale and air pollutants. We found an overall positive significant association between atrial septal defects, NOS and residential distance to major roadways ($p= 0.030$) and increased risk with $PM_{2.5}$ (OR= 1.23, 95% CI: 0.78, 1.90). Gilboa et al. also found a positive significant association between PM_{10} and atrial septal defects in Texas (5). We found a non-significant positive association between patent ductus arteriosus and $PM_{2.5}$ while Agay-Shay et al. found a significant inverse association with isolated patent ductus arteriosus and $PM_{2.5}$ (11). Another study (43) found significant increased odds of patent ductus arteriosus and PM_{10} while utilizing further restrictive criteria for this outcome group based on active surveillance records, but we were unable to replicate this method because of limited information from the Massachusetts birth defects registry. Although there are differences in the composition of PM_{10} and $PM_{2.5}$, this may indicate that general particulate

matter exposure may be associated with risk of patent ductus arteriosus. We did not find strong evidence supporting an association between neural tube defects and traffic-related measures of air pollution. Similarly, Padula et al. found no association with $PM_{2.5}$ or traffic density and neural tube defects (17).

To better understand the association of $PM_{2.5}$ and traffic-related exposure on birth defects, our study includes improved exposure assessment methods, standardized definition of cases, and important confounders. We believe we have captured several important SES measures, but we are less confident about our ability to capture alcohol and smoking behaviors from self-reported measures. We used satellite data to obtain fine spatial distribution estimates of $PM_{2.5}$ during pregnancy for all of Massachusetts. The use of measured $PM_{2.5}$ for each 4 km square grid cell allows for more fine-scale measures of exposure compared to previous studies that have relied on measures from stationary monitoring stations. We believe that using this finer spatial resolution of $PM_{2.5}$ reduced residential exposure misclassification compared to using monitoring stations alone. By including distance to major roadways and traffic density, we are able to assess the influence of local road networks on risk of birth defects.

Although we were able to control for important covariates, we were unable to directly control for folic acid supplementation, a major contributor of neural tube defects (44). As a proxy for folic acid supplementation we used the adequacy of prenatal care index. This index measures prenatal care based on initiation and adequacy of received services. It has been found that adequacy of prenatal care is a useful proxy for folic acid supplementation during pregnancy (45).

We utilized critical windows of exposure specific to birth defect anatomical location. Previous studies have used average exposures over the first trimester of pregnancy or gestational

weeks 3-8. Organ development is time specific, therefore the use of very specific exposure windows can strongly influence the effect estimates and allow for evaluation of exposure-response relationships to be more readily observed (46, 47). We noticed variability in effect estimates when assessing exposure during the first trimester of pregnancy compared to narrow critical windows of exposure for transposition of the great vessel, tetralogy of fallot, endocardial cushion defect, patent ductus arteriosus, coarctation of the aorta, pulmonary artery anomalies, atrial septal defects, NOS, and spina bifida (Table 1.5).

This analysis includes birth defect diagnoses among still births and diagnoses made up to one year of life. However, we are unable to more fully investigate the effects of fetal toxicity as we do not have records of fetal deaths. Fetal deaths may be more sensitive to the effects of air pollution than those who survive to birth with birth defects. Therefore, we may not have captured the conceptions most at risk. We hypothesize that the detected association would be weaker than the true association had all at risk subjects been included in this analysis.

This study only assessed the relationship between birth defects and traffic-related exposures using residential location of mother at time of delivery. Pregnant women are a mobile population and many women will not have been living in the same home during the early prenatal period as they were at the time of birth of the infant. We believe that this may lead to exposure misclassification but it is likely to be non-differential among cases and controls (48). Residential mobility was estimated to be between 3% and 14% in recent American studies based on birth cohorts (48, 49) and mobility rates did not differ among mothers of infants born with birth defects and mothers of infants born without birth defects (48). Additionally, maternal time-activity patterns were not accounted for in our investigation. This may result in exposure misclassification, although it is also likely to be non-differential among cases and controls.

Recent studies have found that outdoor residential levels of exposure act as a good surrogate for personal exposure (50). Both sources of exposure misclassification may increase type 2 error so our null results should be interpreted with caution. Given the multiple comparisons involved in testing a large range of birth defects with three different metrics for traffic-related air pollution, this analysis may result in significant associations due to chance (e.g., type 1 errors). Therefore we have emphasized associations that are observed for more than one measure of traffic-related air pollution. Furthermore, we acknowledge that $PM_{2.5}$ is a complex mixture of particles with varying toxicity and we do not have measures of composition.

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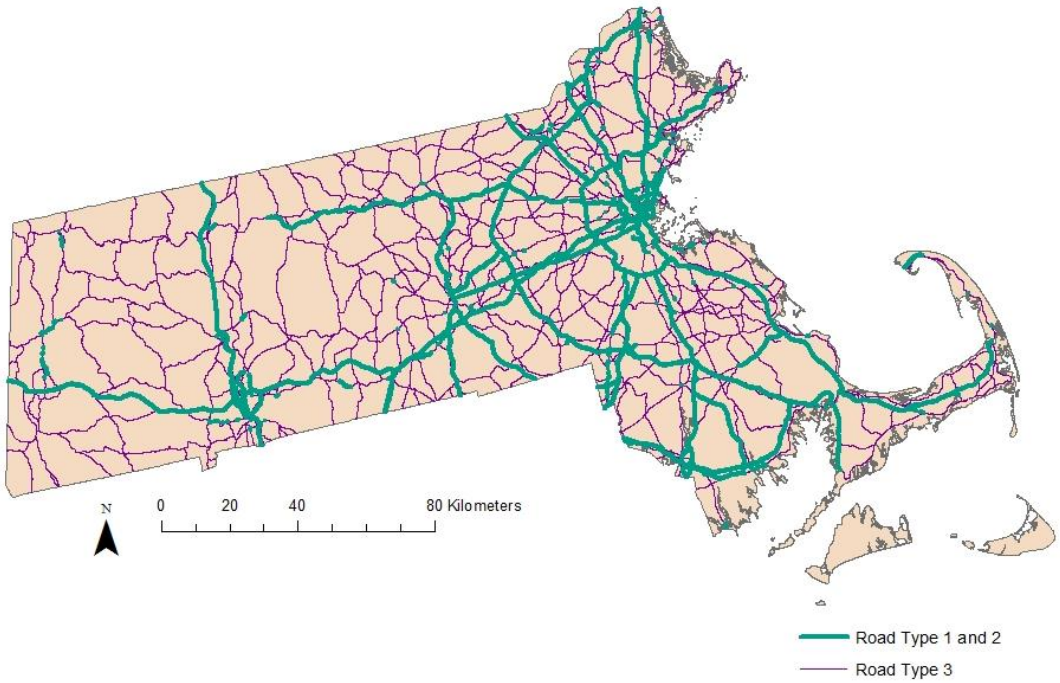

Figure 1.1: Major Roadways in Massachusetts by Road Type.

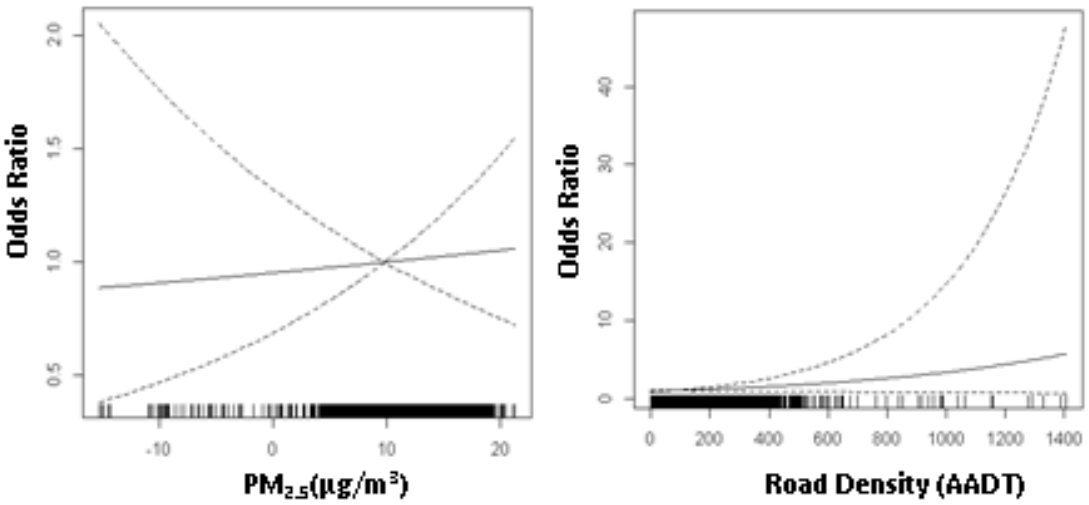

Figure 1.2: Cubic Splines for Pulmonary Valve Atresia and $PM_{2.5}$ and Road Density. Cubic Splines displaying odds ratio (solid line) and 95% confidence intervals (dotted lines) for $PM_{2.5}$ exposure and residential traffic density. All defects yielded linear relationships, similar to those presented above (pulmonary valve atresia) when modeled with $PM_{2.5}$ and road density. Density rugs are provided at the bottom of each plot to display the distribution of values.

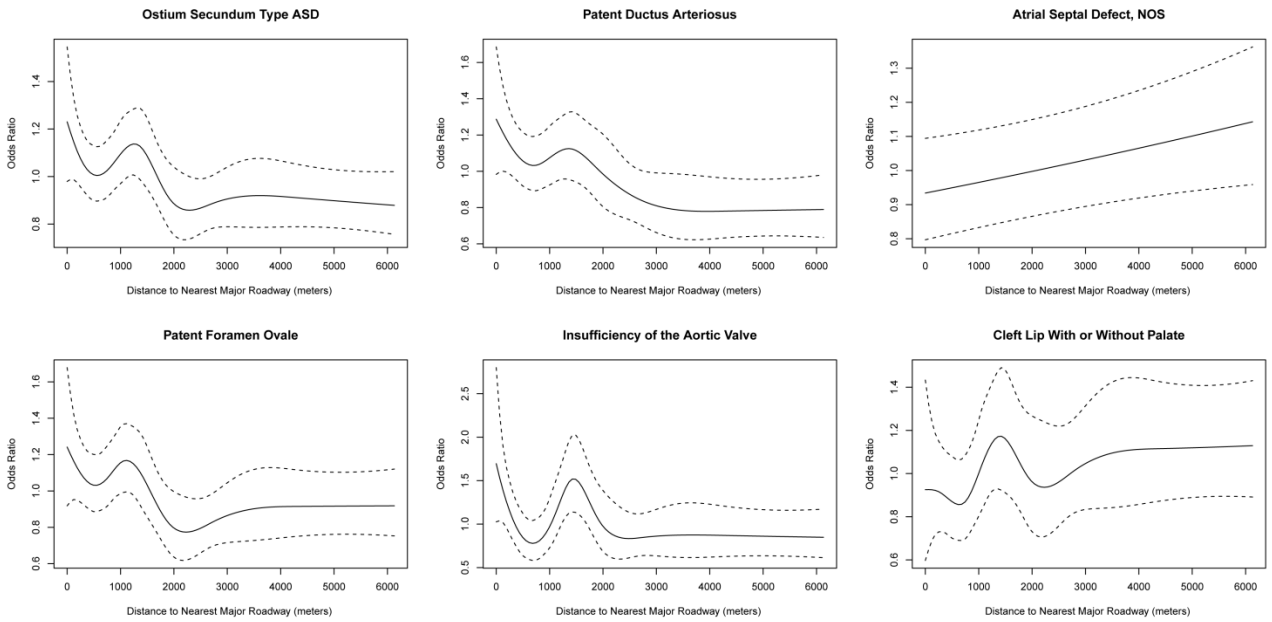

Figure 1.3. Association of Residential Distance to Major Roadways and Risk of Defects. Exposure response curve showing the adjusted odds ratio (solid line) and the 95% confidence interval (dashed line) using cubic splines to model the association of residential distance to a major roadway and birth defects among infants conceived in Massachusetts, 2001-2008. Only birth defects with significant associations are displayed. Estimates are only presented for residential addresses within continental Massachusetts. All models adjusted for maternal race, education, median household income of block group, alcohol consumption during pregnancy, and plurality. Cardiac defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care. Orofacial defects further adjusted for season of conception, infant sex, adequacy of prenatal care, and smoking during pregnancy. Major roadways defined as limited access highways and multi-lane highways (class 1 and 2 roads). Abbreviations: ASD – atrial septal defect; NOS - not otherwise specified.

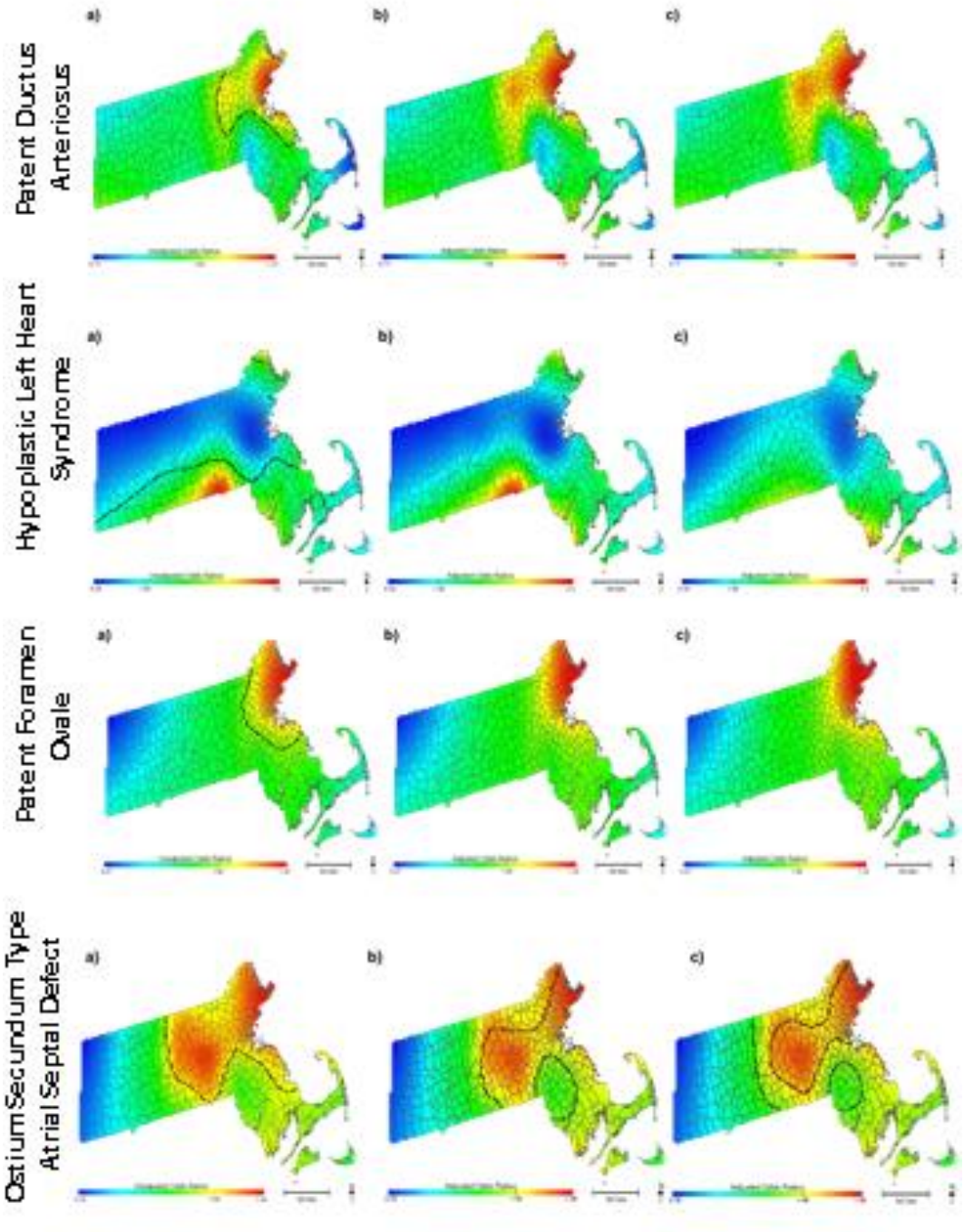

Figure 1.4. Geographic Patterns of Selected Cardiac Defect Risk Among Infants Conceived in Massachusetts, 2001-2008: (a) Unadjusted, (b) Adjusted without $PM_{2.5}$ Exposure, and (c) Adjusted with $PM_{2.5}$ Exposure.
Statistically significant geographic areas of increased or decreased risk of birth defect are indicated using contour lines. Maps adjusted for maternal age, education, race, smoking during pregnancy, alcohol consumption during pregnancy, prenatal care, and season of conception.

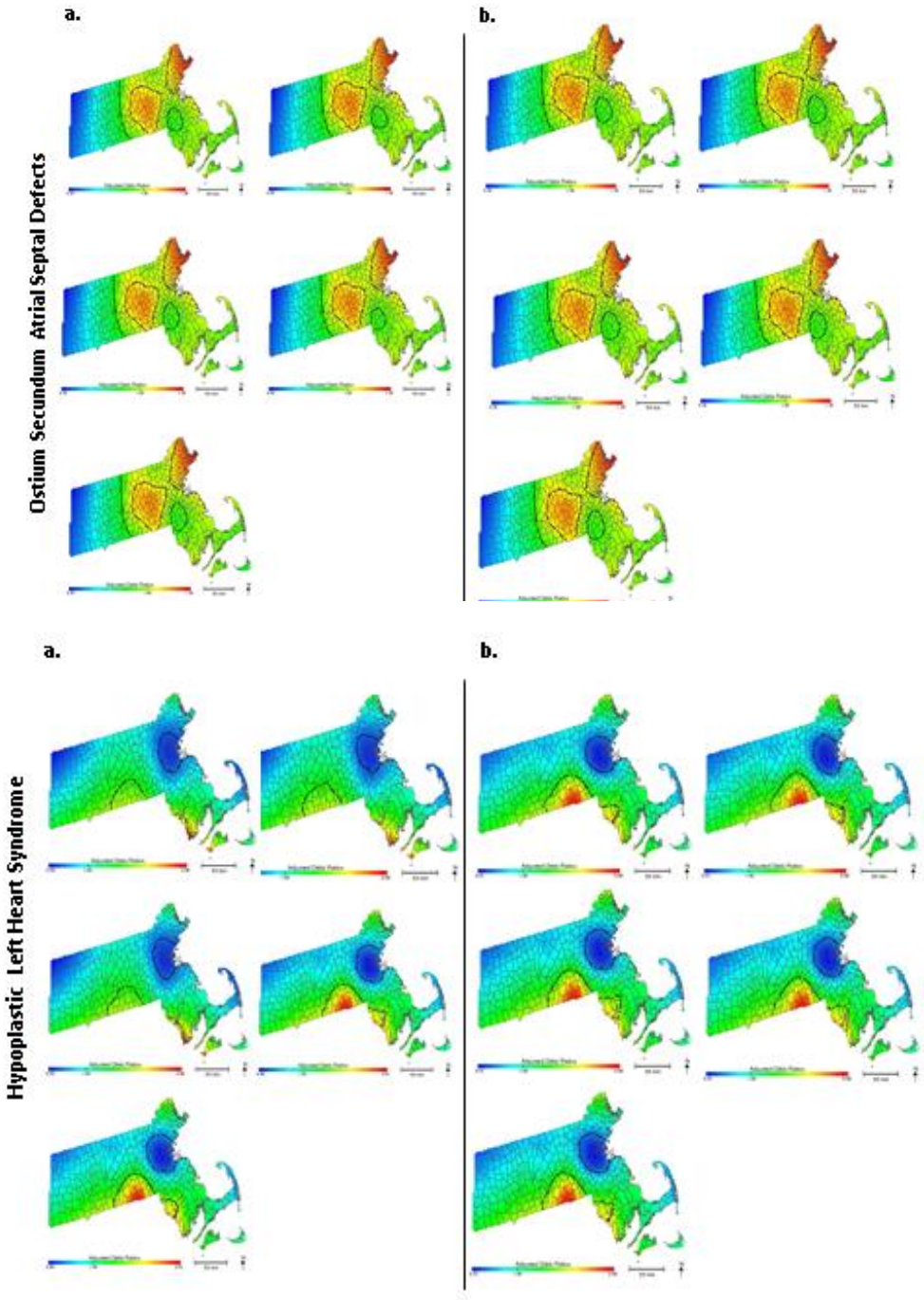

Figure 1.5: Imputed Maps for Ostium Secundum Atrial Septal Defects and Hypoplastic Left Heart Syndrome a.) Adjusted with $PM_{2.5}$ Exposure and b.) Adjusted without $PM_{2.5}$ Exposure.

Results of maps imputed for missing data indicate that spatial patterns were not influenced by missing data.

Table 1.1: International Classification of Disease, 9th Revision Clinical Modification (ICD-9-CM) 6 Digit Codes for Syndromes Associated with Cardiac Defects that were Excluded from Analysis.

Syndrome	ICD-9-CM code
Di George Syndrome	279.110
Down Syndrome	758.000
Marfan Syndrome	759.820
Trisomy 13	758.100
Turner Syndrome	353.500
Noonan Syndrome	237.710
Williams- Beuren Syndrome	759.890
Holt Oram Syndrome	759.890
Alagille Syndrome	759.890
Charge Association	759.890
Char Syndrome	759.890
Ellis-van Creveld Syndrom	756.550
Cardiofaciocutaneous Syndrome	759.890
Costello Syndrome	758.900

Table 1.2: Classification of Birth Defects Adapted from the Birth Defects and Genetic Diseases Branch of the International Classification of Disease, 9th Revision Clinical Modification (ICD-9-CM) 6 Digit Code.

	ICD-9-CM code	Conditions
Cardiac Defects		
Transposition of the Great Vessel	745.100-745.190	Transposition of great vessels, Transposition of great vessels, incomplete with Ventricular Septal Defect (VSD); Taussig-Bing syndrome; L-transposition; Double outlet right ventricle (DORV) with normally related great vessels; DORV with transposed great vessels; DORV, relationship of great vessels not specified; Other specified transposition of great vessels; Unspecified transposition of great vessels.
Tetralogy of Fallot	745.200-745.210	Fallot's tetralogy; Fallot's pentaology and Fallot's tetraology plus atrial septal defects (ASD) ; if dx with tetralogy of fallot and pulmonary valve atresia, case was categorized as tetralogy of fallot only.
Ventricular Septal Defect	745.400-745.498	VSD; Eisenmenger's syndrome; Gerbode defect; Other specified VSD; perimembranous VSD; Muscular VSD; Inlet VSD; VSD, not otherwise specified (NOS); Probable VSD; excluded infant also diagnosed with pulmonary atresia.
Ostium Secundum Atrial Septal Defect (ASD)	745.500-745.590	Nonclosure of foramen ovale, NOS; Patent foramen ovale (PFO); Ostium secundum defect; Other specified atrial septal defect; ASD, NOS.
Edocardial Cushion Defect	745.600-745.690	Ostium primum defects; Single common atrium, cor triloculare biventriculare; common atrioventricular canal with VSD; common antroventricular canal; other specified cushion defect; Endocardial cushion defect, NOS.
Pulmonary Valve Atresia/Stenosis	746.00-746.105	Atresia, hypoplasia of pulmonary valve; stenosis of pulmonary valve (excludes pulmonary infundibular stenosis); pulmonary valve insufficiency or regurgitation; Other specified anomalies of the pulmonary valve; Unspecified anomalies of pulmonary valve.
Aortic Valve Stenosis	746.300	Congenital stenosis of aortic valve (includes congenital aortic stenosis and subvalvular aortic stenosis; excludes supravalvular aortic stenosis)
Hypoplastic Left Heart Syndrome	746.700	Hypoplastic left heart syndrome Atresia, or marked hypoplasia of the ascending aorta and defective development of left ventricle (with mitral valve atresia).
Patent Ductus Arteriosus	747.000-747.008	Patent ductus arteriosus (PDA) for >36 weeks gestation; probable PDA.
Coarctation of Aorta	747.100-	Preductal coarctation of aorta; Postductal coarctation of

	747.190	aorta; Unspecified coarctation of aorta.
Pulmonary Artery Anomalies	747.300-747.380	Pulmonary artery atresia, absence or agenesis; Pulmonary artery atresia with septal defect; Pulmonary artery stenosis; Peripheral pulmonary artery stenosis (> 36 weeks gestation); Aneurysm of pulmonary artery, dilatation of pulmonary artery; Pulmonary arteriovenous malformation or aneurysm; Other specified anomaly of pulmonary artery (includes pulmonary artery hypoplasia and unspecified anomaly of pulmonary artery; excludes cases that were also diagnosed with hypoplasia of left ventricle, stenosis/atresia of mitral and aortic valve -since these are actually hypoplastic left heart syndrome).
Insufficiency of Aortic Valve	746.400-746.490	Aortic valve insufficiency or regurgitation (excludes bicuspid aortic valve, bicuspid aortic valve, other specified anomalies of the aortic valves, supravalvular aortic stenosis; unspecified anomalies of the aortic valves; includes aortic valve atresia).
Single Most Common Defects		
Perimembranous VSD (a subset of VSDs)	745.485	Perimembranous VSD (includes membranous VSD).
Muscular VSD (a subset of VSDs)	745.486	Muscular VSD (includes mid-muscular and apical VSDs).
Single Common Atrium , Cor Tiloculare (a subset of edocardial cushion defects)	745.610	Common atrioventricular canal with VSD (includes common Atrioventricular (AV) canal with muscular VSD.
ASD, NOS (a subset of ASDs)	745.590	ASD,NOS
Patent Foramen Ovale (a subset of ASDs)	745.500	Non-closure of foramen ovale, NOS; Patent foramen ovale (PFO) 1) ≥36 weeks of gestation at birth and defect last noted at ≥6 weeks of age. 2) ≥36 weeks gestation at birth and defect last noted <6 weeks of age, only if another reportable heart defect is present. 3)Never code if <36 weeks gestation
Neural Tube Defects		
Neural Tube Defects	742.000-724.990	Encephalocele; Microcephalus; Reduction deformities of brain; Congenital hydrocephalus; Other specified anomalies of brain; specified anomalies of spinal cord; Other specified anomalies of nervous system; Unspecified anomalies of brain, spinal cord and nervous systems(excluding anencephaly and spina bifida).
Spina Bifida	741.000	Spina Bifida (includes spina bifida aperta, myelocele,

		rachischisis, spina bifida cystica, meningocele, meningomyelocele, myelomeningocele).
Orofacial Defects		
Cleft Lip with & without Palate	749.100-749.290	Cleft lip alone (includes alveolar ridge cleft, cleft gum, harelip); Cleft lip, unilateral; Cleft lip, bilateral; Cleft lip, central; Cleft lip, NOS (fused lip), cleft gum; Cleft lip, unilateral, with any cleft palate; Cleft lip, bilateral, with any cleft palate; Cleft lip, central, with any cleft palate; Cleft lip, NOS, with any cleft palate.
Cleft Palate Only	749.000-749.090	Cleft palate alone; Cleft hard palate, unilateral; Cleft hard palate, bilateral; Cleft hard palate, central; Cleft hard palate, NOS; Cleft soft palate, alone unilateral; Cleft soft palate, alone bilateral; Cleft soft palate, alone central; Cleft soft palate, alone, NOS; Cleft uvula; Cleft palate, NOS (palatoschisis).

Abbreviations: ASD, atrial septal defect; AV, atrioventricular; DORV, double outlet right ventricle; NOS, not otherwise specified; PFO, patent foramen ovale; VSD, ventricular septal defect

Table 1.3. Description of Birth Defect Cases and Randomly Selected Controls in Massachusetts, Conceived 2001-2008

n(%)[a]	Controls (n=7,816)	Cardiac (n=2,729)	Orofacial (n=726)	Neural Tube (n=255)
Infant Sex				
Male	4,032 (51.6)	1,412 (51.7)	411 (56.7)	107 (42.0)
Female	3,784 (48.4)	1,317 (48.3)	314 (43.3)	148 (58.0)
Maternal Age				
<20 years	462 (5.9)	148 (5.4)	46 (6.3)	19 (7.4)
21-24 years	1,227 (15.7)	417 (15.2)	118 (16.2)	38 (14.9)
25-29 years	1,878 (24.0)	586 (21.4)	181 (24.9)	71 (27.8)
30-34 years	2,418 (30.9)	853 (31.2)	211 (29.1)	70 (27.4)
35+ years	1,831 (23.4)	725 (26.5)	169 (23.3)	57 (22.3)
Parity				
0	3,521 (45.0)	1,225 (44.8)	320 (44.1)	123 (48.2)
1	2,665 (34.1)	884 (32.3)	247 (34.0)	84 (32.9)
≥ 2	1,612 (20.6)	617 (22.6)	157 (21.6)	48 (18.8)
Adequacy of Prenatal Care				
Adequate	6,098 (78.0)	2,121 (77.7)	529 (72.9)	190 (74.5)
Intermediate	1,372 (17.5)	462 (16.9)	149 (20.5)	45 (17.6)
Inadequate	247 (3.1)	103 (3.7)	36 (4.9)	14 (5.4)
Unknown	74 (0.9.0)	34 (1.2)	10 (1.3)	5 (1.9)
None	25 (0.3)	9 (0.3)	1 (0.1)	1 (0.3)
Smoking During Pregnancy				
Yes	623 (7.9)	205 (7.5)	74 (10.2)	22 (8.6)
No	7193 (92.0)	2524 (92.4)	651 (89.7)	233 (91.3)
Drinking During Pregnancy				
Yes	137 (1.7)	36 (1.3)	11 (1.5)	1 (0.4)
No	7,674 (98.1)	2,691 (98.6)	713 (98.3)	254 (99.6)
Season of Conception				
Winter	1,883 (24.0)	646 (23.6)	158 (21.7)	55 (21.5)
Spring	1,869 (23.1)	651 (23.8)	209 (28.8)	62 (24.3)
Summer	2,008 (25.6)	657 (24.0)	151 (20.8)	63 (24.7)
Fall	2,056 (26.3)	775 (28.4)	207 (28.5)	75 (29.4)
Gestational Age				
<37 weeks	692 (8.8)	646 (23.7)	99 (13.6)	66 (25.8)
≥37 weeks	7,124 (91.1)	2,083 (76.3)	626 (86.4)	189 (74.1)
Small for Gestational Age				
Yes	845 (10.8)	651 (23.8)	153 (21.1)	95 (37.2)
No	6,961 (89.1)	2,076 (76.0)	572 (78.9)	159 (62.3)
Maternal				

Race/Ethnicity				
Non-Hispanic White	5,379(68.8)	1,837 (67.3)	522 (72.0)	156 (61.1)
Non-Hispanic Black	682(8.7)	284 (10.4)	35 (4.8)	33 (12.9)
Hispanic	1,059(13.5)	375 (13.7)	102 (14.0)	46 (18.0)
Asian/Pacific Islander	530 (6.7)	157 (5.7)	49 (6.7)	11 (4.3)
Other	162 (2.0)	75 (2.7)	17 (2.3)	9 (3.5)
Maternal Education				
<12th grade	793 (10.1)	302 (11.1)	86 (11.8)	30 (11.8)
High school graduation	2,010 (25.7)	710 (26.0)	215 (29.6)	81 (31.8)
Some college	5,002 (64.0)	1,712 (62.7)	424 (58.4)	144 (56.4)
Maternal Language Preference				
English	6,945 (88.8)	2,407 (88.2)	642 (88.5)	209 (81.9)
Spanish	400 (5.1)	164 (6.0)	39 (5.3)	22 (8.6)
Portuguese	181 (2.3)	59 (2.1)	15 (2.0)	10 (3.9)
Other	275 (3.5)	86 (3.1)	25 (3.4)	13 (5.1)
Household Income				
<$20,000	339 (4.4)	126 (4.6)	32 (4.4)	11 (4.3)
$20,000-$69,999	3899 (49.8)	1431 (52.4)	386 (39.4)	158 (62.0)
≥$70,000	3578 (45.8)	1172 (42.9)	307 (42.3)	86 (33.7)
Delivery Source of Payment				
HMO	1,365 (17.4)	437 (16.0)	125 (17.2)	51 (20.0)
Medicaid/Common Health	1,988 (25.4)	741 (27.1)	207 (28.5)	79 (30.9)
Other	4,463 (57.1)	1,550 (56.8)	392 (54.0)	125 (49.0)

[a]Percentages may not sum to 100% due to rounding.

Table 1.4 : Crude and Adjusted Odds Ratios[a] and 95% Confidence Interval for In-Utero Exposure to $PM_{2.5}$ in Massachusetts, and Cardiac, Neural Tube, and Orofacial Defects Among Infants Conceived Between 2001-2008

Defect Type[b]	*N*	Crude OR(95% CI)[c]	Adjusted OR (95% CI)[c]
Isolated Birth Defects	890	0.92 (0.71, 1.21)	0.97 (0.94, 1.00)
Multiple Birth Defects	2571	1.01 (0.84, 1.20)	1.01 (0.85, 1.21)
Total Cases	3,461		
Cardiac			
Transposition of the Great Vessel	233	0.90 (0.59, 1.37)	0.92 (0.60, 1.41)
Tetralogy of Fallot	153	0.99 (0.58, 1.68)	1.00 (0.59, 1.71)
Ostium Secundum ASD	1457	0.96 (0.80, 1.16)	0.98 (0.81, 1.19)
Endocardial Cushion Defect	139	0.99 (0.58, 1.68)	1.18 (0.67, 2.09)
Pulmonary Valve Atresia/Stenosis	436	1.01 (0.73, 1.39)	1.05 (0.76, 1.45)
Aortic Valve Stenosis	93	1.15 (0.58, 2.30)	1.18 (0.58, 2.38)
Hypoplastic Left Heart Syndrome	69	0.70 (0.34, 1.45)	0.73 (0.35, 1.42)
Patent Ductus Arteriosus	675	1.24 (0.95, 1.62)	1.24 (0.94, 1.62)
Coarctation of Aorta	205	1.03 (0.65, 1.63)	1.03 (0.65, 1.64)
Pulmonary Artery Anomalies	172	1.10 (0.67, 1.83)	1.04 (0.64, 1.68)
VSD	864	1.08 (0.86, 1.37)	1.09 (0.86, 1.37)
Perimembranous VSD	494	1.32 (0.98, 1.81)	1.34 (0.98, 1.83)
Muscular VSD	328	0.87 (0.61, 1.25)	0.89 (0.62, 1.27)
Single Common Atrium , Cor Tiloculare	335	1.12 (0.78, 1.62)	1.19 (0.82, 1.72)
Atrial Septal Defect, NOS	235	1.24 (0.80, 1.94)	1.23 (0.78, 1.90)
Patent Foramen Ovale	725	1.15 (0.89, 1.50)	1.18 (0.91, 1.53)
Insufficiency of Aortic Valve	262	1.11 (0.73, 1.68)	1.16 (0.76, 1.76)
Neural Tube			
Neural Tube Defects	199	0.72 (0.47, 1.09)	0.77 (0.46, 1.05)
Spina Bifida	89	1.22 (0.63, 2.37)	1.18 (0.61, 2.30)

Orofacial			
Cleft Lip with & without Palate	406	1.02 (0.71, 1.47)	0.76 (0.50, 1.10)
Cleft Palate Only	251	1.24 (0.78, 1.95)	0.89 (0.54, 1.46)

Abbreviations: ASD, atrial septal defect; CI, confidence interval; NOS, not otherwise specified; OR, odds ratio; $PM_{2.5}$, particulate matter with a diameter of 2.5 μm or less; VSD, ventricular septal defect.

[a] Control group n=7,538, 7,464, 7,562 for cardiac, neural tube, and orofacial defect analysis, respectively.

[b] All models adjusted for maternal race, education, median household income of block group, alcohol consumption during pregnancy, and plurality. Isolated and multiple birth defects further adjusted for maternal age, language preference, parity, and adequacy of prenatal care. Cardiac defects further adjusted for maternal age, language preference, parity, and adequacy of prenatal care. Neural tube defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care, and smoking during pregnancy. Orofacial defects further adjusted for season of conception, infant sex, adequacy of prenatal care, and smoking during pregnancy.

[c] Exposure period of interest is trimester 1 of pregnancy. Control group n=7,538. ORs and 95% CIs correspond to a 10 $\mu g/m^3$ increase of average $PM_{2.5}$ measured at the 4 km grid cell of infant's birth address during weeks 3-7,1-4, and 6-12 of pregnancy for cardiac, neural tube, and orofacial defects, respectively.

Table 1.5. Adjusted Odds Ratios[a] and 95% Confidence Interval for Trimester 1 Exposure to $PM_{2.5}$ in Massachusetts, and Cardiac, Neural Tube, and Orofacial Defects Among Infants Conceived Between 2001-2008.

Defect Type[b]	*n*	Adjusted OR (95% CI)[c]
Cardiac		
Transposition of the Great Vessel	233	1.05 (0.62, 1.78)
Tetralogy of Fallot	153	1.12 (0.59, 2.13)
Ostium Secundum ASD	1457	0.98 (0.78, 1.23)
Endocardial Cushion Defect	139	1.04 (0.54, 2.03)
Pulmonary Valve Atresia/Stenosis	436	1.01 (0.70, 1.48)
Aortic Valve Stenosis	93	1.19 (0.51, 2.77)
Hypoplastic Left Heart Syndrome	69	0.74 (0.34, 1.66)
Patent Ductus Arteriosus	675	1.49 (1.06, 2.09)
Coarctation of Aorta	205	0.90 (0.52, 1.53)
Pulmonary Artery Anomalies	172	0.73 (0.45, 1.20)
VSD	864	1.07 (0.81, 1.41)
Perimembranous VSD	494	1.31 (0.89, 1.90)
Muscular VSD	328	0.90 (0.59, 1.36)
Single Common Atrium , Cor Tiloculare	335	1.12 (0.72, 1.74)
Atrial Septal Defect, NOS	235	1.47 (0.85, 2.56)
Patent Foramen Ovale	725	1.24 (0.90, 1.71)
Insufficiency of Aortic Valve	262	1.14 (0.69, 1.90)
Neural Tube		
Neural Tube Defects	199	0.73 (0.45, 1.18)
Spina Bifida	89	0.88 (0.40, 1.93)
Orofacial		
Cleft Lip with & without Palate	406	0.78 (0.52, 1.14)
Cleft Palate Only	251	0.94 (0.56, 1.56)

Abbreviations: ASD, atrial septal defect; CI, confidence interval; NOS, not otherwise specified; OR, adjusted odds ratio; $PM_{2.5}$, particulate matter with a diameter of 2.5 μm or less; VSD, ventricular septal defect.

[a] Common control group n=7,538.

[b] All models adjusted for maternal race, education, median household income of block group, alcohol consumption during pregnancy, and plurality. Cardiac defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care. Neural tube defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care, and smoking during pregnancy. Orofacial defects further adjusted for season of conception, infant sex, adequacy of prenatal care, and smoking during pregnancy.

[c] ORs and 95% CIs correspond to a 10 $\mu g/m^3$ increase of average $PM_{2.5}$ measured at the 4 km grid cell of infant's birth address during Trimester 1 of pregnancy for cardiac, neural tube, and orofacial defects, respectively.

Table 1.6. Stratified Analysis by Maternal Education Level Displaying Adjusted Odds Ratios and 95% Confidence Interval for In-utero Exposure to $PM_{2.5}$ in Massachusetts, and Cardiac, Neural Tube, and Orofacial Defects Among Infants Conceived Between 2001-2008.

Defect Type[b]	OR (95% CI)[b]	OR (95% CI)[b]	OR (95% CI)[b]	p-value[c] for interaction
	Less than High School	Completed High School	Some College or More	
Cardiac				
Transposition of the Great Vessel	1.41 (0.40, 5.00)	1.20 (0.46, 3.13)	0.69 (0.40, 1.20)	0.224
Tetralogy of Fallot	1.21 (0.26, 5.67)	1.10 (0.38, 3.17)	0.97(0.49, 1.93)	0.854
Ostium Secundum ASD	1.72 (0.99, 3.00)	0.84 (0.60, 1.18)	0.96 (0.75, 1)	0.040
Endocardial Cushion Defect	8.76 (1.43, 53.7)	1.62 (0.54, 4.85)	0.73 (0.35, 1.51)	0.017
Pulmonary Valve Atresia/Stenosis	2.52 (0.97, 6.56)	0.70 (0.39, 1.25)	1.01 (0.66, 1.57)	0.078
Aortic Valve Stenosis	0.95 (0.08, 1.09)	1.27 (0.32, 4.97)	1.16 (0.48, 2.77)	0.856
Hypoplastic Left Heart Syndrome	4.74 (0.13, 16.3)	0.60 (0.23, 1.58)	0.84 (0.28, 2.48)	0.424
Patent Ductus Arteriosus	2.04 (0.90, 4.61)	1.39 (0.83, 2.30)	1.04 (0.73, 1.48)	0.066
Coarctation of Aorta	2.07 (0.28, 15.0)	1.14 (0.49, 2.66)	0.89(0.48, 1.62)	0.494
Pulmonary Artery Anomalies	0.68 (0.31, 1.47)	2.16 (0.79, 5.90)	1.13(0.56, 2.28)	0.180
VSD	1.62 (0.85, 3.08)	1.31 (0.82, 2.10)	0.92 (0.67, 1.25)	0.119
Perimembranous VSD	3.44 (1.32, 8.95)	1.50 (0.81, 2.78)	1.08(0.73, 1.63)	0.030
Muscular VSD	1.55 (0.50, 4.79)	1.34 (0.63, 2.85)	0.66 (0.42, 1.05)	0.130
Single Common Atrium , Cor Tiloculare	4.39 (1.40, 13.7)	0.81 (0.41, 1.61)	1.03 (0.63, 1.69)	0.020
Atrial Septal Defect, NOS	1.43 (0.39, 5.26)	1.88 (0.80, 4.41)	0.98 (0.56, 1.73)	0.495
Patent Foramen Ovale	1.48 (0.75, 2.95)	1.23 (0.72, 2.01)	1.13 (0.80, 1.59)	0.424
Insufficiency of Aortic Valve	1.46 (0.35, 6.19)	1.16 (0.76, 1.76)	1.14 (0.51, 2.52)	0.978
Neural Tube				
Neural Tube Defects	0.62 (0.26, 1.44)	0.89 (0.41, 1.90)	0.68 (0.37, 1.22)	0.677
Spina Bifida	1.45 (0.28, 7.71)	1.39 (0.45, 4.26)	1.23 (0.46, 3.30)	0.442
Orofacial				
Cleft Lip with & without Palate	0.80 (0.34, 1.80)	1.37 (0.60, 3.15)	0.56 (0.33, 0.95)	0.281
Cleft Palate Only	0.50 (0.14, 1.73)	0.84 (0.38, 1.87)	1.03 (0.49, 2.14)	0.038

Abbreviations: ASD, atrial septal defect; CI, confidence interval; NOS, not otherwise specified; OR, adjusted odds ratio; $PM_{2.5}$, particulate matter with a diameter of 2.5 μm or less; VSD, ventricular septal defect.

[a] All models adjusted for maternal race, education, median household income of block group, alcohol consumption during pregnancy, and plurality. Cardiac defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care. Neural tube defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care, and smoking during pregnancy. Orofacial defects further adjusted for season of conception, infant sex, adequacy of prenatal care, and smoking during pregnancy.

[b] ORs and 95% CIs correspond to a 10 $\mu g/m^3$ increase of average $PM_{2.5}$ measured at the 4 km grid cell of infant's birth address during weeks 3-7,1-4, and 6-12 of pregnancy for cardiac, neural tube, and orofacial defects, respectively.

[c] p-value is presented for the null hypothesis that the OR for education level less than high school is equal to the OR for education level college or more.

Table 1.7: Adjusted Odds Ratios[a] and 95% Confidence Interval for Residential Traffic Density[b] and Risk of Cardiac, Neural Tube, and Orofacial Defects Among Infants Conceived Between 2001-2008, and Born in Massachusetts.

Defect Type[c]	*n*	OR (95% CI)
Cardiac		
Transposition of the Great Vessel	243	1.00 (0.94, 1.08)
Tetralogy of Fallot	160	1.05 (0.99, 1.12)
VSD	904	1.02 (0.98, 1.05)
Ostium Secundum ASD	1514	1.03 (1.00, 1.06)
Edocardial Cushion Defect	144	0.94 (0.85, 1.05)
Pulmonary Valve Atresia/Stenosis	462	1.00 (0.88, 1.12)
Aortic Valve Stenosis	96	1.03 (0.92, 1.14)
Hypoplastic Left Heart Syndrome	72	0.97 (0.85, 1.11)
Patent Ductus Arteriosus	703	1.03 (0.99, 1.07)
Coarctation of Aorta	213	1.05 (0.99, 1.12)
Pulmonary Artery Anomalies	179	0.99 (0.92, 1.08)
Perimembranous VSD	514	1.02 (0.97, 1.07)
Muscular VSD	343	0.98 (0.92, 1.05)
Single Common Atrium, Cor Tiloculare	357	0.98 (0.92, 1.04)
Atrial Septal Defect, NOS	242	0.97 (0.89, 1.05)
Patent Foramen Ovale	750	1.05 (1.01, 1.08)
Insufficiency of Aortic Valve	275	1.07 (1.01, 1.12)
Neural Tube		
Neural Tube Defects (excluding Spina Bifida and Anencephaly)	208	1.03 (0.96, 1.10)
Spina Bifida	91	1.05 (0.96, 1.15)
Orofacial		
Cleft Lip with & without Palate	417	0.92 (0.85, 0.98)
Cleft Palate Only	265	0.97 (0.90, 1.05)

Abbreviations: ASD, atrial septal defect; CI, confidence interval; NOS, not otherwise specified; OR, adjusted odds ratio; $PM_{2.5}$, particulate matter with a diameter of 2.5 μm or less; VSD, ventricular septal defects.

[a] Odds Ratio for interquartile range (60.8) annual average daily traffic (AADT) increase in annual average daily traffic; Control group n=7816 for all analysis.

[b] AADT within 200 meters of birth residence (using class 1 and 2 roads).

[c] All models adjusted for maternal race, education, median household income of block group, alcohol consumption during pregnancy, and plurality. Cardiac defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care. Neural tube defects further adjusted for maternal age, language preference, parity, adequacy of prenatal care, and smoking during pregnancy. Orofacial defects further adjusted for season of conception, infant sex, adequacy of prenatal care, and smoking during pregnancy.

Table 1.8: Summary of Spatiotemporal Cardiac Birth Defect Models for Unadjusted and Adjusted Models[a] Among Infants Conceived Between 2001-2008, and Born in Massachusetts.

Birth Defect	Cases (n)	Unadjusted		Adjusted without $PM_{2.5}$[b]		Adjusted with $PM_{2.5}$[c]	
		cOR Range[d]	p-value[e]	aOR Range	p-value[e]	aOR Range	p-value[e]
Cardiac Defects							
Transposition of the Great Vessel	236	0.86-1.23	0.859	0.60-1.40	0.039	1.03-1.50	0.035
Tetralogy of Fallot	156	0.65-1.48	0.234	0.57-1.29	0.106	0.55-1.28	0.105
Ventricular Septal Defect	877	0.63-1.18	0.030	0.66-1.15	0.347	0.66-1.16	0.306
Ostium Secundum ASD	1470	0.32-1.24	<0.001	0.33-1.29	0.007	0.33-1.29	0.004
Edocardial Cushion Defect	141	0.61-1.49	0.297	0.98-2.65	0.180	1.18-3.15	0.204
Pulmonary Valve Atresia/Stenosis	441	0.67-1.42	0.224	0.80-1.75	0.207	0.75-1.63	0.208
Aortic Valve Stenosis	95	0.57-2.78	0.207	0.40-2.05	0.198	0.32-1.69	0.184
Hypoplastic Left Heart Syndrome	74	0.44-2.50	0.033	0.44-2.48	0.067	0.37-2.38	0.144
Patent Ductus Arteriosus	684	0.71-1.21	0.006	0.77-1.22	0.134	0.78-1.23	0.116
Coarctation of Aorta	210	0.49-1.23	0.292	0.64-1.47	0.324	0.57-1.33	0.336
Pulmonary Artery Anomalies	174	0.48-1.19	0.191	0.46-1.1	0.233	0.39-0.92	0.119
Perimembranous VSD	502	0.52-1.19	0.192	0.62-1.32	0.069	0.64-1.34	0.079
Muscular VSD	331	0.55-1.29	0.311	0.87-2.18	0.273	0.94-2.53	0.261
Single Common Atrium , Cor Tiloculare	339	0.65-1.13	0.527	0.73-1.35	0.524	0.67-1.22	0.439
Atrial Septal Defect, NOS	243	0.78-1.87	0.029	0.69-1.69	0.138	0.63-1.78	0.088
Patent Foramen Ovale	731	0.47-1.32	<0.001	0.48-1.36	0.058	0.50-1.40	0.063
Insufficiency of Aortic Valve	266	0.61-1.65	0.782	0.56-1.61	0.080	0.5-1.5	0.075
Neural Tube Defects							
Neural Tube Defects	228	0.84-1.24	0.812	0.71-1.23	0.7957	0.92-1.59	0.807
Spina Bifida	102	0.71-2.19	0.017	0.69-2.13	0.204	0.67-2.07	0.182
Orofacial Defects							
Cleft Lip with & without		0.82-1.47	0.244	0.60-1.01	0.409	0.62-1.03	0.44

Palate	417						
Cleft Palate Only	291	0.63-1.34	0.323	0.79-1.68	0.409	0.66-1.4	0.36

Abbreviations: aOR, adjusted odds ratio; ASD, atrial septal defect; CI, confidence interval; cOR, crude odds ratio; $PM_{2.5}$, particulate matter with a diameter of 2.5 µm or less; NOS, not otherwise specified; VSD, ventricular septal defects

[a] Common control group (n=7,816) for all analysis.

[b] Model adjusted for maternal age, maternal education, race, smoking during pregnancy, drinking during pregnancy, prenatal care, and season of conception.

[c] Model adjusted for variables above and $PM_{2.5.}$

[d] Range of OR point estimates across the prediction grid (geographical location of Massachusetts).

[e] Global hypothesis test that birth defect odds do not depend on the geographic location of participants; permutation tests were used to determine statistical significance.

CHAPTER 2

Acute Air Pollution Exposure and Infant Bronchiolitis and Otitis Media Risk

ABSTRACT

Acute exposure to particulate matter with diameter of 2.5 μm or less ($PM_{2.5}$) may increase risk of infant bronchiolitis and otitis media (OM). Study objectives are to estimate the association between short-term increases in $PM_{2.5}$ concentrations and risk of acute infant bronchiolitis and OM among Massachusetts births born 2001 through 2008 and determine susceptibility factors to identify infants most at risk. This semi-bidirectional case-crossover study included 20,017 infant bronchiolitis and 42,336 OM hospitalizations, observational stays, and emergency department visits. $PM_{2.5}$ was modeled using satellite, remote sensing, meteorological and land use data. Conditional logistic regression was applied to estimate odds ratios (OR) and confidence intervals (CI) per 10-$\mu g/m^3$ increase in $PM_{2.5.}$ Effect modification of risk factors to determine the most susceptible subgroups was assessed. Infant bronchiolitis risk was elevated for $PM_{2.5}$ exposure 0 day (OR=1.03; 95% CI: 0.98, 1.07), 1 day (OR = 1.07; 95% CI: 1.03, 1.11) and 4 days (OR = 1.04; 95% CI: 0.99, 1.08) prior to clinical encounter, but not 7 days. Non-significant associations with OM varied depending on lag (ORs from 0.97-1.02). Preterm infants were at substantially increased risk of bronchiolitis 1 day prior to clinical encounter (OR = 1.17; 95% CI: 1.08, 1.28) and OM 4 and 7 days prior to clinical encounter (OR = 1.09; 95% CI: 1.02, 1.16 and OR = 1.08; 95% CI: 1.02, 1.15, respectively).Our findings suggest that preterm infants are most susceptible to infant bronchiolitis and OM associated with acute $PM_{2.5}$ exposures.

INTRODUCTION

Particulate matter with an aerodynamic diameter of 2.5 microns or less ($PM_{2.5}$) is a widespread air pollutant suspected to be harmful to infants and adults (1-3). Infants may be more susceptible to adverse effects of $PM_{2.5}$ because they are more likely to be active, breathe more air per pound of body mass, and are still physiologically developing (4). It is suspected that $PM_{2.5}$ also plays a role in infant mortality and adverse developmental outcomes, such as low birth weight (1,4-6). In this paper, we investigate the role of acute $PM_{2.5}$ exposure on the risk of infant bronchiolitis, the leading cause of hospitalizations among children during their first year of life (7) and otitis media, the most frequent childhood infection among children less than 3 years of age (8). We also aim to identify infant subgroups most vulnerable to the effects of acute $PM_{2.5}$ exposure. This study is the first environmental epidemiologic analysis of the population-based Pregnancy to Early Life Longitudinal (PELL) cohort, which includes all 619,250 births in Massachusetts from 2001-2008.

Bronchiolitis is a lower respiratory tract infection with variability in symptoms and severity. Some infants experience mild symptoms while others are hospitalized (9). Exposures such as indoor wood burning and environmental tobacco smoke have been associated with risk of hospitalization for bronchiolitis (10-11). Although the literature on infant bronchiolitis and $PM_{2.5}$ is limited, it is suggestive of a possible association (12-14). Analysis in geographic areas with relatively high $PM_{2.5}$ background levels, such as Los Angeles, have found positive associations with risk of bronchiolitis and increased chronic $PM_{2.5}$ exposure (14). Acute $PM_{2.5}$ exposure has been positively correlated with infant bronchiolitis in Italy (15) and associated with risk of infant bronchiolitis among low birthweight infants (13).

Otitis media (OM), or inflammation of the middle ear, is the most common cause for medical care besides a healthy child visit and a major cause for antibiotic use within the first few years of life (16-17). Sixty percent of infants experience at least one episode of OM by one year of life (18). OM can lead to hearing loss which can impact speech and delay cognitive development. Much like bronchiolitis, OM is typically caused by a viral infection and associated with environmental exposures, such as tobacco smoke and indoor wood (19-22). Currently, there is little literature on the association between OM and $PM_{2.5}$. One study detected an association between lifetime $PM_{2.5}$ exposure and OM (23). Another study also detected an association in a geographic locations of relatively low $PM_{2.5}$ levels (mean levels between 3.9-5.5 $\mu g/m^3$) when assessing $PM_{2.5}$ exposure two months prior to the clinical encounter (24).

We conducted a case-crossover study of infant bronchiolitis and OM that utilizes satellite-based $PM_{2.5}$ estimates covering the entire geographic region of Massachusetts at a 4 kilometer (km) gridded spatial resolution. Many previous studies have relied on exposure measurements from the nearest stationary air monitoring station (13,23-24), limiting their study populations (particularly to the urban core) and raising concerns regarding exposure measurement error (26). An additional strength of this study is the large sample size of births from the PELL cohort with linked clinical record data from the entire state, providing good precision for $PM_{2.5}$ effect estimates.

METHODS

Study Population

Cases were obtained from the Pregnancy to Early Life Longitudinal (PELL) study, a Massachusetts data linkage system which links birth and hospital encounter records (27). Cases

of infant bronchiolitis were selected among infants born from 2001-2008 in Massachusetts and were defined as the first clinical encounter (hospitalizations, observational stays, or emergency department visits) with a primary or secondary diagnosis of infant bronchiolitis (*International Classification of Diseases*, Ninth Revision, Clinical Modification (ICD-9-CM) 466.0-466.1). Cases of OM were selected among infants born from 2001-2006 and were defined as the first clinical encounter with a primary or secondary diagnosis (ICD-9-CM 381-382). Infants aged 3 weeks to 12 months and 36 months were included in the analysis for bronchiolitis and OM, respectively, as these are the ages that infants are most susceptible to these illnesses and therefore outcome misclassification due to misdiagnosis will be minimized. We excluded cases less than 3 weeks of age to increase the likelihood that infants in our analyses left the hospital and were exposed to $PM_{2.5}$. We also excluded infants born with birth defects (4%) or whose maternal birth address could not be successfully geocoded (1.3%). The Institutional Review Boards of the University of California at Irvine, Boston University, and the Massachusetts Department of Public Health approved the research.

Exposure Assessment

$PM_{2.5}$ exposures were modeled using satellite remote sensing, meteorological and land use data. The exposure model is described in detail by Girguis et al.(28). Briefly, aerosol optical depth (AOD) measured by satellite instrument was used to estimate $PM_{2.5}$ exposure. AOD is the integral of particle light extinction coefficients from the surface to the top of the atmosphere; it is related to the loadings of fine particles in the atmosphere and is a strong predictor of ground-level $PM_{2.5}$ concentrations as most fine particles are emitted and confined in the boundary layer.

In this study, AOD measurements (available from 9 am to 3 pm local time) were averaged to generate daily mean AOD estimates (29).

We developed a linear mixed effects model with 24-hour average EPA $PM_{2.5}$ measurements from 2001 to 2009 as the dependent variable and AOD, meteorological fields and land use variables as predictors. The model incorporates day-specific random intercepts and slopes for AOD, temperature, and wind speed to account for the temporally varying relationship between $PM_{2.5}$ (based on fixed ground monitors) and AOD (30). This model was run annually for a 4 km modeling grid covering the spatial extent of Massachusetts to estimate daily $PM_{2.5}$ concentrations from 2001 to 2009. Birth addresses of cases were geocoded to the street level and assigned to a 4km grid. Daily $PM_{2.5}$ estimates were assigned to 98% of Massachusetts births included in our study according to their birth grid and dates of exposure.

Covariates.

For the case-crossover study design, each case serves as his/her control. Therefore, only variables that change over short time periods necessitate control in the analysis: temperature, humidity, barometric pressure and whether the event (index) or referent date falls on a holiday. Temperature was obtained at 1 km resolution using methods described by Kloog et al (31). Humidity and barometric pressure were obtained from the NLDAS Phase 2 for each 14km grid of the geographic study location (32). The following national holidays were considered: New Year's Day, Independence Day, Thanksgiving Day, Christmas Day, Memorial Day, and Labor Day.

Statistical Analysis

Conditional logistic regression models were used to calculate odds ratios (ORs) and 95% confidence intervals (CI) for bronchiolitis and OM medical encounters per 10-μg/m^3 increase in $PM_{2.5}$. Hospitalizations, emergency department visits, and observational stays were combined into a single analysis as these cases are similar in etiology and symptoms. Analyses were run separately for emergency department and observational stays versus hospitalizations, since a primary or secondary hospitalization diagnosis might represent more severe cases, which may have different etiologies from those taken to the emergency department or admitted for observational stays.

We used a semi-symmetric bidirectional referent design (33-34) with narrow referent windows to minimize bias due to seasonal and longer-term confounding. Because $PM_{2.5}$ emissions differ by day of the week, referent days were selected to be the same day of the week as the index period, allowing for the referent day to be selected 7 days before or after the clinical event. The semi-symmetric bidirectional referent design randomly assigns an eligible referent day either before or after the clinical encounter. An offset term of log 2 was assigned to referent observations for which one of the potential referent days fell outside the start and end date of our study when $PM_{2.5}$ estimates were not available (35). The offset for all other referent and index days was 0. In order to investigate both the viral incubation and replication periods, we examined the influence of $PM_{2.5}$ on (1) symptom exacerbation using very short exposure lags of 0 and 1 day (for index and referent days), which fall during the viral replication period and (2) susceptibility to infection using longer exposure lags of 4 and 7 days, which span across the viral incubation period. The estimated timeline for respiratory synctal virus (RSV), a common cause of infant bronchiolitis and OM, suggests that after infection, viral incubation occurs for 5 days

and subsequently viral replication occurs for two more days (Figure 2.1). It is suspected that 8 days after infection, symptoms peak and this is the time of a probable clinical encounter (36).

We assessed effect modification by gestational age, birth weight, season of diagnosis, subsequent clinical encounter, insurance payer codes, median income of census block group, infant sex, breastfeeding initiation in hospital at birth, age of infant at time of clinical encounter and maternal race to determine susceptible subgroups. We did this by stratifying our analyses according to each susceptibility variable of interest and obtaining the p-value for the interaction term of the variable of interest and $PM_{2.5}$ by testing product-interaction terms to determine if differences exist between strata. Information on potential effect modifiers was obtained from birth and hospital records.

We also modeled $PM_{2.5}$ at a 1 km resolution (using methods similar to those described above). A sensitivity analysis using 1km $PM_{2.5}$ predictions was run to assess the existence of exposure misclassification due to spatial resolution of exposure prediction models.

RESULTS

Of the 20,017 first time primary or secondary bronchiolitis cases and 42,336 OM cases, respectively, 59.9% and 55.7% were male, 75.8% and 76.5% were born to mothers who received adequate prenatal care, and 72.4% and 71.3% had mothers who initiated breastfeeding in the hospital during birth (Table 2.1).

Due to the presence of cloud or snow, the satellite based exposure models did not produce estimates for all days and locations. Day specific $PM_{2.5}$ values were missing for approximately 10% and 15% of index and referent days for bronchiolitis and OM, respectively. Mean $PM_{2.5}$ values ranged between 9.56-9.76 $\mu g/m^3$ for each lag. With the exception of 1 day lag

among the OM cases, mean and median $PM_{2.5}$ levels were slightly elevated during index versus referent day. Distributions of $PM_{2.5}$ levels during index and referent days are presented in Table 2.2 as the difference between the index and referent measures (referent value subtracted from the index value) for each lag. The difference between index and referent mean $PM_{2.5}$ exposure was between 0.01 and 0.17 $\mu g/m^3$ for bronchiolitis and -0.08 and 0.06 $\mu g/m^3$ for OM. The interquartile range of the difference was between 8.03-8.20 $\mu g/m^3$ and standard deviation of the difference was between 7.21-7.64 $\mu g/m^3$.

Increased $PM_{2.5}$ exposure 1 day prior to clinical encounter (lag 1) is associated with increased risk of bronchiolitis hospitalization (OR = 1.07, 95% CI: 1.03, 1.11) (Table 2.3). The adjusted OR was slightly elevated for acute $PM_{2.5}$ exposure 0 and 4 days prior to hospitalization (lag 0 OR = 1.04, 95% CI: 0.99, 1.08; lag 4 OR=1.03, 95% CI: 0.98, 1.07). The association between bronchiolitis hospitalization and $PM_{2.5}$ exposure 7 days prior to hospitalization (lag 7) was null (OR = 1.00, 95% CI: 0.96, 1.05). The adjusted OR for OM clinical encounter 4 days prior to hospitalization (OR = 1.02, 95% CI: 0.99, 1.05) and 7 days prior to clinical encounter (OR = 1.01, 95% CI: 0.99, 1.04) were slightly elevated. Odds ratios for the day of clinical encounter (OR = 1.00, 95% CI: 0.97, 1.02) and 1 day prior to clinical encounter (OR = 0.97, 95% CI: 0.95, 1.00) for OM were null and inverted with narrow confidence intervals.

Overall, we found elevated ORs for lags 0, 1 and 4 days for infant bronchiolitis and lags 4 and 7 days for OM. Results were similar when using only emergency room visits and observational stays clinical encounters. As such, we report stratified results from all clinical encounters for these specific lags to identify potentially susceptible subgroups. We found that ORs for cold months were similar to warm months across all lags for both infant bronchiolitis and OM (Tables 2.4 and 2.5). Further stratified analyses suggest preterm infants are at increased

risk (OR = 1.17, 95% CI: 1.08, 1.28) for bronchiolitis hospitalization due to increases in $PM_{2.5}$ 1 day prior to clinical encounter compared to full term infants (OR = 1.04, 95% CI: 0.99, 1.09, p-value for interaction; 0.018), but not 4 days prior to hospitalization (Table 2.4). For OM, we also found evidence that preterm infants were at increased risk of OM both 4 days (OR = 1.09, 95% CI: 1.02, 1.16) and 7 days (OR = 1.08, 95% CI: 1.02, 1.15) prior to clinical encounter due to increases in $PM_{2.5}$ compared to full term infants (lag4 OR = 1.01, 95% CI: 0.98, 1.04; lag7 OR = 1.00, 95% CI: 0.97, 1.03, p-interaction: 0.026 and 0.019 for lag 4 and 7, respectively; Table 2.5).

There were no statistical differences in risk for infant bronchiolitis or OM according to birthweight, breastfeeding initiation in the hospital, subsequent clinical encounter, infant sex, age of infant at time of clinical encounter, delivery payment source (insurance), median income of residential block group, or maternal race (Tables 2.4 and 2.5). We found an elevated infant bronchiolitis OR for low birthweight infants, infants who did not breastfeed in the hospital and infants who had multiple clinical encounters 1 day prior to clinical encounter (lag 1), but this was not different from the OR of greater than normal weight infants (p=0.052), infants who did initiate breastfeeding in the hospital (p=0.101) or infants with a single clinical encounter for bronchiolitis (p=0.459).

DISCUSSION

We found that clinical encounters for infant bronchiolitis and OM were positively associated with increases in $PM_{2.5,}$ especially among preterm infants. Increased $PM_{2.5}$ exposure 1 day prior to hospitalization was associated with risk of infant bronchiolitis, and infants who are preterm are at greater risk compared to full term infants. We observed an increased risk of OM only for preterm infants exposed to $PM_{2.5}$ at lag 4 or lag 7 days, although the CIs included 1.

We are aware of only one other study that has examined risk of infant bronchiolitis with acute $PM_{2.5}$ exposure (13). This investigation, using a southern California cohort, also found a positive association between $PM_{2.5}$ and infant bronchiolitis among very preterm infants (<29 weeks) using a 3-5 day average lag (OR=1.26; 95% CI: 1.01–1.57) and a 6-8 day average lag (OR= 1.41; 95% CI: 1.11–1.79). Unlike our findings, all other lags investigated in the Karr et al. (13) study yielded negative results (OR=0.96; 95% CI: 0.94–0.99) across all time periods investigated. This may be due to differing exposure assessment methodologies, differences in $PM_{2.5}$ composition in California and Massachusetts, and/or random error. We found no associations between acute $PM_{2.5}$ exposure and risk of OM when stratified by season and distance to major roadways, which is consistent with other studies (37-38).

The effects of increased acute $PM_{2.5}$ exposure on preterm infants have widespread implications as currently more than 1 in 10 babies are born preterm with increasing rates internationally (39). This finding is even more meaningful given that $PM_{2.5}$ levels in Massachusetts are relatively low compared to international levels (40). Future studies are needed to better assess the effects of higher levels of acute $PM_{2.5}$ exposure and risk of infant bronchiolitis or OM clinical encounter on infants, with emphasis on preterm infants. To explain the increased risk observed among preterm infants, we further hypothesize that lungs and ears of preterm infants are not fully developed leaving preterm infants with an increased $PM_{2.5}$ related risk of infant bronchiolitis compared to their full term counterparts (13).

Increased risk of OM among preterm infants was associated with increased $PM_{2.5}$ exposures 4 or 7 days prior to clinical encounter corresponding to the viral replication period, indicating that $PM_{2.5}$ exposure may increase susceptibility to infection among preterm infants There is evidence that $PM_{2.5}$ exposure may decrease mucus clearance by the cilia (41), causing

inflammation leading to increased susceptibility to OM. For infant bronchiolitis, elevated risk was observed 1 or 4 days prior to clinical encounter, corresponding to the estimated viral incubation period, indicating possible influence of exposure on disease susceptibility. $PM_{2.5}$ exposure also may result in epigenetic effects leaving infants more susceptible to infant bronchiolitis or OM (42).

We did note elevated and decreased risk estimates when assessing OM hospitalizations only. We suspect this difference is due to the small number of OM hospitalizations observed. Only 3% of OM cases were hospitalizations, therefore risk may not be accurate and precision estimates are wide. When assessing bronchiolitis hospitalizations alone, we saw similar estimates to our pooled analysis.

We used satellite data to obtain fine spatial distribution estimates of $PM_{2.5}$ for all of Massachusetts. Using exposure models with 4km exposure resolution, we performed additional case-crossover analyses using 1km exposure models (43-45). We present findings from the 4 km model results here because they provided a better fit to the health data according to Akaike Information Criterion and more realistic exposure distributions compared to the 1 km exposure models (with fewer outliers). Epidemiological results for the 1km model were similar to those for the 4km model (Table 2.6).

Our study only included infants who were born in Massachusetts and subsequently had a clinical encounter for infant bronchiolitis or OM. However, we do not have reason to believe our findings could not be generalized to children born in other geographic regions with similar $PM_{2.5}$ levels, especially since similar results were seen in California (13), Italy (15), and Canada (38).

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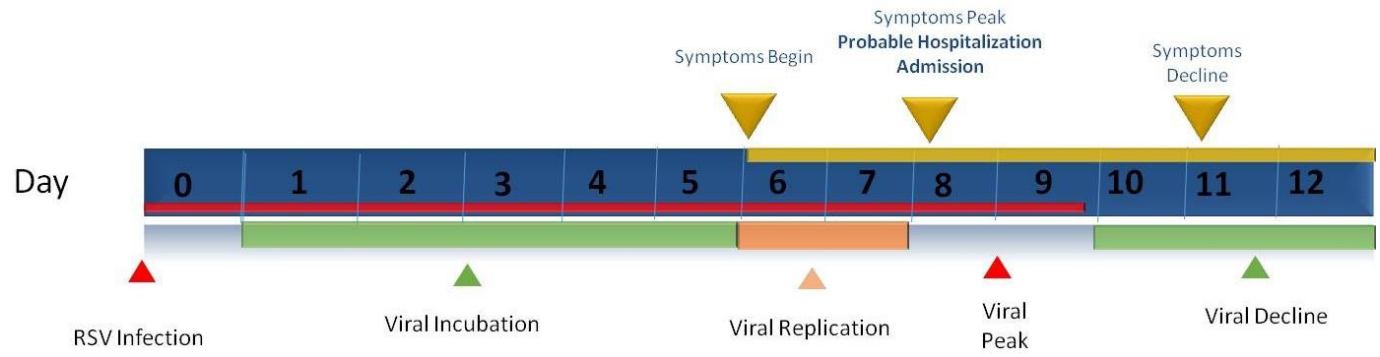

Figure 2.1. Respiratory Syncytial Virus (RSV) Timeline.

$PM_{2.5}$ will be measured 1(lag 7), 4 (lag4), 7 (lag1) and 8 (lag 0) days after estimated respiratory syncytial virus (RSV) infection.

Table 2.1. Demographic Characteristics of Infant Bronchiolitis and Otitis Media Cases Diagnosed in Massachusetts, 2001-2009 Included in Analysis.

	Bronchiolitis n (%)[a]	**Otitis Media** n (%)[a]
Total Cases	20,017	42,336
Infant Sex		
Male	11,985 (59.9)	23,591 (55.7)
Female	8,032 (40.1)	18,745 (44.3)
Maternal Age		
<20 years	2,057 (10.3)	4,997 (11.8)
21-24 years	4,356 (21.8)	9,913 (23.4)
25-29 years	4,796 (23.9)	10,316 (24.4)
30-34 years	5,204 (26.0)	10,395 (24.5)
35+ years	3,604 (18.0)	6,715 (15.9)
Parity		
0	6,923 (34.6)	19,021 (44.9)
1	7,362 (36.8)	13,902 (32.8)
2 or more	5,679 (28.3)	9,345 (22.1)
Missing	53 (0.2)	68 (0.2)
Adequacy of Prenatal Care		
Adequate	15,163 (75.8)	32,406 (76.5)
Intermediate	3,936 (19.7)	8,261 (19.5)
Inadequate	617 (3.1)	1263 (2.9)
Unknown	251 (1.2)	299 (0.7)
None	50 (0.2)	107 (0.2)
Smoking During Pregnancy		
Yes	2,445 (12.2)	5,106(12.1)
No	17,550 (87.7)	37,190 (87.9)
Missing	22 (0.1)	40 (0.1)
Drinking During Pregnancy		
Yes	311 (1.6)	670 (1.6)
No	19,684 (98.3)	41,629 (98.3)
Missing	22 (0.1)	37 (0.1)
Season of Conception		
Winter	3,307 (16.5)	10,440 (24.7)
Spring	4,372 (21.8)	11,078 (26.2)
Summer	6,438 (32.2)	10,898 (25.7)
Fall	5,892 (29.4)	9,902 (23.4)
Missing	8 (0.1)	18 (0.1)
Gestational Age		
>37 weeks	17,123 (85.5)	38,067 (89.9)
≥37-32 weeks	2,266 (11.3)	3,559 (8.4)
<32 weeks	620 (3.1)	692 (1.6)
Missing	8 (0.1)	18 (0.1)
Small for Gestational Age		

Yes	2,484 (12.4)	4,855 (11.5)
No	17,436 (87.1)	37,288 (88.1)
Missing	97 (0.5)	193 (0.5)
Maternal Race/Ethnicity		
Non-Hispanic White	12,012 (60.0)	25,900 (61.2)
Non-Hispanic Black	2,003 (10.0)	4,099 (9.7)
Hispanic	4,642 (23.1)	9,301 (22.0)
Asian/Pacific Islander	798 (3.9)	1,729 (4.1)
Other	547 (2.7)	1,282 (3.0)
Missing	15 (0.1)	25 (0.1)
Maternal Education		
$<12^{th}$ grade	3,549 (17.7)	7,714 (18.2)
High school graduation	6,489 (32.4)	14,892 (35.1)
Some college	9,945 (49.7)	19,670 (46.4)
Missing	34 (0.1)	60 (0.1)
Breastfeeding		
Yes	14,488 (72.4)	30,170 (71.3)
No	5,475 (27.4)	12,084 (28.5)
Missing	54 (0.3)	82 (0.2)
Maternal Language Preference		
English	17 095 (85.4)	35,797 (84.5)
Spanish	1,832 (9.2)	3,682 (8.7)
Portuguese	576 (2.9)	1,621 (3.8)
Other	442 (2.2)	1,121 (2.6)
Missing	72 (0.4)	115 (0.3)
Household Income		
<$20,000	1,502 (7.5)	3,091 (7.3)
$20,000-$70,000	11,220 (56.1)	24,865 (58.7)
≥$70,000	7,291 (36.4)	14,372 (34.0)
Missing	4 (0.02)	8 (0.02)
Delivery Source of Payment		
Health Maintenance Organization	9,071 (45.3)	18,014 (42.5)
Medicaid/CommonHealth	7,467 (37.3)	17,143 (40.5)
Other	3,428 (17.1)	7,097 (16.8)
Missing	51 (0.3)	82 (0.3)

[a]Percentages may not sum to 100% due to rounding.

Table 2.2, Distribution of $PM_{2.5}$ as the Difference Between the Index[a] and Referent[b] Measures for Each Lag, 2001-2009.

$PM_{2.5}$ µg/m^3	N	Mean	Standard Deviation	Median	Interquartile Range
Bronchiolitis					
Lag 0	16,359	0.17	7.34	0.13	8.20
Lag 1	16,357	0.06	7.28	0.48	8.17
Lag 4	16,281	0.11	7.37	0.10	8.11
Lag 7	16,295	0.01	7.21	-0.02	8.10
Otitis Media					
Lag 0	37,040	0.01	7.64	0.03	8.11
Lag 1	37,114	-0.08	7.50	-0.06	8.12
Lag 4	37,090	0.06	7.61	0.08	8.23
Lag 7	37,117	0.03	7.63	0.03	8.03

[a]Index days are days lagged in reference to date of clinical encounter of a case.

[b]Referent day for each case is randomly assigned as one week before or after index day.

Table 2.3. Associations Between 10 ug/m^3 Increase in $PM_{2.5}$ and Infant Bronchiolitis and Otitis Media on the Day of Clinical Encounter (Lag 0), One Day Prior to Clinical Encounter (Lag 1), Four Days Prior to Clinical Encounter (Lag 4) or Seven Days Prior to Clinical Encounter (Lag 7).

OR (95%CI)	Lag 0	Lag 1	Lag 4	Lag 7
Infant Bronchiolitis				
Crude Model	1.02 (0.98, 1.07)	1.07 (1.03, 1.11)	1.04 (1.00, 1.09)	1.00 (0.96, 1.05)
Adjusted Model[a]	1.03 (0.98, 1.07)	1.07 (1.03, 1.11)	1.04 (0.99, 1.08)	1.00 (0.96, 1.05)
Emergency Room and Observational Stays Only	1.01 (0.96, 1.06)	1.08 (1.02, 1.13)	1.03 (0.98, 1.09)	1.03 (0.98, 1.09)
Hospitalizations Only	1.05 (0.98, 1.12)	1.06 (0.99,1.13)	1.05 (0.98, 1.13)	0.95 (0.88, 1.02)
Otitis Media				
Crude Model	1.00 (0.98, 1.03)	0.97 (0.95, 1.00)	1.02 (0.99, 1.05)	1.01 (0.98, 1.03)
Adjusted Model[a]	1.00 (0.97, 1.02)	0.97 (0.95, 1.00)	1.02 (0.99, 1.05)	1.01 (0.99, 1.04)
Emergency Room and Observational Stays Only	0.97 (0.95, 1.00)	0.99 (0.96, 1.02)	1.02 (0.99, 1.05)	1.01 (0.99, 1.04)
Hospitalizations Only	1.14 (0.99, 1.32)	0.98 (0.84, 1.14)	1.08 (0.93, 1.25)	0.97 (0.84, 1.12)

[a] Model adjusted for lagged temperature, barometric pressure, humidity, and holiday indicator.

Table 2.4. Associations Between 10 ug/m^3 Increase in $PM_{2.5}$ and Infant Bronchiolitis One Day Prior to Clinical Encounter (Lag 1) and Four Days Prior to Clinical Encounter (Lag 4) Stratified by Susceptibility Risk Factors.

OR (95%CI)[a]	Lag1	p-interaction[b]	Lag 4	p-interaction[b]
Season of Dx[c]				
Cold	1.08 (1.03, 1.13)	0.188	1.04 (1.00, 1.09)	0.489
Warm	1.00 (0.80, 1.24)	-	1.01 (0.90, 1.13)	-
Gestational Age				
Term >37 weeks	1.04 (0.99, 1.09)	0.018	1.04 (1.00, 1.09)	0.978
Preterm ≤ 37 weeks	1.17 (1.08, 1.28)	-	1.04 (0.97, 1.13)	-
Birthweight				
Low Birth Weight <2500 g	1.12 (0.98, 1.27)	0.052	1.09 (0.95, 1.24)	0.595
Normal Weight ≥2500 g	1.06 (1.02, 1.11)	-	1.03 (0.99, 1.08)	-
Breastfeeding Initiation in Hospital at Birth				
Yes	1.05 (0.99, 1.10)	0.101	1.03 (0.98, 1.08)	0.531
No	1.13 (1.05, 1.22)	-	1.06 (0.97, 1.15)	-
Subsequent Infant Bronchiolitis Clinical Encounter				
No	1.07 (1.02, 1.12)	0.459	1.04 (0.99, 1.07)	0.375
Yes	1.15 (1.01, 1.31)	-	0.98 (0.85, 1.13)	-
Infant Sex				
Male	1.08 (1.02, 1.14)	0.716	1.05 (1.00, 1.11)	0.501
Female	1.06 (0.99, 1.13)	-	1.02 (0.95, 1.09)	-
Age of Infant at Time of Clinical Encounter				
3 weeks-6months	1.06 (0.99, 1.12)	0.650	1.09 (1.02, 1.15)	0.027
6 months-1year	1.08 (1.02, 1.15)	-	0.99 (0.93, 1.05)	-
Delivery Payment Source				
Health Maintenance Organization	1.04 (0.98, 1.10)	-	1.04 (0.98, 1.10)	-
Medicaid/CommonHealth	1.09 (1.02, 1.17)	0.331	1.04 (0.97, 1.11)	0.979
Other	1.11 (1.00, 1.23)	0.330	1.04 (0.96, 1.15)	0.842
Median Income of Census Block Group				
<$20 000	0.97 (0.83, 1.14)	0.242	0.97 (0.83, 1.14)	0.586
$20 000-$70 000	1.09 (1.03, 1.15)	0.534	1.04 (0.99, 1.10)	0.987
>$70 000	1.06 (0.99, 1.14)	-	1.04 (0.97, 1.12)	-
Maternal Race				
Non-Hispanic White	1.06 (1.00, 1.12)	-	1.06 (1.01, 1.12)	-
Non-Hispanic Black	1.11 (0.97, 1.27)	0.441	1.13 (0.99, 1.30)	0.368

Hispanic	1.06 (0.97, 1.15)	0.845	0.96 (0.88, 1.05)	0.054
Asian	1.27 (1.02, 1.57)	0.128	0.98 (0.79, 1.21)	0.432
Other	1.07 (0.83, 1.36)	0.805	0.99 (1.75, 1.30)	0.608

[a] Model adjusted for lagged temperature, barometric pressure, humidity, and holiday indicator.
[b]p-interaction generated from interaction term of susceptibility risk factor and $PM_{2.5}$ in full model.
[c]Warm months are May through October and cold months are January through April and November through December

Table 2.5. Associations Between 10 ug/m^3 Increase in $PM_{2.5}$ and Otitis Media Four Days (Lag 4) and Seven Days (Lag 7) Prior to Clinical Encounter Stratified by Susceptibility Risk Factors.

OR (95% CI)[a]	Lag4	p-interaction[b]	Lag 7	p-interaction[b]
Season of Dx[c]				
Cold	1.02 (0.98, 1.06)	0.963	1.01 (0.97, 1.04)	0.879
Warm	1.01 (0.98, 1.04)	-	1.02 (0.98, 1.06)	-
Gestational Age				
Term >37 weeks	1.01 (0.98, 1.04)	0.026	1.00 (0.97, 1.03)	0.019
Preterm ≤ 37 weeks	1.09 (1.02, 1.16)	-	1.08 (1.02, 1.15)	-
Birthweight				
Low Birth Weight <2500 g	1.01 (0.91, 1.12)	0.868	0.97 (0.88, 1.08)	0.377
Normal Weight ≥2500 g	1.02 (0.99, 1.05)	-	1.01 (0.99, 1.04)	-
Breastfeeding Initiation in Hospital at Birth				
Yes	1.02 (1.00, 1.06)	0.508	1.02 (0.99, 1.05)	0.555
No	1.01 (0.96, 1.06)	-	1.00 (0.95, 1.05)	-
Subsequent Otitis Media Clinical Encounter				
No	1.02 (0.99, 1.05)	0.624	1.01 (0.98, 1.04)	0.678
Yes	0.99 (0.82, 1.20)	-	0.99 (0.81, 1.20)	-
Infant Sex				
Male	1.04 (1.00, 1.08)	0.205	0.99 (0.96, 1.03)	0.146
Female	1.00 (0.96, 1.04)	-	1.04 (1.00, 1.08)	-
Age of Infant at Time of Clinical Encounter				
0-1 year	1.02 (0.99, 1.07)	-	1.04 (1.00, 1.08)	-
1-2 years	1.02 (0.98, 1.07)	0.972	0.98 (0.94, 1.03)	0.0555
2-3 years	1.00(0.93, 1.07)	0.489	1.00(0.93, 1.07)	0.270
Delivery Payment Source				
Health Maintenance Organization	1.02 (0.98, 1.07)	-	0.99 (0.95, 1.04)	-
Medicaid/CommonHealth	1.01 (0.97, 1.17)	0.593	1.04 (1.00, 1.09)	0.097
Other	1.03 (0.97, 1.11)	0.848	0.99 (0.93, 1.06)	0.958
Median Income of Census Block Group				
<$20 000	1.01 (0.91, 1.12)	0.949	1.04 (0.94, 1.15)	0.419
$20 000-$70 000	1.01 (0.98, 1.05)	0.764	1.01 (0.97, 1.04)	0.446
>$70 000	1.03 (0.98, 1.08)	-	1.01 (0.97, 1.06)	-
Maternal Race				
Non-Hispanic White	1.03 (1.00, 1.07)	-	1.00 (0.96, 1.03)	-
Non-Hispanic Black	1.00 (0.92, 1.09)	0.557	1.03 (0.94, 1.12)	0.575

Hispanic	1.02 (0.96, 1.08)	0.700	1.03 (0.98, 1.09)	0.247
Asian	1.02 (0.89, 1.16)	0.810	1.02 (0.89, 1.16)	0.776
Other	0.89 (0.76, 1.05)	0.136	1.12 (1.95, 1.31)	0.149

[a] Model adjusted for lagged temperature, barometric pressure, humidity, and holiday indicator.
[b]p-interaction generated from interaction term of susceptibility risk factor and $PM_{2.5}$ in full model.
[c]Warm months are May through October and cold months are January through April and November through December

Table 2.6. Associations Between 10 ug/m^3 Increase in $PM_{2.5}$ and Infant Bronchiolitis and Otitis Media on the Day of Clinical Encounter (Lag 0), One Day Prior to Clinical Encounter (Lag 1), Four Days Prior to Clinical Encounter (Lag 4) or Seven Days Prior to Clinical Encounter (Lag 7) Using 1km Exposure Model.

OR (95%CI)	Lag 0	Lag 1	Lag 4	Lag 7
Infant Bronchiolitis				
Crude Model	1.01 (0.92, 1.10)	1.06 (0.97, 1.15)	1.00 (0.91, 1.08)	1.00 (0.92, 1.09)
Adjusted Model[a]	1.01 (0.93, 1.10)	1.06 (0.97, 1.15)	1.00 (0.91, 1.08)	1.00 (0.92, 1.09)
Otitis Media				
Crude Model	1.02 (0.97, 1.08)	1.01 (0.95, 1.06)	1.04 (0.98, 1.10)	0.97 (0.92, 1.03)
Adjusted Model[a]	1.02 (0.96, 1.08)	1.01 (0.95, 1.06)	1.04 (0.98, 1.10)	0.98 (0.92, 1.03)

[a] Model adjusted for lagged temperature, barometric pressure, humidity, and holiday indicator.

CHAPTER 3

Chronic $PM_{2.5}$ Exposure and Risk of Infant Bronchiolitis and Otitis Media Clinical Encounters

ABSTRACT

Chronic exposure to particulate matter less than 2.5μm in diameter ($PM_{2.5}$) can cause an inflammatory response leaving infants more susceptible to illness. Our objective is to estimate associations of chronic $PM_{2.5}$ exposure on infant bronchiolitis and otitis media (OM) clinical encounters. We obtained all first time bronchiolitis (n=18,029) and OM (n=40,042) clinical encounters among children less than 12 and 36 months of age, respectively, diagnosed from 2001-2009 in Massachusetts and two controls per case matched on birthdate and gestational age from the Pregnancy to Early Life Longitudinal (PELL) data linkage system. We applied conditional logistic regression to estimate odds ratios (OR) and confidence intervals (CI) per 2-$\mu g/m^3$ increase in lifetime $PM_{2.5}$ exposure. Effect modification was assessed by age of the infant at clinical encounter, gestational age, frequency of clinical encounter, and income. We also examined the association between residential distance to major roadways, traffic density, and risk of infant bronchiolitis and OM. $PM_{2.5}$ was not associated with infant bronchiolitis (OR = 1.02, 95% CI: 1.00, 1.04) and inversely associated with OM (OR= 0.97, 95% CI: 0.95, 0.99). There was no evidence of effect modification. Compared to infants living near low traffic density, infants living near the highest traffic density had elevated risk of bronchiolitis (OR = 1.23, 95% CI: 1.14, 1.31) but not OM (OR = 0.98, 95% CI: 0.93, 1.02) clinical encounter. We did not find strong evidence to support an association between $PM_{2.5}$ exposure and infant bronchiolitis or OM. Risk of bronchiolitis was increased among infants living near high traffic density.

INTRODUCTION

Infant bronchiolitis is a lower respiratory tract infection and the leading cause of hospitalizations among children during the first year of life (1). Most bronchiolitis cases are caused by viral infection, specifically respiratory syncytial virus (RSV) infection. Although most infants are RSV seropositive, some infants experience mild symptoms whereas others are hospitalized (2). Otitis media (OM), or inflammation of the middle ear, is one of the most frequent infections among children less than 3 years of age (3), the most common cause for medical care besides a healthy child visit, and a major cause for antibiotic use within the first few years of life (4). OM can be caused by viral and bacterial infections. Much like bronchiolitis, OM results from a complex combination of pathogens, environmental exposures (such as tobacco smoke and indoor wood burning), and heredity (6-9).

Symptomatic infants with bronchiolitis or OM have shown immunoregulatory and proinflammatory responses during the course of their illness (10-12). Toxicological studies have demonstrated that chronic exposure to traffic related air pollution, such as particulate matter with a diameter of 2.5 microns or less ($PM_{2.5}$) can trigger an inflammatory response in rats (13-15) and humans (16). To date, there have been only a few studies investigating chronic $PM_{2.5}$ exposure and bronchiolitis (17-19) or OM (20-21), indicating evidence of a possible association (17, 20-21). Estimating the association between $PM_{2.5}$ exposure and clinical encounters for infant bronchiolitis and OM may help better elucidate factors contributing to the occurrence and symptom severity of these common child morbidities.

In this study we estimated associations between chronic $PM_{2.5}$ exposure and risk of bronchiolitis during infancy and OM during early childhood using data from the Pregnancy to Early Life Longitudinal (PELL) data linkage system, which includes all Massachusetts (MA) birth records linked to subsequent clinical encounters (hospitalizations, emergency room visits, and observational stays). To rigorously control for confounding, we conducted analyses using two control groups. The main analysis accounts for temporal trends and baseline health of the children based on gestational age and birthdate using matched controls. The secondary analysis uses sibling controls to better control for confounding by unmeasured time-invariant factors that are shared among family members, such as parental propensity to seek health care, indoor pollution (such as wood burning), and genetics.

METHODS

Study Population

Eligible study participants were obtained from PELL, a partnership between the Boston University School of Public Health, the Massachusetts (MA) Department of Public Health, and the Centers for Disease Control and Prevention (22). Cases of infant bronchiolitis were selected among infants born between 2001-2008 and are defined as the first clinical encounter (hospitalization, observational stay, or emergency department visit) with a primary or secondary diagnosis of infant bronchiolitis (*International Classification of Diseases,* Ninth Revision, Clinical Modification (ICD-9-CM) 466.0-466.1) experienced by infants greater than 3 weeks and less than 12 months of age. Cases of OM were selected among children born between 2001-2006 and are defined as the first clinical encounter with a primary or secondary diagnosis (ICD-9-CM 381-382) experienced by children greater than 3 weeks and less than 36 months of age. A minimum age of 3 weeks increases likelihood that infants have left the hospital and are exposed

to $PM_{2.5}$ at home. Children are most susceptible to infant bronchiolitis and OM up until 12 months (1) and 36 months of age (3), respectively. Cases with a different zip code at birth and time of clinical encounter (20%) were excluded to minimize exposure misclassification due to residential mobility. The Institutional Review Boards of the University of California at Irvine and the MA Department of Public Health approved this research.

To assess the influence of chronic $PM_{2.5}$ exposure on infant bronchiolitis and OM, a nested case-control design using random controls matched on birth date and gestational age was used. Two matched controls were selected for each case among infants with a non-respiratory related clinical encounter in PELL. Controls were eligible if they did not have a bronchiolitis or OM event before they were the same age as the case when diagnosed, were born within 6 days of the case, had the same gestational age (week) as the case, and had the same zip code at time of birth and time of PELL clinical encounter.

A secondary nested case control analysis was performed with sibling controls, utilizing PELL's data system tracking of families over time. Because important individual level risk factors such as indoor air pollution (including tobacco smoke), house dust, breastfeeding intensity and duration, frequency of wood burning in the home, and proclivity of parents to take their children to the emergency department are not assessed in PELL, using a sibling control helps adjust for these confounders as these variables are likely to be similar among siblings. Siblings of cases were selected if they were single births with discordant outcomes from time of birth up until the case age. Sibling matched controls is a technique frequently used in cancer epidemiology but has rarely been used in air pollution epidemiology (23).

Among all cases and controls, we excluded infants born with birth defects (4%) or whose birth address could not be successfully geocoded (2%).

Exposure Assessment

We developed a three-stage statistical model to predict $PM_{2.5}$ throughout Massachusetts at a 4km resolution. For details see Girguis et al. (24). Briefly, the first stage accounts for the temporally varying relationship between $PM_{2.5}$ and satellite based aerosol optical depth (AOD), a measurement of light transmission through atmospheric aerosols, after adjustment for relative humidity, wind speed, elevation, major roads, forest cover, and point emissions (25-26). The second stage explains the spatially varying relationship between $PM_{2.5}$ and AOD accounting for geographic location. The third stage combines the first and second stage model to estimate daily $PM_{2.5}$ concentrations for grid cells (including those grid cells where AOD data was unavailable) accounting for temporal and spatial variations.

For each case and control, daily $PM_{2.5}$ concentrations were averaged from birth until the age of case at time of clinical encounter. Birth addresses of infants were geocoded to the street level and assigned to a 4km grid cell. Average daily $PM_{2.5}$ values were assigned to each child according to their birth grid cell and dates of exposure. We only included average $PM_{2.5}$ exposure measures of children who had $PM_{2.5}$ measure for over 70% of their exposure window.

Residential distance to major roadways and traffic density were calculated for each case and control. Using geographic information system software (ArcGIS, version 10.0; ESRI), we calculated the shortest distance between each birth address and the nearest Class 1 (limited access highways) or 2 (multilane highways without limited access) road segment to obtain residential distance to major roadways. Traffic density was calculated by summing the annual average daily traffic (AADT) for a 200 meter grid of Class 1 and 2 road segments (27). For further details, see Girguis et al (24).

Covariates

By matching on temporal variables such as birth date and gestational ages, secular time trends are accounted for ensuring children are compared across the same time period for the same duration. The following covariates were considered as potential confounders in the time-matched analysis: plurality, parity, maternal race/ethnicity, maternal education, maternal language preference, delivery payment source, smoking during pregnancy, alcohol consumption during pregnancy, adequacy of prenatal care (measured by the Adequacy of Prenatal Care Utilization Index), marital status, maternal age, breastfeeding initiation in hospital, risky pregnancy, and birthweight. We used geocoded addresses to determine median household income and proportion of homes that use wood for fuel by census block group from the American Community Survey 2006-2010, 5 year estimates. Directed Acyclic Graphs (DAGs) (Figure 3.1) were used for covariate selection and subsequent change-in-estimate procedures (5%) were used to simplify the final model (28-29).

Since sibling control studies only necessitate adjustment for potential confounders that differ between siblings, the following variables were considered for adjustment in the sibling control models: parity, smoking during pregnancy, alcohol consumption during pregnancy, adequacy of prenatal care (measured by the Adequacy of Prenatal Care Utilization Index), maternal age, breastfeeding initiation in hospital, risky pregnancy, birthweight, birth year, and gestational age. Such control selection provides aggressive control for family level risk factors and reduces residual confounding by family-level risk factors.

Statistical Analysis

Conditional logistic regression models with a cluster function for census block group (generalized estimating equation models) were used to estimate odds ratios (ORs) and 95% confidence intervals (CI) for bronchiolitis and OM clinical encounters per 2 $\mu g/m^3$ increase in lifetime $PM_{2.5}$. Bronchiolitis and OM hospitalizations, emergency department visits, and observational stays were combined into a single analysis as most of these cases are similar. A sensitivity analysis was run with hospitalizations only since a primary or secondary hospitalization diagnosis would likely represent more severe cases with comorbidities which may have different etiologies from those taken to the emergency department or admitted for observational stays.

Effect modification was assessed by income, frequency of clinical encounters, gestational age, and age of child at time of diagnosis in the age matched analysis. In the sibling matched analysis, effect modification was assessed by maternal language preference and maternal education. The mean difference of age between siblings was 2.5-3 years; therefore there will be temporal variation in exposure measures due to secular trends and possible residential mobility. To account for variables influenced by where the family lives, a sensitivity analysis was completed using only residentially stable sibling pairs with the same birth zip code. This increases the likelihood that siblings would be taken to the same emergency department or hospital and their medical care and diagnosis criteria would be similar.

Studies have suggested that concentrations of traffic-related pollutants demonstrate consistent pollutant gradients where concentrations fall to background concentrations within a few hundred meters away from roads (30). To assess the relationship between distance to major roadways and risk of bronchiolitis and OM in our cohort, we used penalized splines to model distance (meters). Traffic density was modeled using quartiles.

We also fit a conditional poison regression to model counts of clinical encounter and assess rates of morbidity using the gnm R package (31). Counts of clinical encounters included the total number of bronchiolitis clinical encounters experienced by each child during the first 12 and 36 years of life, respectively. Controls were non-nested randomly selected infants matched on date of birth and gestational age among individuals in PELL with a non-respiratory clinical encounter. Analysis was adjusted for the same variables as the matched analysis above.

We became aware of a difference in geocoding methods conducted by the Massachusetts Department of Public Health (MADPH) starting in 2004. In 2004, instead of geocoding unsuccessful birth address geocodes to the centroid of the zip code, MADPH did not geocode the address at all. To assess the influence of such exposure misclassification, we conducted a sensitivity analysis of just children born from 2004-2009.

RESULTS

Demographic characteristics of cases and controls are presented in Table 3.1. A larger proportion of cases were male (60%) while controls were more evenly distributed across sex. For both cases and controls, the majority of mothers were between 30-34 years old. Among OM cases and controls, 45% were first born infants, while 35% and 45% of bronchiolitis cases and controls, respectively, were first born infants. Approximately half of the mothers attended at least some college and 86-90% of infants were full term.

Mean $PM_{2.5}$ was 9.7 and 9.6 $\mu g/m^3$ for bronchiolitis cases and controls, respectively, and 10.1 and 10.0 $\mu g/m^3$ for OM cases and controls respectively (Table 3.2). The maximum lifetime $PM_{2.5}$ average was between 19.2-19.9 $\mu g/m^3$ and IQR was 2.2 $\mu g/m^3$ for bronchiolitis cases and controls and 1.8 $\mu g/m^3$ for OM cases and controls.

Crude analysis, controlling for the matching factors only, indicated positive associations between lifetime $PM_{2.5}$ exposure and bronchiolitis (OR = 1.05, 95% CI: 1.02, 1.07) and OM (OR = 1.08, 95% CI: 1.06, 1.10) (Table 3.3). After adjusting for risky pregnancy, maternal age, birthweight, smoking during pregnancy, maternal education, adequacy of prenatal care, parity, income and insurance type, the association between $PM_{2.5}$ and bronchiolitis was attenuated (OR = 1.02, 95% CI:1.00, 1.04). Further adjustment for wood burning in the home yielded a null association (OR = 1.00, 95% CI: 0.98, 1.03). After adjustment, an inverse association was observed between OM and $PM_{2.5}$ exposure (OR = 0.96, 95% CI: 0.94, 0.98). When only using hospitalization clinical encounters, adjusted risk estimates were elevated for OM (OR = 1.07, 95% CI: 0.96, 1.19) and bronchiolitis (OR = 1.09, 95% CI: 1.05, 1.13) hospitalizations. Results were similar among infants and children born from 2004-2009.

Results of effect modification by age at time of clinical encounter, income, frequency of clinical encounter and gestational age are displayed in Table 3.4. None of the stratified analyses indicated meaningful differences across groups based on interaction p-values and Wald test p-values (used when comparing more than 2 groups). Infants from households in the lowest quartile of median income by census block group (<$36,543) had stronger associations with increased lifetime exposure to $PM_{2.5}$ and OM (OR = 1.03, 95% CI: 0.96, 1.11) compared to households of the highest quartile of median income by census block group (>$78,929) (OR = 0.92, 95% CI: 0.85, 0.98), but with overlapping confidence intervals. In our analysis modeling the number of clinical encounters experienced for each child, we found no significant association between $PM_{2.5}$ exposure and the rate of bronchiolitis (results not shown).

We found a positive association with traffic density near the home and bronchiolitis, but not with OM (Table 3.5). Compared to individuals living in the least dense quartile, those living

in the second (OR = 1.10, 95% CI: 1.05, 1.17), third (OR = 1.24, 95% CI: 1.17, 1.32) and fourth quartiles (OR = 1.23, 95% CI: 1.14, 1.31) had elevated ORs for bronchiolitis. Similar to results from traffic density analyses, bronchiolitis ORs decrease with increasing residential distance to major roadways (Figure 3.2). There is a negative non-linear association between OM and distance to major roadways (Figure 3.3).

Results of our secondary analysis using sibling matched controls were consistent with the age matched results (Table 3.6). See Appendix A for demographic and exposure distribution of sibling cases and controls. After adjustment for variables that could differ across time, including season of conception, parity, birth year, maternal age, and adequacy of prenatal care, we found no association between lifetime $PM_{2.5}$ exposure and risk of bronchiolitis (OR= 1.00, 95% CI: 0.98, 1.03) or OM (OR = 0.96, 95% CI: 0.91, 1.00). Results were similar for residentially stable sibling pair subgroup analysis. The only evidence of effect modification in the sibling analyses was between maternal education and OM, with higher ORs among children whose mothers had less than a high school education.

DISCUSSION

We report results from our large nested case-control study on the association of $PM_{2.5}$ and traffic related air pollution with bronchiolitis and OM clinical encounters. This study utilizes a unique data linkage system to apply two control groups: a random sample matched on birthdate and gestational age and a sibling matched design to carefully control for common sources of unmeasured confounding. The first study design controls for secular trends in air pollution, while sibling matched controls better control for nurture related factors which are commonly unaccounted for in traditional environmental epidemiology analyses. Our findings suggest little

evidence of consistent associations between chronic $PM_{2.5}$ exposure during early life and bronchiolitis clinical encounter risk, but positive significant associations with residential traffic metrics. We did observe consistently protective associations with OM clinical encounters.

We present both the crude and adjusted results to highlight the influence of confounders. In all analyses, there exists a disparity between the crude and adjusted estimates regardless of matching strategy, especially for OM models. In the time matched analysis, we found that income, as measured by the median income of census block group, shifted the risk estimate across the null for OM and most strongly influenced the effect estimate towards the null for bronchiolitis. To account for correlated income among infants within the same census block group and minimize Berkson Error from utilizing group level data (income and wood burning), we used marginal models by including a cluster function for census block groups. As we only partially controlled for income, we suspect there remains residual confounding by income or socioeconomic status. In the sibling matched analysis, for both infant bronchiolitis and OM, year of birth and parity of the mother at time of birth most strongly shifted the effect estimate towards the null for infant bronchiolitis and across the null for OM, indicating the presence of strong temporal trends. Although we included year of birth in the model to account for temporal trends, we suspect that the sibling analysis may be subject to residual confounding or selection bias by time and birth order (Appendix B)(32).

Analyses of only hospitalization clinical encounters indicated elevated significant associations with infant bronchiolitis hospitalizations and lifetime $PM_{2.5}$ exposure. Infant bronchiolitis hospitalizations made up 36% of our cases (with emergency room visits and observational stays making up 50% and 14%, respectively). Such findings may be due to multiple comparisons or may indicate that $PM_{2.5}$ exposure may be associated with only the most

extreme cases of bronchiolitis. Previous analyses (17-19) which found null and positive associations were conducted with only hospitalization cases. We also examined emergency room and observational stay clinical encounters separately and found risk and precision estimates were similar to our main findings. Our results suggest that future studies would benefit from investigating associations between $PM_{2.5}$ exposure and different clinical encounter types if sufficient data are available.

Final models were adjusted for wood burning. Wood burning in the home is used as an alternate fuel source among individuals living in rural areas and contributes to $PM_{2.5}$ (33-34) (Appendix C). However, it is still unclear if wood burning increases risk of bronchiolitis (35) or OM (6,36) independent of the effects of $PM_{2.5}$. Given that wood burning is associated with risk of bronchiolitis and OM independent of $PM_{2.5}$ (as seen in our study when both variables were included in the model), then wood burning is a classic confounder and estimates generated from models adjusted for wood burning are more accurate.

Our findings are consistent with studies that assessed the association between lifetime $PM_{2.5}$ exposure and risk of infant bronchiolitis. Two of the three previous studies also found null associations between $PM_{2.5}$ and bronchiolitis in Washington, USA and Canada (18-19). Another study conducted in the South Coast Air Basin, a geographic region with high background $PM_{2.5}$ level (mean $PM_{2.5}$ =24 $\mu g/m^3$) detected a positive association between lifetime $PM_{2.5}$ exposure and infant bronchiolitis (OR= 1.09, 95% CI: 1.04, 1.14) (17). The South Coast Air Basin statistical models did not adjust for income, which in our analysis, strongly influenced the effect estimates towards the null. Differences in results also may be due to differing levels and

composition of $PM_{2.5}$ in the South Coast Air Basin compared to Massachusetts (37) or differing exposure assessment methods.

Our findings indicate inverse associations between $PM_{2.5}$ exposure and risk of OM. The direction of the estimate was consistent across epidemiological designs. In Canada, a geographic location with low background $PM_{2.5}$ levels (mean $PM_{2.5}$= 3.9-5.5), associations were similar to ours (OR= 0.91, 95% CI: 0.89, 0.93) when land-use regression was used to model $PM_{2.5}$ (21). The same study, using multipollutant models including $PM_{2.5}$ (measured using inverse distance weight) and wood burning predictions, found positive significant associations (OR= 1.02, 95% CI: 1.01, 1.04) for $PM_{2.5}$ and OM (21). Although this study only accounted for physician visits, which were not accounted for in our study, and utilized more sophisticated measures of wood burning, similar to our study, they did see effect estimates for $PM_{2.5}$ decrease when including wood burning in the model. Another study found a marginal positive association in a Netherlands cohort and a null association in a German cohort between $PM_{2.5}$ and OM among young children living in areas of higher background $PM_{2.5}$ levels (mean $PM_{2.5}$ =16.4 and 13.4 ug/m3) (20).

This study has several strengths, including the large sample size, the ability to link cases within the PELL data system to examine associations using both sibling and time matched controls, control of important confounders such as time, parity, income and wood burning, and the use of sophisticated satellite based $PM_{2.5}$ exposure measures which yield exposure predictions across the entire geographic region of MA. Such exposure assessments reduce risk of differential exposure misclassification compared to previous studies that have relied on measures from stationary monitoring systems to estimate exposure over large areas.

Although satellite based $PM_{2.5}$ exposure describes local exposure near the home. A limitation is incomplete temporal coverage due to snowy conditions (38), satellite error, cloud coverage (39), or broken satellites. Therefore, we only included average measures which had exposure estimates for over 70% of the days assessed (< 10% of observations excluded). Another limitation of this analysis was to the potential for spurious associations due to multiple testing across outcomes and study designs. Lastly, we were limited because we were not able to include primary care visit diagnosis of bronchiolitis or OM which is a likely place for repeat visits and would capture a more stable patient population. Therefore, our findings are only generalizable to hospitalizations, emergency room visits and observational stays. In summary, our results do not support a positive association between PM2.5 exposure and risk of bronchiolitis and OM, although there is suggestion of increased susceptibility among infants born to mothers with lower socioeconomic status and education.

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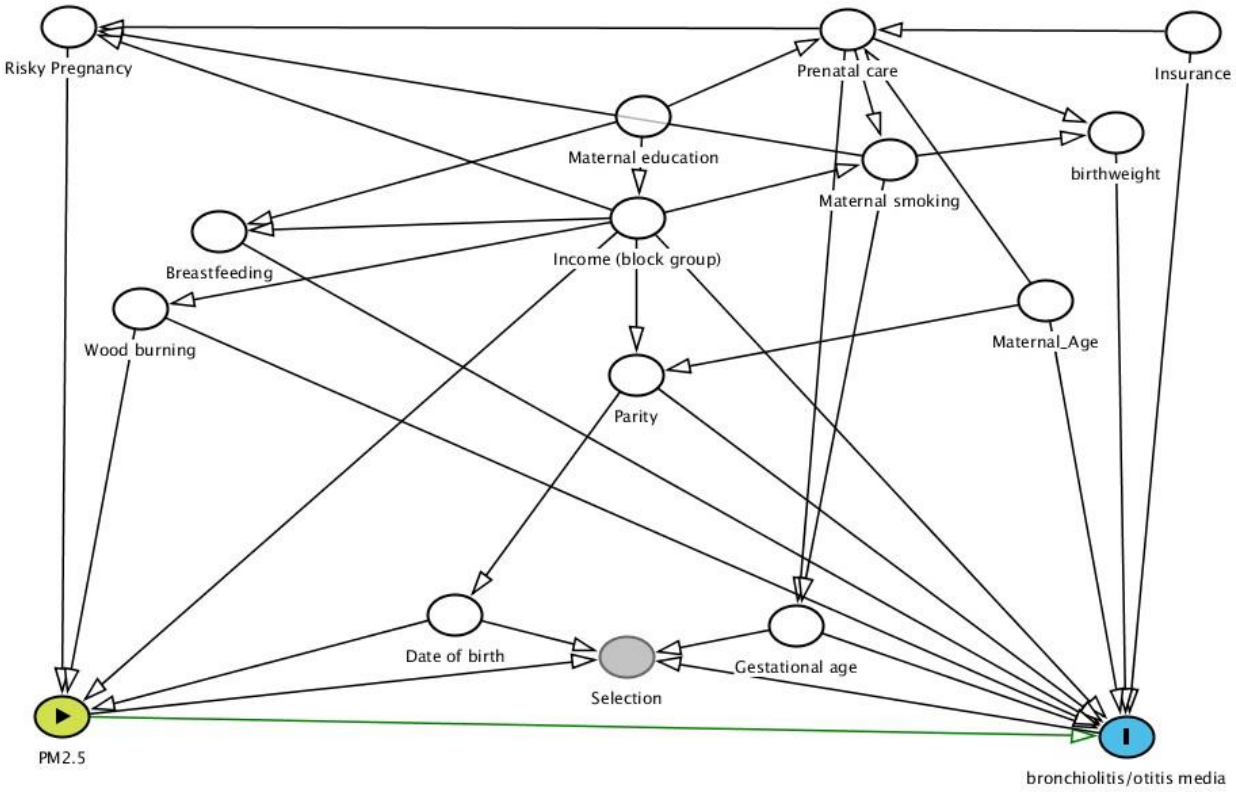

Figure 3.1: Directed Acyclic Graph (DAG) of Birth Date (+/- 6 days) and Gestational Week Matched Case Control Design.

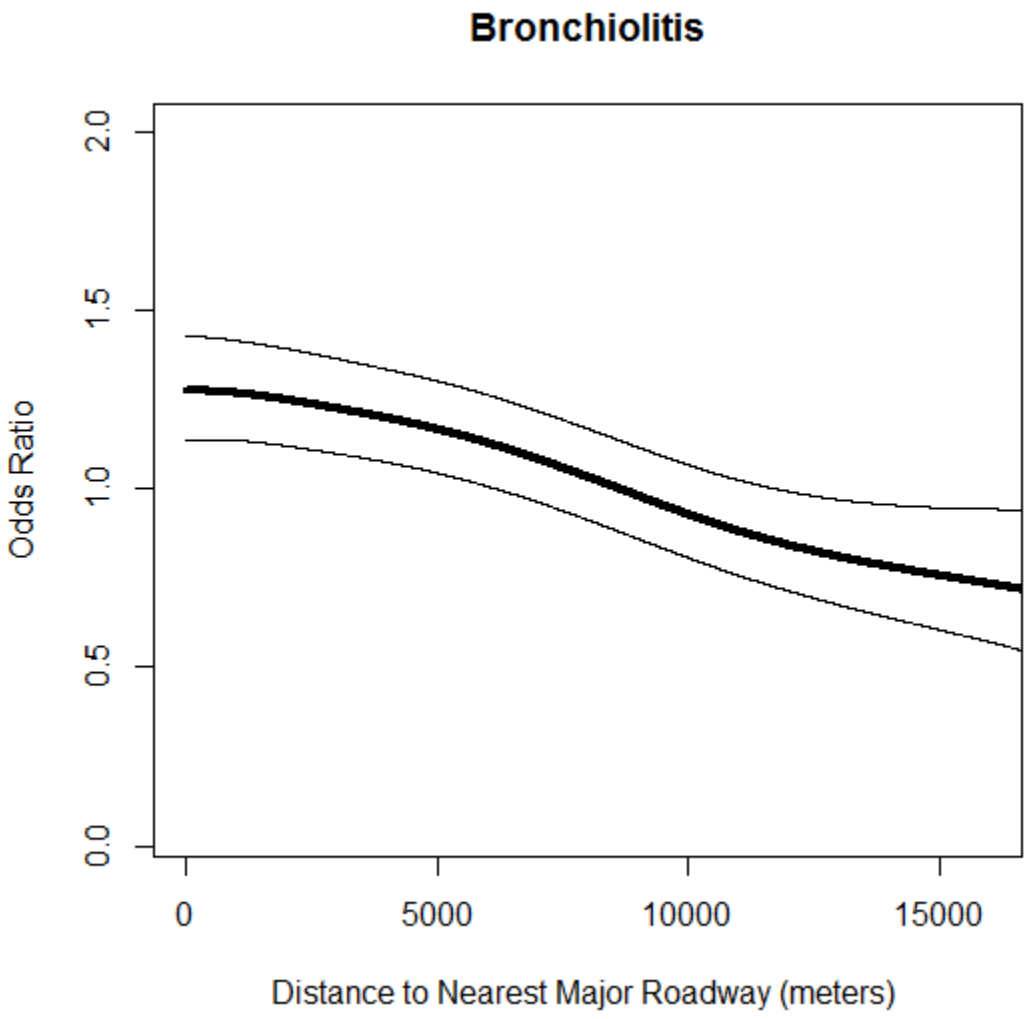

Figure 3.2: Exposure Response Curve Using Penalized Splines to Determine Effect of Distance to Major Roadways and Risk of Infant Bronchiolitis. Association of residential distance to major roadways and risk of infant bronchiolitis showing the adjusted odds ratio (thick solid line) and the 95% confidence interval (thin solid line) using penalized splines to model the association of residential distance to a major roadway and infant bronchiolitis clinical encounter among infants diagnosed in Massachusetts between 2001–2009. Smoothed distance term is significant (P=0.002). Estimates are only presented for residential addresses within continental Massachusetts. All models matched on birth date and gestational week and adjusted for risky pregnancy, maternal age, birthweight, smoking during pregnancy, maternal education, adequacy of prenatal care, parity, income and insurance type Major roadways defined as limited access highways and multi-lane highways (class 1 and 2 roads).

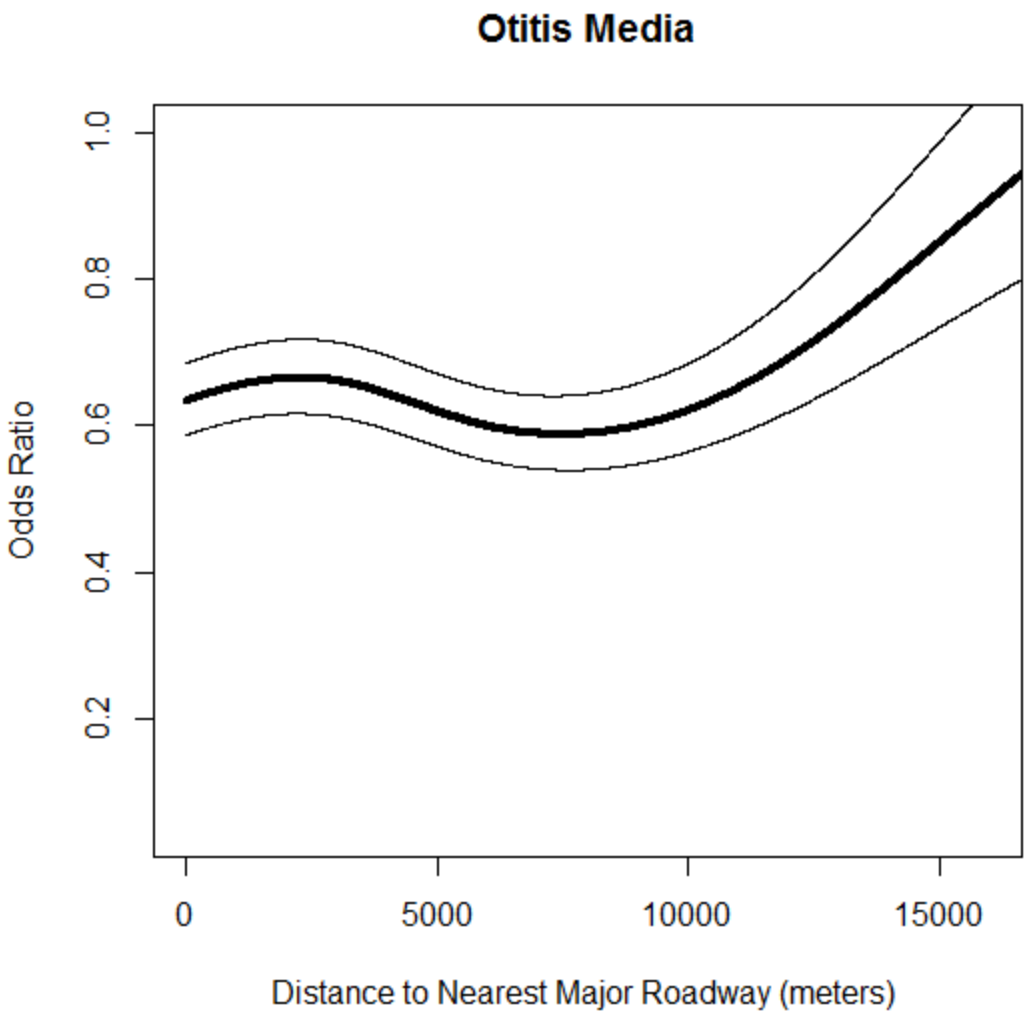

Figure 3.3: Exposure Response Curve Using Penalized Splines to Determine Effect of Distance to Major Roadways and Risk of Otitis Media. Association of residential distance to major roadways and risk of otitis media showing the adjusted odds ratio (thick solid line) and the 95% confidence interval (thin solid line) using penalized splines to model the association of residential distance to a major roadway and otitis media clinical encounter among infants diagnosed in Massachusetts between 2001–2009. Smoothed distance term is significant (P=0.01). Estimates are only presented for residential addresses within continental Massachusetts. All models matched on birth date and gestational week and adjusted for risky pregnancy, maternal age, birthweight, smoking during pregnancy, maternal education, adequacy of prenatal care, parity, income and insurance type Major roadways defined as limited access highways and multi-lane highways (class 1 and 2 roads).

Table 3.1: Demographic Characteristics of Infant Bronchiolitis and Otitis Media Cases Diagnosed in Massachusetts, 2001-2009 and Random Controls Matched on Birthdate (+/-6 days) and Gestational Week, Included in Analysis.

	Bronchiolitis Cases N (%)[a]	Bronchiolitis Controls N (%)[a]	Otitis Media Cases N (%)[a]	Otitis Media Controls N (%)[a]
Total	18,029	35,816	40,042	79,747
Infant Sex				
Male	10,809 (60.0)	18,892 (52.8)	22,336 (59.9)	44,485 (55.8)
Female	7,220 (40.0)	16,924 (47.3)	17,706 (40.1)	35,262 (44.2)
Maternal Age				
<20 years	1,852 (10.3)	2495 (7.0)	4,689 (11.7)	9,347 (11.7)
21-24 years	3,928 (21.8)	5,817 (16.2)	9,342 (23.3)	18,619 (23.4)
25-29 years	4,365 (24.2)	8,593 (24.0)	9,772 (23.4)	19,439 (24.4)
30-34 years	4,700 (26.1)	10,856 (30.1)	9,859 (24.6)	19,657 (24.7)
35+ years	3,184 (17.7)	8,053 (22.5)	6,380 (16.9)	12,685 (15.9)
Parity				
0	6,291 (34.9)	15,987 (44.7)	17,985 (44.9)	35,817 (44.9)
1	6,625 (36.8)	12,315 (34.4)	13,181 (32.9)	26,262 (32.9)
2	5,072 (28.1)	7,448 (20.8)	8825 (22.0)	17,567 (22.0)
Missing	41 (0.2)	66 (0.2)	51 (0.1)	101 (0.1)
Adequacy of Prenatal Care				
Adequate	13,663 (75.8)	27,977 (78.1)	30,646 (76.5)	61,051 (76.6)
Intermediate	3,537 (19.6)	6,404 (17.9)	7,815 (19.5)	15,550 (19.5)
Inadequate	558 (3.1)	1,040 (2.9)	1,208 (3.0)	2,408 (3.0)
Unknown	224 (1.2)	310 (0.9)	270 (0.7)	538 (0.7)
None	47 (0.3)	85 (0.2)	103 (0.3)	200 (0.3)
Smoking During Pregnancy				
Yes	2,201 (12.2)	3,104 (8.6)	4,818 (12.0)	9,600 (12.0)
No	15,828 (87.8)	15,828 (91.3)	35,189 (87.9)	70,078 (87.9)
Missing	16(0.1)	16 (13.1)	35 (0.1)	69 (0.1)
Season of Conception				
Winter	3,064 (17.0)	6,074 (16.9)	10,170 (25.4)	20,235 (25.4)
Spring	4,329 (24.0)	8,520 (23.8)	10,740 (26.8)	21,382 (26.8)
Summer	6,027 (33.4)	12,006 (33.5)	10,201 (25.5)	20,334 (25.5)
Fall	4,614 (25.6)	9,216 (25.7)	8,931 (22.3)	17,796 (22.3)
Missing				
Gestational Age				
≥37 weeks	15,415 (85.5)	30,842 (86.1)	36,010 (89.9)	71,977 (90.2)
36-32 weeks	2,050 (11.4)	4,107 (11.5)	3,380 (8.4)	6,760 (8.5)
<32 weeks	564 (3.1)	867 (2.4)	652 (1.6)	1,010 (1.3)
Missing				
Maternal Race/Ethnicity				

NH White	10,729 (59.5)	24,654 (68.8)	24,543 (61.3)	48,923 (61.4)
NH Black	1,829 (10.1)	2,957 (8.3)	3,886 (9.7)	7,688 (9.6)
Hispanic	4,244 (23.5)	5,162 (14.4)	8,759 (21.9)	17,447 (21.9)
Asian/Pacific Islander	710 (3.9)	2,213 (6.2)	1,619 (4.0)	3,229 (4.1)
Other	508 (2.8)	808 (2.3)	1,213 (3.0)	2,416 (3.0)
Missing	9 (0.0)	22 (0.1)	22 (0.1)	44 (0.1)
Maternal Education				
<12th grade	3,209 (17.8)	4,066 (11.4)	7,255 (18.1)	14,469 (18.1)
High school graduation	5,862 (32.5)	10,004 (27.9)	14,085 (35.2)	28,026 (35.1)
Some college	8,931 (49.5)	21,677 (60.5)	18,646 (46.6)	37,143 (46.6)
Missing	24 (0.1)	69 (0.2)	56 (0.1)	109 (0.1)
Maternal Language Preference				
English	15,334 (85.1)	31,488 (87.9)	33,882 (84.6)	67,450 (84.9)
Spanish	1,701 (9.4)	213 (6.0)	3,456 (8.6)	6,904 (8.7)
Portuguese	533 (3.0)	964 (2.7)	1,540 (3.9)	3,074 (3.9)
Other	401 (2.2)	1,126 (3.1)	1,059 (2.6)	2,110 (2.7)
Missing	60 (0.3)	107 (0.3)	105 (0.3)	209 (0.3)
Household Income[b]				
<$20,000	1,367 (7.6)	1,721 (4.8)	2,929 (7.3)	5,828 (7.3)
$20,000-$70,000	10,160 (56.4)	18,607 (51.9)	23,513 (58.7)	46,823 (58.7)
>$70,000	6,499 (36.1)	15,480 (43.2)	13,592 (33.9)	27,080 (34.0)
Missing	3 (0.0)	8 (0.0)	8 (0.0)	16 (0.0)
Delivery Source of Payment				
HMO	8,134 (45.1)	19,649 (54.9)	17,074 (42.6)	34,016 (42.7)
Medicaid/Commonhealth	6,777 (37.6)	9,950 (27.8)	16,157 (40.4)	32,178 (40.4)
Other	3,078 (17.1)	6,150 (17.2)	6,747 (16.9)	13,426 (16.8)
Missing	40 (0.2)	67 (0.2)	64 (0.2)	127 (0.2)
Use Wood for Fuel[b]				
Yes	4113 (22.3)	10227 (28.5)	10294 (25.7)	23562 (29.5)
No	13915 (77.1)	25574 (71.4)	29749 (74.2)	56187 (70.5)
Birth Year				
2001	2,413 (13.4)	4,793 (13.4)	23,991 (20.0)	15,978 (20.0)
2002	1,783 (9.9)	3,554 (9.9)	17,111 (14.3)	11,393 (14.3)
2003	2,189 (12.1)	4,346 (12.1)	20,402 (17.0)	13,580 (17.0)
2004	2,059 (11.4)	4,086 (11.4)	19,538 (16.3)	13,008 (16.3)
2005	2,412 (13.4)	4,804 (13.4)	19,534 (16.3)	13,003 (16.3)
2006	2,663 (14.8)	5,264 (14.7)	19,213 (16.0)	12,785 (16.0)
2007	2,220 (12.3)	4,404 (12.3)	-	-
2008	2,290 (12.7)	4,565 (12.8)	-	-

[a]Percentages may not sum to 100% due to rounding.
[b]Measured at the census block group level.

Table 3.2: Distribution of $PM_{2.5}$[a] Exposure in Massachusetts for Infant Bronchiolitis and Otitis Media Cases and Controls.

$PM_{2.5}$ µg/m^3	Bronchiolitis		Otitis Media	
	Cases	Controls	Cases	Controls
Mean (standard deviation)	9.7 (2.5)	9.6 (2.2)	10.1 (1.6)	10.0 (1.5)
Maximum	19.9	20.1	19.2	17.8
Median	9.9	9.8	10.3	10.2
Interquartile range	2.2	2.2	1.8	1.8

[a]$PM_{2.5}$ average from birth to time of clinical encounter for cases and from birth to age (days) of matched case at time of clinical encounter for controls.

Table 3.3: Odds Ratios (OR) and 95% Confidence Intervals (95% CI) for 2 ug/m^3 Increase in Lifetime Average $PM_{2.5}$ Exposure and Infant Bronchiolitis and Otitis Media.

	Bronchiolitis		Otitis Media	
	N	**OR (95%CI)**	**N**	**OR (95%CI)**
Crude[a]	53,845	1.05 (1.02, 1.07)	119,789	1.08 (1.06, 1.10)
Adjusted[b]	53,492	1.02 (1.00, 1.04)	119,072	0.97 (0.95, 0.99)
Adjusted + wood burning[b,c]	53,492	1.00 (0.98, 1.03)	119,072	0.96 (0.94, 0.98)
Hospitalizations only[b]	19,374	1.09 (1.05, 1.13)	3,976	1.07 (0.96, 1.19)
Born 2004-2009[b,d]	34,767	1.01 (0.99, 1.03)	58,290	0.97 (0.94, 0.99)

[a] Crude models adjusted for matching variables; date of birth (+/- 6 days) and gestational week.
[b]Adjusted for risky pregnancy, maternal age, birthweight, smoking during pregnancy, maternal education, adequacy of prenatal care, parity, income and insurance type;matched on date of birth (+/- 6 days) and gestational week.
[c]Wood burning is a blockgroup level variable obtained from census data.
[d]Analysis limited only to infants born between 2004-2009.

Table 3.4: Odds Ratio (OR) and 95% Confidence Interval (95% CI) for 2 ug/m^3 Increase in Lifetime Average $PM_{2.5}$ Exposure and Infant Bronchiolitis and Otitis Media by Susceptibility Risk Factors.

	Bronchiolitis				Otitis Media		
	OR (95%CI)[a]	**N**	**p-value-interaction**		**OR (95%CI)[a]**	**N**	**p-value-interaction**
Gestational age				**Gestational age**			
<37 weeks	1.05 (0.97, 1.13)	5,989		<37 weeks	0.97 (0.95, 0.99)	8,933	
≥37 weeks	1.02 (1.00, 1.05)	47,667	0.81	≥37 weeks	1.00 (0.90, 1.10)	110,370	0.77
Frequency of clinical encounter				**Frequency of clinical encounter**			
1	1.04 (1.01, 1.07)	42,763		1	0.97 (0.95, 0.99)	117,190	
≥2	1.02 (0.96, 1.08)	7,370	0.29	≥2	0.82 (0.70, 0.96)	2,481	0.08
Income[b]				**Income[b]**			
<$37,188	0.98 (0.92, 1.04)	10,981		<$36,543	1.03 (0.96, 1.11)	22,579	
$37,189-$56,579	0.98 (0.91, 1.05)	12,821		$36,544-$55,125	0.94 (0.87, 1.00)	26,763	
$56,580-$81,740	0.99 (0.91, 1.07)	14,615		$55,126-$78,929	0.97 (0.91, 1.04)	32,101	
>$81,740	1.07 (0.97, 1.18)	15,413	0.09[c]	>$78,929	0.92 (0.85, 0.98)	38,349	0.08[c]
Age at time of clinical encounters				**Age at time of clinical encounters**			
0-6 months	1.04 (1.01, 1.07)	34,454		<1 year	0.98 (0.95, 1.01)	51,143	
6-12 months	1.02 (1.00, 1.04)	19,391	0.05	1-2 years	0.97 (0.94, 1.00)	49,657	
				2-3 years	0.94 (0.90, 0.98)	20,040	0.05[c]

[a] Adjusted for risky pregnancy, maternal age, birthweight, smoking during pregnancy, maternal education, adequacy of prenatal care, parity, income and insurance type; matched on date of birth (+/- 6 days) and gestational week.
[b]Income categorized by quartiles based on control distribution.
[c] p-value based on the Wald Test.

Table 3.5: Odds Ratio (OR) and 95% Confidence Interval (95% CI) Residential Traffic Density and Infant Bronchiolitis and Otitis Media.

	Bronchiolitis		Otitis Media	
Traffic Density[b]	**OR (95% CI)[a]**	**N**	**OR (95% CI)[a]**	**N**
Q1 (least dense)	1.00	8,462	1.00	20,688
Q2	1.10 (1.05, 1.17)	15,494	1.01 (0.98, 1.06)	35,577
Q3	1.24 (1.17, 1.32)	17,084	1.03 (0.99, 1.07)	37,381
Q4 (most dense)	1.23 (1.14, 1.31)	12,803	0.98 (0.93, 1.02)	26,128

[a] Adjusted for risky pregnancy, maternal age, birthweight, smoking during pregnancy, adequacy of prental care, maternal education, parity, income and insurance type; matched on date of birth (+/- 6 days) and gestational week.

[b]Traffic density modeled in quartiles (Q1-Q4) based on annual average daily traffic (AADT); Q1 is 0-12.7, Q2 is 12.8-23.0, Q3 is 23.1-79.0, and Q4 is 79.1-652 AADT. Categorized by quartiles based on control distribution.

Table 3.6: Odds Ratio (OR) and 95% Confidence Interval (95% CI) for 2 ug/m^3 Increase in Lifetime Average $PM_{2.5}$ Exposure and Infant Bronchiolitis and Otitis Media Using Sibling Matched Controls.

	Bronchiolitis		Otitis Media	
	OR (95% CI)	**p-interaction**	**OR (95% CI)**	**p-interaction**
Crude	0.88 (0.86, 1.05)		1.12 (1.07, 1.16)	
Adjusted[a]	1.00 (0.98, 1.03)		0.96 (0.91, 1.00)	
Residentially Stable Pairs[c]	0.96 (0.92, 1.01)		0.96 (0.91, 1.02)	
Maternal Education				
Less than high school	0.99 (0.92, 1.07)		1.01 (0.90, 1.13)	
High school	0.95 (0.88, 1.02)		0.99 (0.91, 1.09)	
More than high school	0.98 (0.93, 1.04)	0.96[b]	0.85 (0.78, 0.92)	0.005[b]
Maternal Language Preference				
English	0.98 (0.93, 1.02)		0.95 (0.90, 1.00)	
Not English	0.97 (0.87, 1.06)	0.65	0.95 (0.85, 1.06)	0.98

[a]Adjusted for season of conception, parity, year of birth, maternal age, gestational age, maternal education, and adequacy of prenatal care; matched by mother.
[b] p-value based on the Wald Test.
[c]Only residentially stable pairs included by comparing birth zip-code.

CONCLUSIONS

This dissertation assessed the effect of traffic-related air pollution through various periods of the early life course using a Massachusetts state birth cohort from 2001-2009. In addition to investigating different temporal windows of exposure, this dissertation also assessed various durations of exposure (acute and chronic). Traffic-related air pollution was assessed using novel satellite based $PM_{2.5}$ predictions in addition to traffic proximity measures including; residential distance to major roadways and traffic density near the home.

Cardiac, neural tube, and oral facial clefts defect risk and in-utero $PM_{2.5}$ exposure were investigated using specific critical windows of exposure. Traffic proximity measures were also assessed. Because $PM_{2.5}$ has an uneven spatial distribution, potentially giving rise to variation in disease occurrences, spatial analysis was used to detect geographic variations of birth defect risk. Additionally, spatial patterns were analyzed to determine the influence of $PM_{2.5}$ exposure on geographic risk and identify areas of increased or decreased risk. Results suggest that in utero $PM_{2.5}$ exposure may be associated with risk of patent foramen ovale, patent ductus arteriosus, and perimembranous ventricular septal defects. Findings also support a possible relationship between ostium secundum atrial septal defects and insufficiency of the aortic valve with traffic proximity measures. Spatial analyses show that there are geographic regions with increased risk of ostium secundum atrial septal defects in Massachusetts, even after accounting for $PM_{2.5}$ exposure. There is limited evidence to suggest that cleft lip with or without palate may have an inverse association with $PM_{2.5}$ exposure and traffic-related air pollution in general. To fully understand the influence of $PM_{2.5}$ on birth defects, larger studies of these specific birth defect groups using valid $PM_{2.5}$ estimates are needed.

Acute $PM_{2.5}$exposure was evaluated using a case crossover study, to determine if short term increases in $PM_{2.5}$ 0,1,4, and 7 days prior to clinical encounter are associated with infant bronchiolitis and OM risk. Additional analysis was conducted to determine risk factors which increase susceptibility to disease risk due to increases in $PM_{2.5}$ exposure. Increased risk of infant bronchiolitis was detected with increased acute $PM_{2.5}$ exposure 1 and 4 days prior to clinical encounter. This suggests that $PM_{2.5}$ exposure may play a role in bronchiolitis susceptibility and severity. Findings indicate that preterm infants are at increased risk of infant bronchiolitis with increasing $PM_{2.5}$ levels. Evidence to support an association between $PM_{2.5}$ exposures and OM diagnosis was not found, except among preterm infants. Such findings indicate that future investigations of the influence of $PM_{2.5}$ exposure on infant health outcomes should examine preterm births closely as they may be most affected.

Chronic exposure to $PM_{2.5}$ and risk of infant bronchiolitis and OM clinical encounter was assessed using two matched nested case control designs. The primary design was matched by date of birth and gestational age to account for temporal trends and the second was sibling matched to control for time invariant variables between siblings. Traffic proximity measures were also assessed. Results do not support a positive association between $PM_{2.5}$ exposure and risk of bronchiolitis or OM, although there is suggestion of increased susceptibility among infants born to mothers with lower socioeconomic status and education. Findings also indicate an association between infant bronchiolitis and traffic proximity measures.

This dissertation provides evidence of an association between traffic-related air pollution and cardiac defects as well as for infant bronchiolitis and OM, especially among preterm infants in a geographic location with relatively low $PM_{2.5}$ exposure. Given that exposure is higher in

other areas of the USA and internationally, traffic-related air pollution exposure is a major public health problem and should be considered in future investigations.

APPENDIX A-Chpater 3: Demographic and Exposure Distributions Among Sibling Cases and Controls

Table A.1: Demographic Characteristics of Infant Bronchiolitis and Otitis Media Cases Diagnosed in Massachusetts from 2001-2009 and Sibling Matched Controls Included in Analyses.

	Bronchiolitis Cases N (%)[a]	Bronchiolitis Sibling Controls N (%)[a]	Otitis Media Cases N (%)[a]	Otitis Media Sibling Controls N (%)[a]
Total	8,349	8,349	12,828	12,828
Infant Sex				
Male	5,002 (59.9)	4,240 (50.8)	7,237 (56.4)	6,407 (50.0)
Female	3,342 (40.0)	4,092 (49.0)	5,591 (43.6)	6,421 (50.1)
Maternal Age				
<20 years	745 (8.9)	881 (10.6)	1,302 (10.1)	1,057 (8.2)
21-24 years	1,849 (22.7)	1,887 (22.6)	3,048 (23.8)	3,075 (24.0)
25-29 years	2,022 (24.2)	2,103 (25.2)	3,189 (24.9)	3,253 (25.4)
30-34 years	2,324 (27.8)	2,270 (27.2)	3,508 (27.4)	3,552 (27.7)
35+ years	1,364 (16.3)	1,208 (14.5)	1,781 (13.9)	1,891 (14.7)
Parity				
0	2,004 (34.6)	3,112 (37.3)	4,566 (35.6)	3,793 (29.6)
1	3,628 (36.8)	2,826 (33.9)	4,987 (38.9)	5,433 (42.4)
2	2,694 (28.4)	2,391 (28.6)	3,251 (25.3)	3,574 (27.9)
missing	23 (0.2)	20 (0.2)	24 (0.2)	28 (0.2)
Adequacy of Prenatal Care				
Adequate	6,382 (75.4)	6,378 (76.4)	9,953 (77.6)	9,948 (77.6)
Intermediate	1,607 (19.3)	1,598 (19.1)	2,403 (18.7)	2,359 (18.4)
Inadequate	232 (2.8)	266 (3.2)	362 (2.8)	363 (2.8)
Unknown	101 (1.2)	81 (1.0)	81 (0.6)	115 (0.9)
None	27 (0.3)	26 (0.3)	29 (0.2)	43 (0.3)
Smoking During Pregnancy				
Yes	984 (11.8)	946 (11.3)	1,421 (11.1)	1,449 (11.2)
No	7,353 (88.1)	7,392 (88.6)	11,397 (88.8)	11,360 (88.6)
missing	12 (0.1)	11 (0.1)	10 (0.1)	19 (0.1)
Season of Conception				
Winter	1,376 (16.5)	2,067 (24.8)	3,244 (25.3)	3,193 (24.9)
Spring	1,914 (22.9)	2,398 (28.7)	3,482 (27.1)	3,445 (26.9)
Summer	2,858 (34.2)	2,160 (25.9)	3,277 (25.6)	3,260 (25.4)
Fall	2,193 (26.3)	1,705 (20.4)	2,821 (22.0)	2,922 (22.8)
missing	8 (0.1)	19 (0.2)	4 (0.0)	8 (0.1)
Gestational Age				

<37 weeks	7,220 (86.5)	7,460 (89.4)	11,658 (90.9)	17,123 (90.3)
≥37-32 weeks	902 (10.9)	710 (8.5)	983 (7.7)	2,266 (7.8)
<32 weeks	214 (2.6)	160 (1.9)	183 (1.4)	620 (1.8)
missing	8 (0.1)	19 (0.2)	4 (0.0)	8 (0.1)
Maternal Race/Ethnicity				
NH White	5,157 (61.8)	5,171 (61.9)	8,294 (64.7)	8,301 (64.7)
NH Black	762 (9.1)	751 (9.0)	1,143 (8.9)	1,137 (8.9)
Hispanic	1,906 (22.8)	1,890 (22.6)	2,593 (20.2)	2,595 (20.2)
Asian/Pacific Islander	328 (3.9)	323 (3.9)	482 (3.8)	479 (3.7)
Other	188 (2.3)	207 (2.5)	308 (2.4)	309 (2.4)
Missing	8 (0.1)	7 (0.1)	8 (0.1)	7 (0.1)
Maternal Education				
<12th grade	1,430 (17.1)	1,441 (17.3)	2,218 (17.3)	2,051 (16.0)
High school graduation	2,567 (30.8)	2,539 (30.4)	4,002 (31.2)	4,132 (32.2)
Some college	4,339 (51.9)	4,358 (52.2)	6,590 (51.4)	6,622 (51.6)
missing	13 (0.2)	11 (0.1)	18 (0.1)	23 (0.2)
Maternal Language Preference				
English	7,326 (87.8)	7,281 (87.2)	11,255 (87.7)	11,320 (88.2)
Spanish	684 (8.2)	703 (8.4)	911 (7.1)	885 (6.9)
Portuguese	146 (1.8)	137 (1.6)	290 (2.3)	269 (2.1)
Other	171 (2.1)	192 (2.3)	338 (2.3)	307 (2.4)
missing	22 (0.3)	36 (0.4)	34 (0.3)	47 (0.4)
Household Income				
<$20,000	669 (8.0)	623 (7.5)	916 (7.1)	939 (7.3)
$20,000-$70,000	4,445 (53.2)	4,500 (53.9)	7,057 (55.0)	7,080 (55.2)
≥$70,000	3,235 (38.8)	3,226 (38.6)	4,853 (37.8)	4,809 (37.5)
missing	0 (0)	0 (0)	2 (0.0)	0 (0)
Delivery Source of Payment				
HMO	3,904 (46.8)	3,926 (47.0)	9,071 (46.8)	6,013 (46.9)
Medicaid/Commonhealth	3,044 (36.5)	3,030 (36.3)	7,467 (37.2)	4,556 (36.3)
Other	1,382 (16.6)	1,375 (16.5)	3,428 (15.7)	2,131 (16.6)
missing	19 (0.2)	18 (0.2)	51 (0.3)	28 (0.2)
Birth Year				
2001	864 (10.4)	1,139 (13.6)	2,578 (20.1)	2,089 (16.3)
2002	757 (9.1)	850 (10.2)	1,955 (15.2)	1,613 (12.6)
2003	1,063 (12.7)	1,092 (13.1)	2,341 (18.3)	2,293 (17.9)
2004	1,047 (12.5)	1,288 (15.4)	2,153 (16.8)	2,252 (17.6)
2005	1,175 (14.1)	1,208 (14.5)	1,980 (15.4)	2,094 (16.3)
2006	1,360 (16.3)	1,049 (12.6)	1,821 (14.2)	2,487 (19.4)
2007	1,014 (12.2)	867 (10.4)		
2008	1,069 (12.8)	856 (10.3)		

[a]Percentages may not sum to 100% due to rounding.

Table A.2: Distribution of $PM_{2.5}$ Exposure in Massachusetts for Infant Bronchiolitis and Otitis Media Sibling Pairs.

$PM_{2.5}$ µg/m^3	Bronchiolitis		Otitis Media	
	Cases	Controls	Cases	Controls
Mean (standard deviation)	9.8 (2.4)	9.8 (2.4)	10.1 (1.6)	10.06 (1.58)
Maximum	19.7	19.1	18.2	16.59
Median	9.9	10.0	10.2	10.21
Interquartile range	2.3	2.2	1.7	1.78

[a]$PM_{2.5}$ average from birth to time of clinical encounter for cases and from birth to age (days) of sibling case at time of clinical encounter for controls.

APPENDIX B-Chapter 3: Bias in Sibling Matched Designs

Siblings as a comparison group in matched case-control studies are free from confounding from all time invariant measured and unmeasured factors that are shared by siblings (1). Such methods are ideal for studies, such as this, that are subject to confounding from factors such as indoor air pollution (including tobacco smoke and wood burning), parental proclivities, nutrition, genetics, and socioeconomic status. Although this design is ideal for unmeasured confounding, there is concern regarding selection bias in environmental epidemiology studies of air pollution when there secular trends in exposure of interest exist (2).

When assessing $PM_{2.5}$ exposure and risk of bronchiolitis and OM in the sibling matched design, birth year and parity strongly influenced risk estimates, indicating the possible presence of secular $PM_{2.5}$ trends in MA. To investigate secular trends in MA, $PM_{2.5}$ predictions were averaged across all grid cells per day from 2001-2009. The plot of the raw data indicates a slight decline in $PM_{2.5}$ from 2001-2009 (Figure B.1a). To understand if there was a significant trend, a simple linear regression model was used to model average $PM_{2.5}$ (dependent variable) and time (explanatory variable). Results further confirmed this decline indicating that as time (measured in days) increases, average $PM_{2.5}$ decreases by 0.001 $\mu g/m^3$ ($p<0.0001$). Using the mgcv package in R, a generalized additive model (GAM) with a cubic spline for time, further confirmed a significant decline in $PM_{2.5}$ ($P<0.0001$) over time in MA (Figure B.1b).

Because of secular trends, older sibling controls theoretically would have higher average $PM_{2.5}$ values compared to the younger sibling case, biasing effect estimates towards the null. To quantify the difference in effect estimates by case and control age difference, results were stratified by age difference of sibling cases and controls (Table B.1). Results provide evidence of effect modification according to if controls were older or younger than the case. Overall, we

found elevated ORs for infant bronchiolitis (OR=1.28, 95% CI: 1.16, 1.41) and otitis media (OR=1.19, 95% CI: 0.68, 2.07) when sibling controls are younger and protective ORs for bronchiolitis (OR= 0.81, 95% CI: 0.74, 0.89) and otitis media (OR= 0.67, 95% CI: 0.44, 1.03) when sibling controls are older than cases. These differences in risk estimate were statistically significant ($p<0.001$).

From 2001-2009, $PM_{2.5}$ levels have been significantly decreasing. Such secular trends should be accounted for when assessing the influence of chronic $PM_{2.5}$ on disease risk, especially when using a sibling matched design. Future studies are needed to confirm the consistent presence of bias when using sibling matched designs in air pollution epidemiology studies and future studies should interpret results from sibling matched designs with caution.

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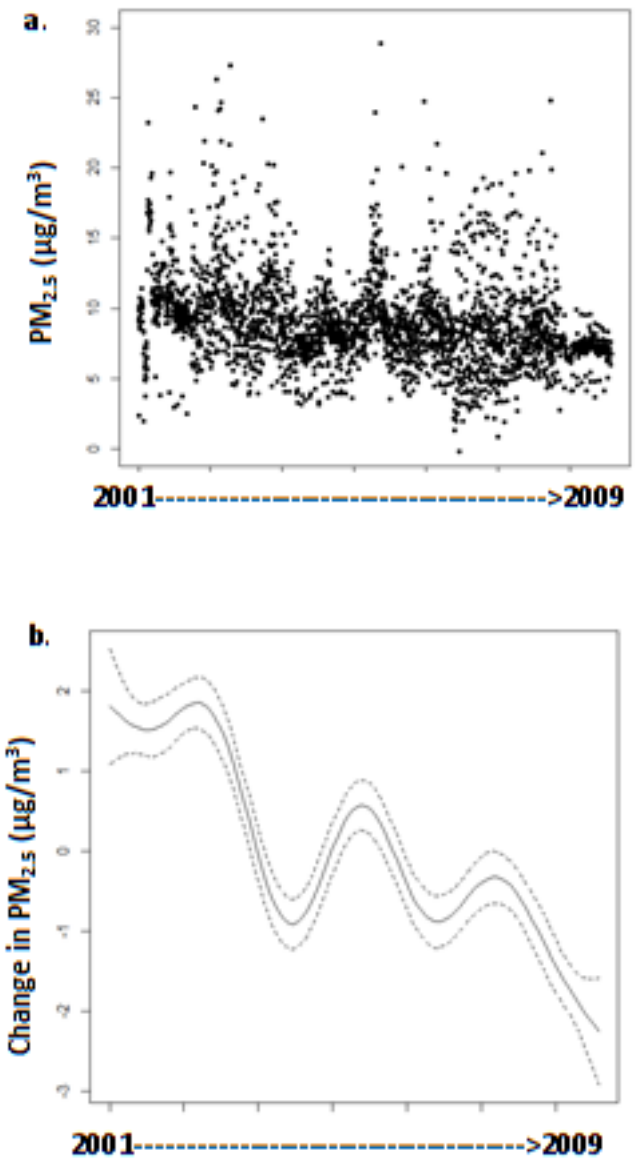

Figure B.1: Secular trends of $PM_{2.5}$ in Massachusetts from 2001-2009.
Scatter plot of average $PM_{2.5}$ in Massachusetts from 2001-2009 (a) and plotted generalized additive model (GAM) of $PM_{2.5}$ with a cubic spline for time (b) demonstrating a decline in $PM_{2.5}$ over the study period.

Table B.1: Odds ratio (OR) and 95% confidence interval (95% CI) for 2 ug/m^3 increase in lifetime average $PM_{2.5}$ exposure and infant bronchiolitis and otitis media by sibling control birth order.

	Bronchiolitis			**Otitis Media**		
	OR (95% CI) [a]	**n**	**p-interaction**	**OR (95% CI)** [b]	**n**	**p-interaction**
Sibling Control Older	0.81 (0.74, 0.89)	11,844		0.67 (0.44, 1.03)	12,806	
Sibling Control Younger	1.28 (1.16, 1.41)	8,160	< 0.0001	1.19 (0.68, 2.07)	12,850	< 0.0001

[a]Adjusted for season of conception, year of birth, maternal age, gestational age, maternal education and adequacy of prenatal care.
[b]Adjusted for season of conception, maternal age, gestational age, maternal education and adequacy of prenatal care.

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APPENDIX C-Chapter 3: Use of Wood for Fuel in Massachusetts

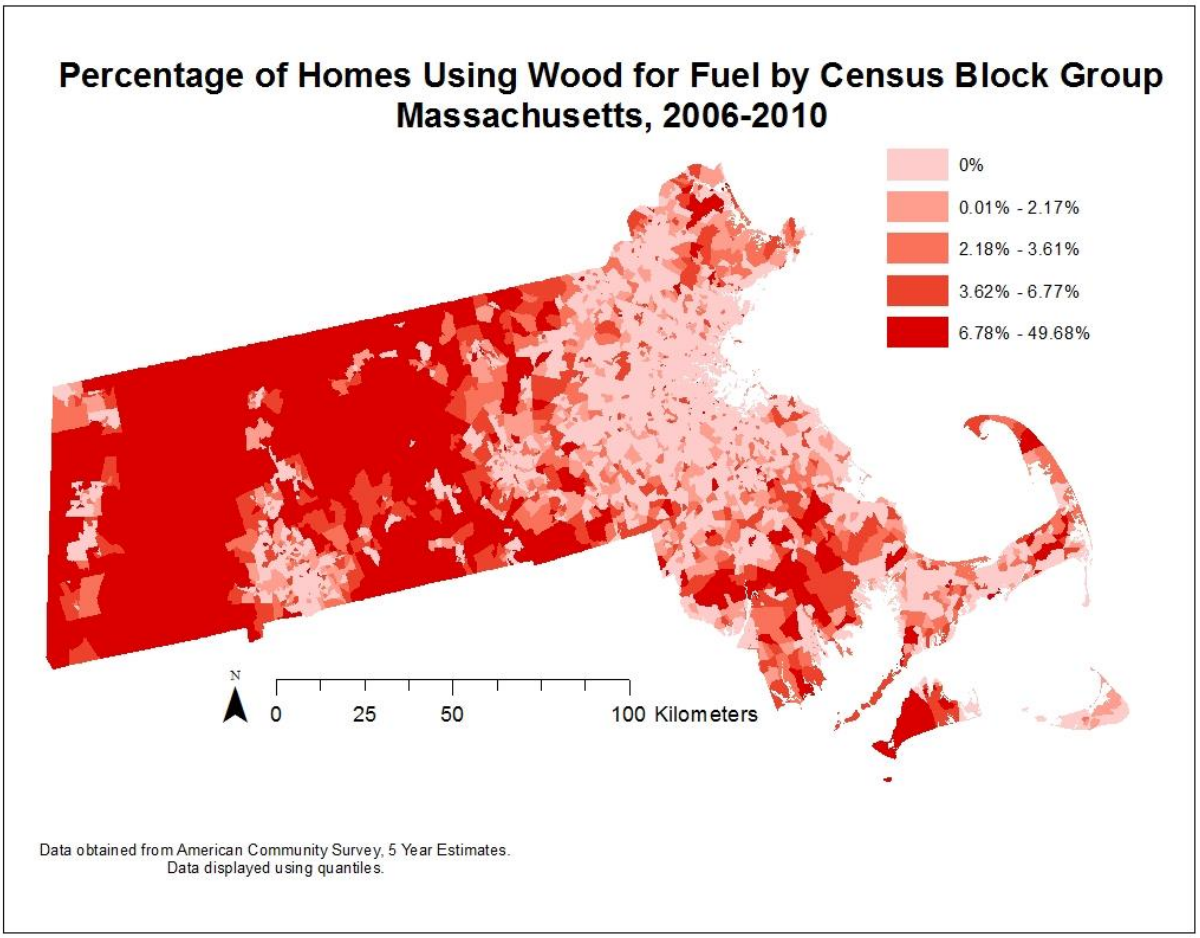

Figure C.1: Spatial Distribution of Wood Used for Fuel as Proportion by Census Block Group in Massachusetts.

Spatial variation of wood used for fuel indicates increased use of wood for fuel in the west and limited use of wood for fuel in the east. Mean for the state is 1.6 (standard deviation is 4.1).