AIR POLLUTION AND PULMONARY TUBERCULOSIS DISEASE IN CALIFORNIA

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Abstract

GENEÉ S. SMITH: air pollution and pulmonary tuberculosis disease in California
(Under the direction of Marilie Gammon)

Mycobacterium tuberculosis is the causative agent of pulmonary tuberculosis (TB) disease, but environmental factors may influence disease susceptibility and or progression from infection to disease. Ecologic analyses, including a preliminary study conducted, suggest a possible link between active TB and ambient air pollution. Animal models demonstrate that air pollution is associated with a reduction in cytokines that normally prevent latent TB infection (LTBI) from progressing into clinical disease. The primary objective of this investigation was to evaluate the association between ambient air pollution (as measured by SO_2, NO_2, CO, O_3, $PM_{2.5}$ and PM_{10}) and incident pulmonary TB (PTB). An additional aim was to determine whether cigarette smoking is a risk factor for PTB disease in this population, and whether smoking influences the association between air pollution and increased risk of developing PTB. To address these aims a case-control study nested among members of Kaiser Permanente Northern California (KPNC) was undertaken. Using electronic clinical databases (ECD), incident cases of PTB diagnosed among adults between the years 1996-2010 were selected, and controls without a history of PTB matched to the cases on age, gender, and race/ethnicity. Cigarette smoking, current and historical residential addresses, and other covariates of interest were abstracted from the ECD. To estimate individual-level pollutant exposure geocoded addresses were linked to inverse distance

weighted surfaces of average monthly pollutant concentrations, produced by the California Air Resources Board. Logistic regression models were used to evaluate the relationship between individual air pollutant and incident PTB, stratifying on history of cigarette smoking. These results are the first from an analytic epidemiologic investigation undertaken to formally evaluate the hypothesis that individual-level estimates of air pollution concentrations are associated with an increased risk of incident PTB. Considering the high air pollution levels and increasing TB rates internationally, their potential association warrants study.

DEDICATION

To my mother, who told me I could when I thought I could not. Thanks for your continued love, support, and prayers.

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LIST OF ABBREVIATIONS

CA	California
CARB	California Air Resources Board
CI	confidence interval
CO	carbon monoxide
COPD	chronic obstructive pulmonary disease
DOTS	Directly Observed Therapy Shortcourse
ECD	electronic clinical database
EPA	Environmental Protection Agency
HIV	human immunodeficiency virus
INF	interferon
IGRA	interferon gamma release assay
IQR	interquartile range
KPNC	Kaiser Permanente Northern California
MDR	multiple drug resistant
NO_2	nitrogen dioxide
O_3	ozone
OR	odds ratio
PM_{10}	particulate matter (particles less than 10μm in aerodynamic diameter)
$PM_{2.5}$	particulate matter (particles less than 2.5μm in aerodynamic diameter)
PTB	pulmonary tuberculosis
SES	socioeconomic status

SO_2	sulfur dioxide
TB	tuberculosis
TNF	tumor necrosis factor
TST	tuberculosis skin test
U.S.	United States
XDR	extensively drug resistant

SPECIFIC AIMS

Understanding cofactors that may increase TB susceptibility is necessary to inform intervention programs aimed at reducing TB transmission. High levels of ambient air pollution are common in many high TB prevalence areas, and have been previously associated with several adverse health outcomes. Air pollution may increase the risk of progression of LTBI to PTB through changes in the adaptive immune system. Utilizing data from Kaiser Permanente Northern California (KPNC), a large and diverse integrated health care delivery system, I evaluated the association between individual estimates of ambient air pollution and incident pulmonary tuberculosis (PTB) disease among California (CA) residents. **Specific Aims** were as follows:

Specific Aim 1. Estimate the magnitude of the association between long-term, 24-month exposure to air pollution (SO_2, NO_2, CO, O_3, PM_{10} and $PM_{2.5}$ levels) and development of incident active PTB.

Hypothesis: Adults are at higher risk of developing PTB if they reside in areas with high air pollution levels, measured by SO_2, NO_2, CO, O_3, PM_{10} and $PM_{2.5}$ levels.

Rationale: In an unpublished ecologic study conducted in North an association was observed between prevalent TB cases and residing in a county with high levels of air pollution assessed using measures of PM_{10} and $PM_{2.5}$ as recorded by United States (U.S.)

Environmental Protection Agency (EPA) monitoring sites. Building upon this finding, a nested case-control study was proposed to determine whether the risk of developing incident PTB is associated with individual-level estimates of air pollution concentrations. Cases and controls were identified from the KPNC membership.

Specific Aim 2. Evaluate whether there is a positive association between cigarette smoking and incident PTB disease, and to test whether smoking is a confounder and/or effect modifier of the association between air pollution levels and TB.

Hypothesis: Smokers will be at a higher risk of developing TB, and thus smokers who reside in areas with high air pollution levels may be at an even higher risk of developing TB than nonsmokers.

Rationale: The few epidemiologic studies which have focused on whether smoking is associated with TB are inconsistent. Those that have found an association, the odds ratio (OR) estimates range from 0.80 (95% confidence intervals (CI): 0.34-1.89) to 4.62 (95%CI: 2.44-8.73) for smokers as compared with non-smokers (Lin 2007). Because smoking is a potential confounder or effect modifier of the primary aim, this association was evaluated among the same Kaiser TB patients and control population. To assess smoking status, data was abstracted from the electronic clinical data maintained by KPNC.

To address these specific aims, the proposed study utilized a case-control design nested within the KPNC population. Cases were KPNC members with a first time diagnosis of PTB disease between 1996 and 2010. Controls, also members of KPNC, were matched 2:1

by age, gender, and race/ethnicity. Primary exposures of interest included monthly averages of ambient air pollution as measured by local monitors. Pollutant concentrations at monitors located closest to residential address was utilized to estimate individual-level exposure. Logistic regression models were used to derive the effect estimate for the association between average 24-month air pollution estimates and risk of developing incident PTB. This study was the first to formally evaluate the proposed primary aim in a population-based epidemiologic study.

CHAPTER 1

BACKGROUND

Epidemiology of Tuberculosis

Tuberculosis (TB) dates back to ancient history, confirmed with evidence of TB found in the remains Egyptian mummies from as far back as 4000BCE (Rothschild 2001, Zink 2003). In the 1600s, one out of every four deaths in Europe was due to TB. It was not until that 1882 Robert Koch identified *Mycobacterium tuberculosis* as the bacillus responsible for TB.

Improvements in living conditions, the introduction of effective public health preventive measures and the development of effective antibiotics, helped to bring tuberculosis somewhat under control during the second half of the 20th century (Tremblay 2007). However, the surfacing of HIV/AIDS and increase in drug-resistant TB, led to a resurge of cases in many areas (Paolo 2004). As a result, the World Health Organization (WHO) declared TB a global health emergency and implemented Directly Observed Treatment Short course (DOTS). Over 183 countries currently use the DOTS strategy in efforts to eliminate TB across the globe.

Today the first line of antibiotic treatments for TB include isoniazid, rifampicin, ethambutol, and pyrazinamide. Second-line drugs used in TB treatment are often less

effective than first-line drugs and cause toxic side effects; these include include streptomycin, cycloserine, capreomycin, ethionamide, and levofloxacin among others. Treatment generally lasts approximately 6 months and is highly effective in abating TB. Although public health has come a long way to control TB, we are still a long way from elimination. Rates have dramatically declined in the US throughout the years, but TB has remained a significant public health problem in the less developed countries (WHO 2010).

It is estimated that approximately one-third of the world's population is currently infected with *Mycobacterium tuberculosis*; 10 percent of which will develop tuberculosis disease during their lifetime. According to the World Health Organization between 2002 and 2020, roughly one billion people will develop tuberculosis infection, over 150 million will become ill, and 36 million will die from the disease (WHO 2010).

Most of the infected will have no symptoms, but those who become ill from the disease often experience prolonged debilitating symptoms including coughing, sputum production, chest pain, loss of appetite, weight loss, fatigue, night sweats, chills and fever (NIEHS 1998). Initial symptoms are generally mild, often being mistaken for the flu or a lingering cold, but do not subside. Symptoms worsen with progression of the disease, resulting in severe coughing and blood in the sputum, an indication of damage to blood vessels in the lungs. Left untreated, 40-60% of active TB cases result in death (Yancey 2008).

TB Infection and Disease

TB is a contagious disease caused by *Mycobacterium tuberculosis* (WHO 2010). The

most common form of the disease is pulmonary tuberculosis, however, tuberculosis can attack almost any part of the body (Kumar 2007).

TB is spread from person to person through the air, usually when infected people cough and sneeze and others inhale infectious droplets circulating in the air. TB cannot be transferred from items touched by an infected person, but in rare cases may be spread through open wounds and from mother to child before and during birth (CDC 2009). Infection occurs when inhaled mycobacteria are engulfed by alveolar macrophages and survive in the phagosomes of these cells. In a person with an intact immune system, the cells of the immune system assemble a wall around the infected macrophages, preventing symptomatic infection. This state is referred to as latent tuberculosis infection. When the body's defenses are not capable or lose the ability to wall off the infection, the host develops active tuberculosis. Persons with active pulmonary tuberculosis are usually symptomatic and capable of passing the infection on to other people (WHO 2010).

TB infection can be present for years, with the individual being asymptomatic. However, in some instances TB infection may progress into disease in a matter of weeks, especially in young children and HIV infected individuals. In either case, the infection becomes active as a result of the immune system's inability to contain the infection. This modification to the immune system occurs when there is a change in health status due to HIV infection, substance abuse, diabetes mellitus, cancer, kidney disease, or a number of other reasons (CDC 1997).

TB Risk Factors

Individuals at higher risk of developing active tuberculosis disease include the elderly, the chronically ill including people with HIV/AIDS, the poor, immigrants, minorities, and healthcare and prison staff.

Many of the elderly were previously exposed to TB years ago and have been able to fight off the infection; however, as they age, many suffer from chronic diseases and malnutrition, which weaken the immune system. In addition, the elderly often live in long term care facilities where they are at greater risk for exposure to others with TB. It is often difficult to recognize their TB symptoms as weight loss and fatigue, are common in the elderly.

Individuals with chronic illnesses, such as cancer, diabetes and kidney disease, often have weakened immune systems and as a result are at greater risk for developing TB. Even those that are malnourished and have extremely low body weight are at increased risk for TB. Persons with HIV/AIDS are at extremely high risk for progressing from latent to active TB. While only 10% of healthy individuals infected with TB will develop the disease over a lifetime, this same percentage of HIV positive individuals infected with TB will develop active TB disease every year if not treated with anti-retrovirals. The fact that some HIV/AIDS infected people are drug addicts or homeless already prone to infection only compounds the problem.

Roughly 6-7% of all people with TB in the United States are homeless and the prevalence of active TB among the homeless is almost twenty times to of the nation as a

whole (Yancey 2008). The homeless frequently live and sleep in overcrowded shelters and many suffer from malnourishment, drug abuse and/or alcoholic addictions and have little or no access to healthcare.

In the US, TB rates are higher among foreign-born persons (**Figure 1.1**). Many immigrants come from countries where TB is endemic and prevention of latent TB is not a priority. In addition, language and communication barriers often cause problems as immigrants as it may hinder immigrants' ability to understand instructions, take medicine correctly, and return for follow up visits.

The large proportion of TB cases arising from blacks, however, cannot be explained by migration (**Figure 1.2**). While TB is known to be more prevalent among minorities, Blacks in the US, carry a very disproportionate burden of the disease. Increased risk for TB is not only a problem for Blacks, but for Hispanics, Native Americans, and Asians as well. Many argue that the increased risk for TB associated with minorities is actually a bi-product of lower SES because minorities are more likely to experience conditions such as malnutrition, homelessness, and limited access to healthcare, all of which are risk factors for TB.

TB can also be an occupational hazard. Prison staffs are generally at high risk of TB infection because many of these facilities lack isolation rooms, have poor ventilation, are often overcrowded and have inadequate medical services. In addition, individuals from various communities reside with one another for varying periods, which can result in susceptible populations having regular contact with persons with existing TB disease (CDC 2009).

TB Prevalence

In 2009 there were almost 9.4 million incident cases and 14 million prevalent cases of TB worldwide, resulting in more than 1.3 million deaths among HIV-negative people and 0.38 million deaths among HIV-positive people attributed to TB (WHO 2010). The largest number of newly detected TB cases occurs in the South-East Asia Region, making up 35% of incident cases globally. The estimated incidence rate in sub-Saharan Africa is more than 350 cases per 100,000. Tuberculosis control has been greatly complicated by the appearance of drug-resistant strains, including those resistant to isoniazid and rifampicin (multiple drug resistant, MDR), those resistant to isoniazid, rifampicin, floroquinolone, and one or more injectable drugs (extensively drug resistant, XDR) and totally drug resistant (TDR). Cases of MDR and XDR tuberculosis are very expensive and difficult to treat and have raised the specter of a strain that is almost completely untreatable. The problem of MDR and XDR tuberculosis is acute in Russia, South Africa, and other countries where resources and modern infection control facilities are limited (Amukoye 2008).

In 2009 there were 11,545 TB cases (resulting in a rate of 3.8 cases per 100,000 people) reported in the US. While the numbers of TB cases reported and the case rate have both steadily decreased throughout the years, TB still has a public health impact on many in the US. California currently contributes, and has for years, the largest number of cases to the nation's TB total. California contributed 21% of the 11,545 reported cases of TB in the United States for 2009. With an incident rate of 6.4 cases per 100,000 population, compared to a national rate of 3.8 cases per 100,000 population, California has the second highest TB rate in the nation (CDC 2009).

Air Pollution and Tuberculosis

Epidemiology of Air Pollution

Air pollution refers to substances in the air capable of causing harm to humans and the environment (**Table 1.1**). This general term is used to describe not just one constituent, but a complex mixture sometimes containing hundreds of components. Some pollutants, like smog, are visible to the human eye, while others, such as carbon monoxide (CO), go undetected. These undesirable elements may be in solid, liquid, or gaseous form and can be natural or man-made. Naturally occurring air pollution can be formed by volcano eruptions, forest fires, dust storms, and even interactions among other pollutants. Air pollution caused by humans, however, is a much larger problem, because this pollution is avoidable and modifiable. Pollutants produced by human activity are numerous in sources including, but not limited to, power plants, manufacturing facilities, marine vessels, aircraft, motor vehicles, controlled burning, chemicals, waste deposition in landfills, furnaces and other fuel-burning heating devices and fumes from paint and aerosol sprays (CARB 2010). Because there are so many causes and sources of air pollution, this mixture is often difficult to elucidate, particularly in outdoor air pollution where many of the coexisting pollutants are also highly correlated (Briggs 1997).

Although, most of the same ambient pollutants are present throughout all cities, states, and countries, they originate from different causes, and exist in varying quantities. This often depends on the geographical location, temperature, wind and other climatic

factors, as well as industrial factors. A prime example of how these factors can affect levels of air pollution can be seen in California, where more than 90% of residents are exposed to unhealthy levels of outdoor air pollution during some portion of the year (CARB 2010). Here, wind patterns and mountain ranges allow air pollution to be trapped into cities, often resulting in smog. These naturally occurring events conducive to the formation of ambient air pollution are also compounded by an abundance of motor vehicle traffic (more cars per household than any other state), major ports, and oil refineries (**Figure 1.3**) (BAAMQD 2010).

While levels of air pollution have continually dropped in the US and other developed countries in recent years, levels in developing countries remain high and are even increasing in many areas. The persistence of ambient air pollution remains a major public health problem as millions die each year from causes directly related to air pollution. There has been an abundance of research studies showing some relationship between ambient air pollution and adverse health effects. In previous research even relatively low concentrations of air pollution have been shown to be associated with adverse health effects (WHO 2011).

A substantial amount of scientific evidence has been amassed that suggests that exposure to ambient air pollution is responsible for a large burden of illness in the US and abroad. Throughout the years, research has shown air pollution to be associated outcomes ranging from cough, exacerbation of asthma and overall declines in lung function to low birth weight, COPD, elevated respiratory and cardiovascular hospitalizations, and even mortality (Holgate 1999). The tremendous burdens air pollution has placed on public health prompt us to delve deeper into understanding the extent of the problem.

People in urban areas of developing countries are exposed to the highest levels of outdoor air pollution in the world. Each year outdoor air pollution imposes an estimated burden of hundreds of thousands of deaths and millions of years of healthy life lost from cardiovascular disease, selected respiratory diseases, and lung cancer (Iwai 2005, Downs 2007, Fullerton 2008). In addition to respiratory effects noted above, ambient air pollution has also previously been linked with pneumococcal disease, caused by bacteria which can lead to pneumonia and meningitis, so this type of association between air pollution and infectious disease is not unprecedented (Kim 1996).

The immune response has long been a sensitive indicator for detecting adverse effects of ambient air pollution (Wang 1989, Wei 2001). Wang *et al*. studied the immune function of children living in various ambient and indoor air pollution exposure levels. The immune function of pupils living in mildly polluted areas but whose family used coal for cooking was inhibited more than was the immune function of pupils residing in heavily polluted areas but whose family used gas for cooking. The immune function of pupils living in mildly polluted areas and using gas for cooking faired the best. While indoor air pollution may play a role in the immune function of children, ambient air pollution could also affect immune processes and should not be neglected (Wang 1989, Chen 2004).

Air Pollution and Tuberculosis: Animal Studies

Combustion-source air pollution is recognized to influence resistance to infection with *Mycobacterium tuberculosis* through effects on airway resistance, and macrophage function in animal models. Hiramatsu and colleagues (2005) investigated effects of the diesel exhaust inhalation on murine mycobacterium infection *in vivo*. In this experiment mice were

exposed to diesel exhaust for 1, 2, or 6 months. Following the last exposure day, exposed and unexposed mice were aerially infected with *Mycobacterium tuberculosis* and seven weeks post infection, the lung tissues were examined. After 6 months of diesel exhaust exposure, the expression levels of interleukin-10, interferon-gamma, and inducible NO synthase mRNAs were decreased, and the mycobacterial load in the mice increased approximately four-fold (Hiramatsu 2005).

Air pollution exposures in murine models were shown to reduce tumor necrosis factor (TNF)-α and interferon-gamma (IFN-γ) production (Saito 2002b). This is noteworthy because TNF-α and IFN-γ play a central role in containing and inhibiting the growth of mycobacterium (Flad 1995, Döffinger 2004, Fremond 2005). These findings suggest that ambient air pollution affects immune processes that can affect TB. Further, the inhibition of TNF-α by drugs in clinical trial has been linked to TB reactivation, implying that the association in mice may be applicable to humans (Jacobs 2007).

Air Pollution and Tuberculosis: Human Studies

In the last twenty years, rates of tuberculosis have been on the rise, mostly in parts of the developing world. On the observation that industrialization coincides with TB, historical statistics of TB and energy were compared dating back to the 1940s (Tremblay 2007). Investigators found that coal consumption and TB rates appear to follow the same trends in the U.S., Canada, and China. However, an explanation for the link between industrialization and rates of tuberculosis remains to be discovered. A paper by Tremblay hypothesizes that pollution caused by the combustion of coal during industrialization has provoked previous

epidemics in the West and may contribute to the continual burden of TB in developing parts of the world.

A different study explored the seasonal fluctuations of TB incidence, and found that along with various climatic factors, atmospheric pollutants (including NO, CO, and SO_2) affected TB incidence (Shilova 2004). These results are suggestive, but to date **there are no published epidemiological studies that have specifically evaluated the association between estimated individual-level outdoor air pollution concentrations and incident PTB.** If air pollution exposure increases the risk of infection, illness, or death from tuberculosis, then the attributable burden of disease would be even greater. Given that tuberculosis infection is endemic in many developing countries, even a small increase in the risk for tuberculosis disease could translate into a large attributable burden (Cohen 2007). Thus, research on outdoor air pollution and tuberculosis is important.

Air pollution from outdoor sources, including motor vehicles, industry, and solid waste burning, is associated with increased morbidity and mortality from respiratory infections in children and adults (Lin 2007). In recent years, air pollution has been identified as a possible risk factor for tuberculosis. Mishra et al. linked biomass cooking fuels and tuberculosis based on a 1992–1993 survey of 90,000 households in India. This study found that women in households using biomass cooking fuels are three times more likely to report tuberculosis than those in households using cleaner fuels (Mishra 1999). Miners in coal and gold mines are also prone to tuberculosis, although it is considered, at least in part, an effect of silicosis (Rockette 1977).

Several epidemiologic studies have suggested a connection between outdoor air

pollution and tuberculosis. In Shenyang, China an ecological analysis was used to study chronic effects of air pollution on mortality in 1992. The analysis showed a significant association between daily mortality and daily ambient levels of total suspended particulates (TSP) and SO_2. Considerable increases in these pollutants led to significant increases in mortality from COPD (chronic obstructive pulmonary disease), cerebrovascular disease, cardiovascular disease, cancer (all sites combined), and tuberculosis disease, as well as total mortality (Xu 1995).

The proposed nested case-control study will be the first epidemiologic investigation to formally evaluate the hypothesis that individual-level estimates of air pollution concentrations will be associated with an increased risk of incident pulmonary TB. My hypothesis is that given the ubiquitous exposure to *Mycobacterium tuberculosis,* the causative agent of PTB, other co-factors are likely to play a role in influencing susceptibility to and progression of TB disease. Experimental data *in vivo* and *in vitro* and ecologic analyses undertaken in human populations support the hypothesis that air pollution, by increasing inflammation and decreasing immune response, will increase the risk of developing pulmonary TB disease **(Figure 1.4**).

Smoking and Tuberculosis

Epidemiology of Smoking

The practice of tobacco smoking has existed for thousands of years; however, with the passage of time, we have determined just how hazardous this activity can be. Many

equate smoking with cancer, but according to the CDC, smoking causes harm to almost every organ in the human body. Cigarette smoking wreaks havoc on the general health of smokers and is commonly the cause of disease, especially cancer, heart disease, and respiratory diseases (CDC 2010).

Although most people think of active smoking when considering potential health effects, passive smoking can also be a health hazard. Passive smoking can be experienced through secondhand smoke (SHS), also known as environmental tobacco smoke (ETS). SHS can result from a smoke remaining following the burning of a cigarette, cigar, or pipe, or from smoke exhaled by individuals in the process of smoking. ETS exposures occur most frequently in homes, workplace, and restaurants. Chemical analyses have shown over 4,000 different compounds present in ETS, many of which are carcinogens (CDPH 2009). Cigarette smoking, including the resulting SHS exposure, is the number one cause of preventable deaths in the US, accounting for 443,000 (or 1 out of every 5) deaths each year (CDC 2010).

Approximately one in five adults (18+ years) in the US smokes cigarettes. The practice of smoking is more common among men than women, 24% vs. 18%, and is often associated with socioeconomic status. The US economy suffers a loss of about $96 billion in health care costs associated with smoking related diseases each year. The prevalence of adult (18+ years) cigarette smoking ranges from 9.3% to 26.5% across the US (CDC 2009). In California, the adult smoking prevalence has dropped from 25.9% percent in 1984 to 11.9% in 2010 (**Figure 1.5**), the second lowest in the nation (CDPH 2010). While rates of smoking are declining in developed nations, tobacco consumption in the developing world is rising at a rate of 3.4% annually. Smoking related diseases will be the cause of one in ten adult deaths

globally and still, approximately 10 million cigarettes are sold worldwide every minute (WHO 2002).

Cigarette Smoking and Tuberculosis: Animal Studies

Cigarette smoking is known to increase the risk of respiratory tract infection by several mechanisms, including inducing inflammation, increasing mucosal perviousness, impairing mucociliary clearance, enhancing pathogen adherence, disrupting the respiratory epithelium, and altering immune function (Benowitz 2010).

Several studies have been conducted that suggest there may be a link between exposure to cigarette smoke and increased susceptibility to TB disease. A study conducted by Chan et al. found an association between the number of cigarettes smoked in the household and the risk of developing active TB among exposed individuals (Chan 2010).

In an animal experiment (Shang 2011), mice were exposed to cigarette smoke for 14 weeks and thereafter exposed to aerosolized *Mycobacterium tuberculosis*. Results showed that mice exposed to cigarette smoke had more mycobacteria isolated from the lungs and spleens after 14 and 30 days than control mice. Cigarette smoke exposed mice also had worse lung lesions, less macrophages producing IL-12 and TNF-α, and fewer INF-γ and TNF-α producing CD4 and CD8 cells.

Shang et al. used 2 different approaches to corroborate these findings. In the first approach, they infected macrophages isolated from the cadaveric lungs of smokers and nonsmokers with TB. The result showed increased mycobactium isolated from the macrophages of the smokers compared with nonsmokers**.** Investigators in this study also

infected macrophages from two healthy nonsmokers with TB alone or with 10% cigarette smoke extract, 1 lg/mL nicotine, or 30 lmol/L acrolein. After only four days of infection, there was significantly more mycobacterium TB bacilli in the cells with cigarette smoke extract, nicotine, or acrolein compared with that in the alveolar macrophages infected with TB alone. Levels of TNF-α were decreased by 42%, 41%, and 28% by the cigarette smoke extract, nicotine, and acrolein, respectively (Shang 2011).

Investigators believe that nicotine in the cigarette smoke prevents macrophages in the lungs from producing TNF-α, and in the absence of this cytokine, reactivation of TB is more likely to occur (Davies 2006). Considering the increasing number of studies pointing to a plausible connection, this hypothesis warrants further examination.

Cigarette Smoking and Tuberculosis: Human Studies

Cigarette smoking is increasing worldwide, particularly in resource-poor countries. The geographic overlap between areas of the world with a high prevalence of cigarette smoking and regions with the most cases of latent and active TB cases is striking. China and India not only parallel in top rankings in the number of smokers, but also rank as the two countries with the largest number of active TB cases (1.4 million and 1.9 million, respectively) (Chan 2010, Hassmiller 2006).

A large case-control study from India found that smoking-attributable deaths from TB were greater than the smoking-attributable deaths from vascular disease or cancer (Benowitz 2010) and a Hong Kong study of over 15,000 female never smokers found that household exposure to SHS increased susceptibility to TB disease (Leung 2004). Lin *et al.* (2007)

conducted a meta-analysis to explore the link between tobacco smoking and the risk of TB attributed infection, disease, and death. Included in the analysis were observational studies reporting effect estimates and 95% confidence intervals on how tobacco smoking, is associated with tuberculosis. Substantial evidence was found that tobacco smoking is positively associated with all tuberculosis outcomes, and that passive smoking and indoor air pollution from biomass fuel combustion are also possibly associated with tuberculosis disease (Lin 2007).

Several studies have found that cigarette smoking to be a risk factor for tuberculosis infection, tuberculosis disease, severe forms of tuberculosis disease, and death from tuberculosis, however, a few have not found this association. The odds ratio (OR) estimates range from 0.80 (95% confidence intervals (CI): 0.34-1.89) to 4.62 (95%CI: 2.44-8.73) for smokers as compared with non-smokers (Gajalakshmi 2003, Hassmiller 2006, den Boon 2007, Lin 2007). Despite current building evidence, CDC does not record smoking in the present surveillance system. This study may offer additional evidence that cigarette smoking is an important factor in the susceptibility to developing tuberculosis disease.

Preliminary Study

For my master's essay, I utilized Poisson regression models to examine the impact of outdoor air pollutants (PM_{10} and $PM_{2.5}$) on rates of pulmonary tuberculosis disease in NC residents for 1993-2007. This ecologic study is the first in the U.S. to provide support for a potential link between air pollution and TB, in a geographic location where both the

prevalence of the exposure and the outcome are lower than have been reported in studies conducted in developing countries.

All physician-reported cases of tuberculosis disease during the study period were considered for this analysis, and annual averages of county level particulate matter (**Table 1.2**) were used to define case exposure to air pollution. During the study period there were 5319 reported cases of pulmonary tuberculosis disease total in NC.

The overall rate of pulmonary tuberculosis disease in North Carolina was 4.41 per 100,000 person-years from 1993 to 2007. **Figure 1.6** illustrates how the rates and frequency of diagnosed tuberculosis disease cases in North Carolina have declined nearly monotonically since 1993.

Table 1.3 shows rate ratios for pulmonary TB in relation to PM_{10} and $PM_{2.5}$ after adjustment for age, gender, race, and year of diagnosis and then after adjustment in a co-pollutant model. Elevated $PM_{2.5}$ tended to have larger incidence rate ratios than PM_{10}, although there was inconsistency across the highest PM categories. Before adjustment for confounding variables, all quintiles of particulate air pollution above the lowest had elevated incidence rate ratios, suggesting that any exposure above the lowest quintile was associated with a higher rate of tuberculosis disease. However, adjustment for confounders weakened the observed associations.

This analysis was hampered by the lack of PM monitors within North Carolina. Official monitoring of $PM_{2.5}$ by the U.S. EPA in the state did not occur until 1999, and as a

result, no data for this pollutant could be analyzed before that year. Additionally, the number of PM_{10} monitors in NC declined by one-third in 1998 and then by half in 2003.

The true effects of air pollution on tuberculosis may be other than estimated here, for numerous reasons. Primarily, individuals are typically exposed to a mixture of air pollutants, whereas these data include information only on PM_{10} and $PM_{2.5}$. The estimated effects may be attenuated to the extent that a mix of pollutants could trigger tuberculosis disease, rather than solely PM_{10} and $PM_{2.5}$. In addition, county level measures, adapted from local monitors, are not an ideal measure of exposure to air pollution. Interpretation also must consider temporal ambiguity complicated by an unknown latency period. It should also be noted that the trends for particulate matter air pollution and reported TB disease during the period both declined similarly throughout the period. While there could be many causes of this correlation, the possibility that TB disease rates could be influenced by pollutants outside of county lines should be considered.

Additionally, data were not available that would allow for the assessment of several important covariates: occupational exposures, HIV status, and behaviors such as tobacco smoking, alcohol consumption, and injection drug use. For example, smoking could affect the relationship between particulate air pollution and tuberculosis disease. TB is a disease of poverty and is often linked with urban living; however, the association of specific socio-economic factors like urbanicity and tuberculosis is not clear. Information is also lacking in this study concerning the length of residence in a particular county. Available data could not account for individuals migrating in and out of a county and varying air pollutant exposures

as a result. This history is important, because the effects of air pollution on the risk of tuberculosis disease could be cumulative over time.

None of the rate ratios was meaningfully below 1.0, consistent with the possibility of a threshold effect. This investigation was a first step in exploring the association between ambient air pollution and TB, and motivating us to further investigate the study hypothesis by improving upon the study design and exposure assessment methods.

Summary

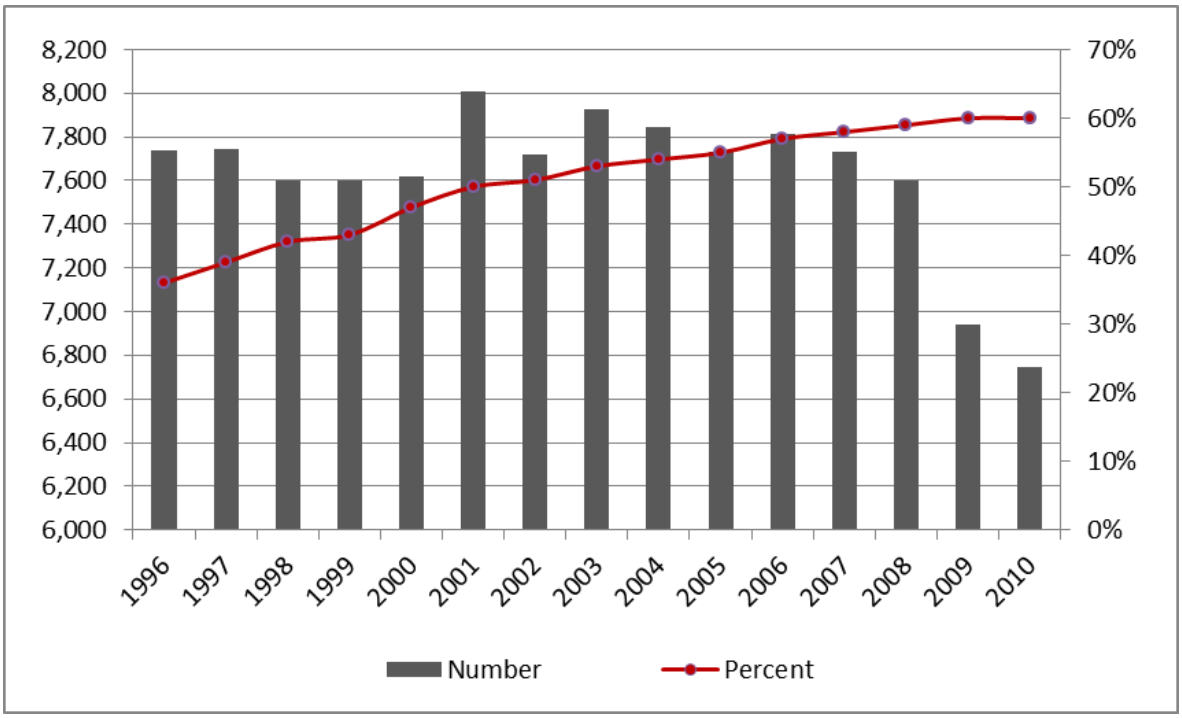
Mycobacterium tuberculosis is the causative agent of TB disease. Laboratory experiments demonstrate that it is biologically plausible that environmental factors may influence progression from TB infection to active disease (Saito 2002a, Saito 2002b, Hiramatsu 2005, Bonay 2006). Experimental results are supported by ecologic analyses in humans. Comprehensive understanding of the disease's risk factors is essential to the control of TB. Although recent TB studies in humans have focused on the influence of HIV, alcohol, and other co-factors (den Boon 2007, Kolappan 2007), none have assessed the impact of personal ambient air pollution concentrations. Because of the high PTB burden, the increasing air pollution levels worldwide, and the biologic plausibility of a possible link between the two, the putative association between modifiable environmental exposures and TB deserves exploration.

Current evidence supports the idea that exposure to ambient air pollution and cigarette smoke both act as TB disease promoters. Therefore, in this dissertation, I will

investigate incident pulmonary tuberculosis disease in relation to levels of ambient air pollution and PTB in relation to cigarette smoking. The research questions in this dissertation are of considerable public health importance as they address an important relation that is missing in existing TB literature.

Chapter 1 Figures and Tables

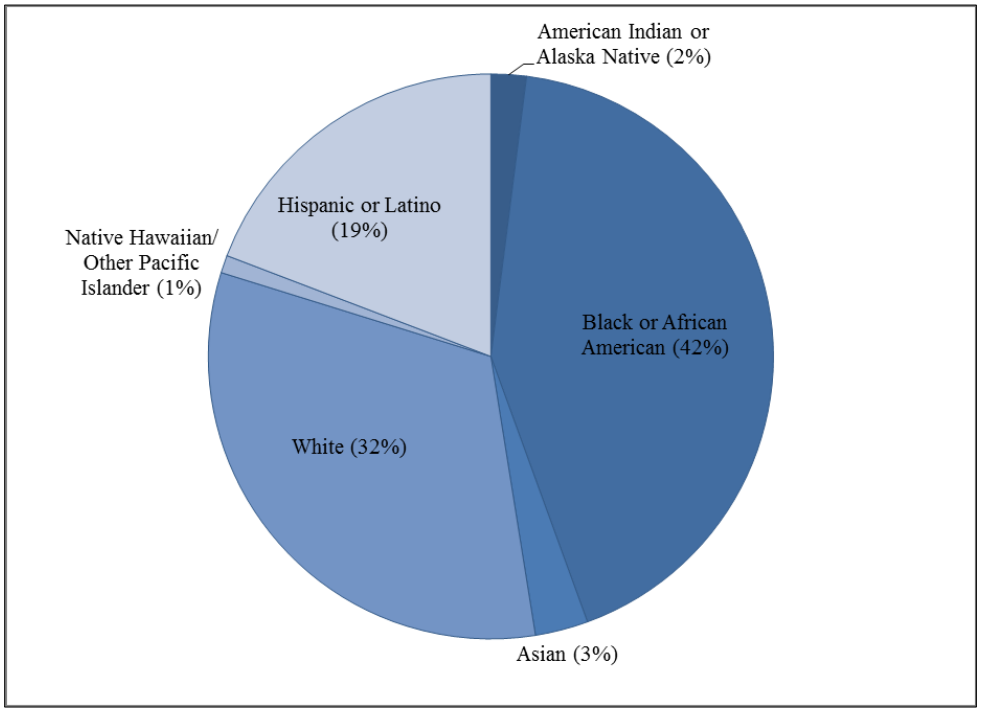
Figure 1.1 Trends in TB Cases in Foreign-born Persons, United States, 1996–2010.



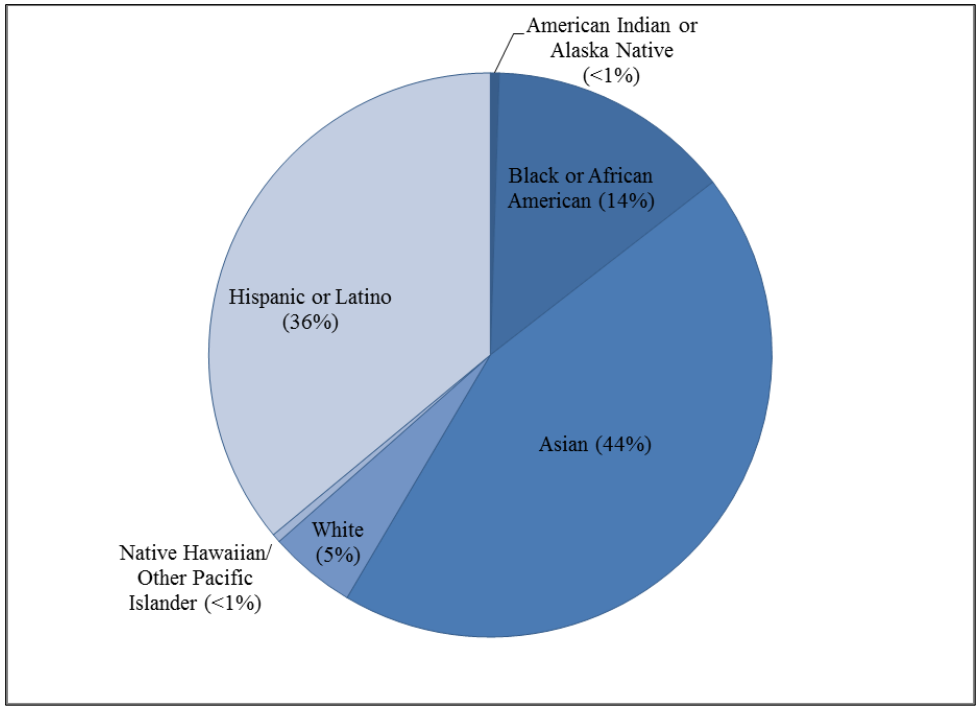
Data Source: Centers for Disease Control and Prevention (http://www.cdc.gov/tb/statistics/reports/2011/table5.htm)

Figure 1.2 Reported TB Cases by Origin and Race/Ethnicity*, United States, 2009.

(a) U.S. born

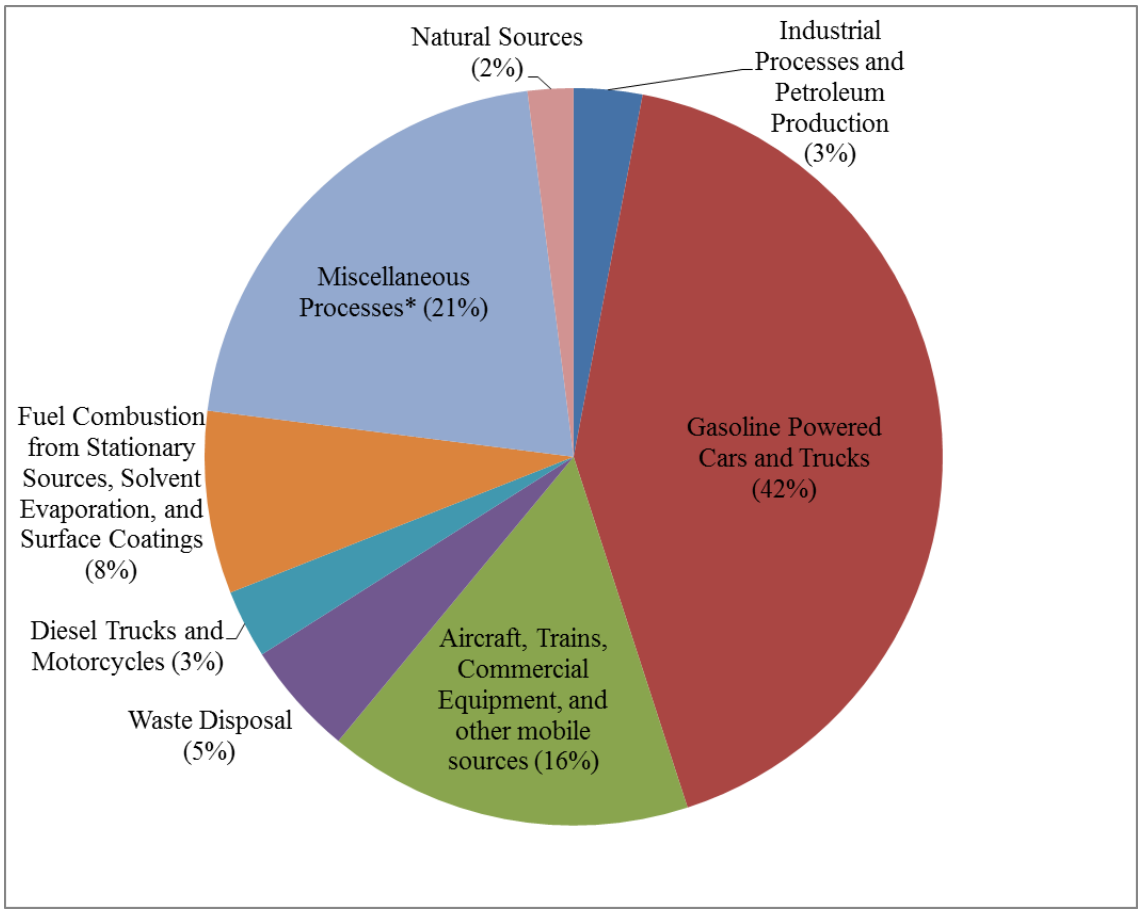


(b) Foreign born



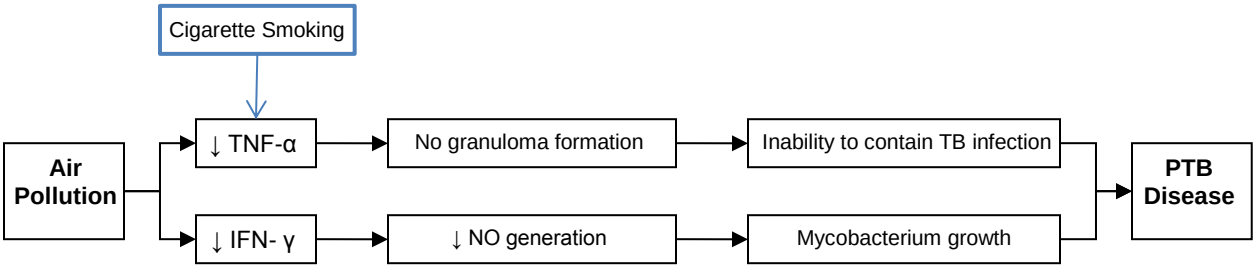
*All races are non-Hispanic. Persons reporting two or more races accounted for less than 1% of all cases.

Figure 1.3 Sources of all air pollutants measured in California



*Residential fuel combustion, farming operations, construction, road dust, wind-blown dust, fires, waste burning, utility equipment and other miscellaneous processes.
Data Source: California Air Resources Board Emissions Inventory report
(**http://www.bar.ca.gov/80_BARResources/02_SmogCheck/Air_Pollution_Sources.htm)l**

Figure 1.4 Conceptual Diagram for the association of air pollution, smoking, and pulmonary tuberculosis (PTB).



Adapted from: Tremblay (2007).

Table 1.1 Health effects of common air pollutants.

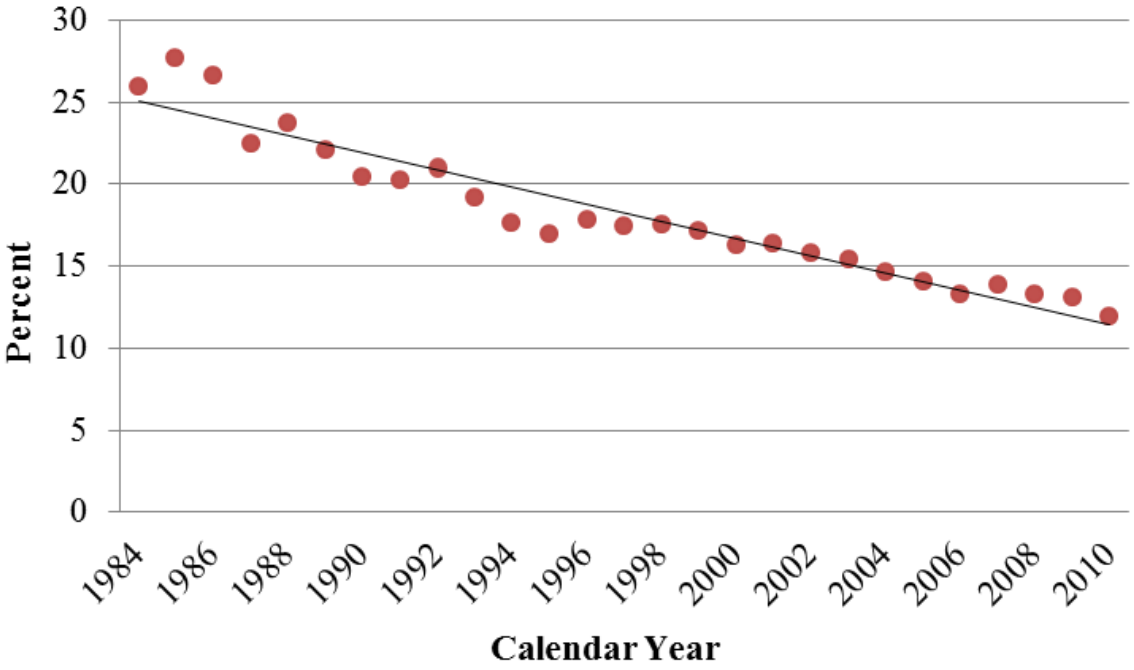
POLLUTANT	HEALTH EFFECTS	EXAMPLES OF SOURCES
Particulate Matter ($PM_{2.5}$ and PM_{10})	Hospitalizations for worsened heart diseases Emergency room visits for asthma Premature death	Cars and trucks (especially diesels) Fireplaces, woodstoves Windblown dust from roadways, agriculture and construction
Ozone (O_3)	Cough, chest tightness Difficulty taking a deep breath Worsened asthma symptoms Lung inflammation	Precursor sources*: motor vehicles, industrial emissions, and consumer products
Carbon Monoxide (CO)	Chest pain in heart patients** Headaches, nausea** Reduced mental alertness** Death at very high levels**	Any source that burns fuel such as cars, trucks, construction and farming equipment, and residential heaters and stoves
Sulfur Dioxide (SO_2)	Increases lung disease and breathing problems for asthmatics. Reacts in the atmosphere to form acid rain.	Coal or Oil Burning Power Plants and Industries, Refineries, Diesel Engines
Nitrogen Dioxide (NO_2)	Increased response to allergens	Any source that burns fuel such as cars, trucks, construction and farming equipment, and residential heaters and stoves

Data Source: http://www.arb.ca.gov/research/health/fs/fs1/fs1.htm

*Ozone is not generated directly by these sources. Rather, chemicals emitted by these precursor sources react with sunlight to form ozone in the atmosphere.

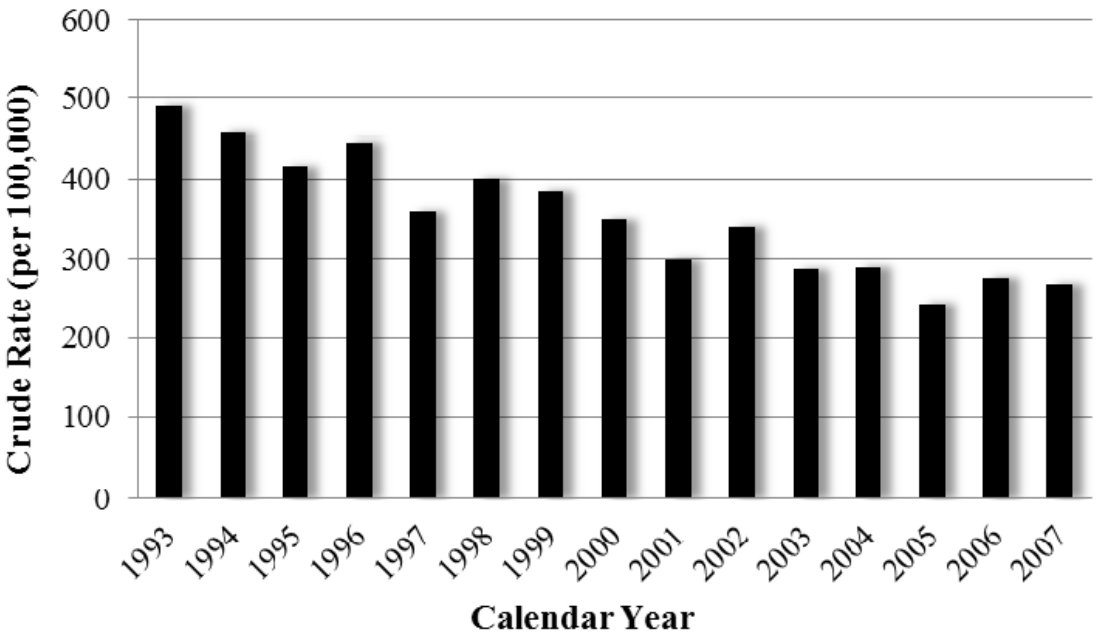
**Health effects from CO exposures occur at levels considerably higher than ambient.

Figure 1.5 Smoking prevalence among California adults, 1984-2010



Data Source: http://www.cdph.ca.gov/Pages/NR11-031SmokingChart.aspx

Figure 1.6 Overall annual rate of pulmonary TB in NC, 1993-2007



Data Source: NC Department of Health and Human Services

Table 1.2 Mean ± standard deviation of particulate matter air pollution concentrations in North Carolina, 1993-2007.

PM_{10} ($\mu g/m^3$)				
Year	**Mean**	**SD**	**Min**	**Max**
1993	24.63	2.91	17.75	29.90
1994	22.44	2.98	16.63	29.67
1995	22.41	3.46	16.46	33.81
1996	23.25	3.29	16.26	32.72
1997	22.38	2.90	16.06	28.49
1998	23.74	2.47	19.85	28.04
1999	22.53	2.96	16.29	27.81
2000	22.29	3.49	16.72	29.81
2001	20.94	2.67	15.53	26.55
2002	19.82	2.38	15.53	23.90
2003	19.31	2.13	14.21	22.28
2004	18.98	3.03	14.85	24.68
2005	18.91	2.89	12.82	23.90
2006	19.51	2.71	15.94	26.19
2007	19.39	3.83	9.60	26.82
$PM_{2.5}$ ($\mu g/m^3$)				
Year	**Mean**	**SD**	**Min**	**Max**
1999	15.30	1.47	11.93	17.76
2000	14.84	1.64	11.95	17.72
2001	13.56	1.40	9.76	16.39
2002	13.17	1.25	10.16	15.74
2003	12.43	1.56	8.74	15.26
2004	12.94	1.32	10.37	15.34
2005	13.34	1.33	10.32	15.73
2006	12.77	1.38	9.73	15.24
2007	12.38	1.43	8.90	14.68

SD= standard deviation
Data Source: U.S. EPA's Aerometric Information Retrieval System (AIRS)

Table 1.3 Comparison of crude and adjusted incidence rate ratios of pulmonary TB for ambient PM exposure in North Carolina.

		Crude IRR (95%CI)		Adjusted* IRR (95%CI)	
PM_{10}[†]	<19.05	Ref		Ref	
(μg/m^3)	19.06-21.08	1.20	(1.07, 1.34)	1.21	(0.97, 1.51)
	21.09-22.78	1.09	(0.97, 1.23)	0.96	(0.76, 1.20)
	22.79-24.92	1.17	(1.05, 1.31)	1.00	(0.79, 1.26)
	≥24.93	1.44	(1.30, 1.60)	1.22	(0.98, 1.52)
$PM_{2.5}$[‡]	<11.94	Ref		Ref	
(μg/m^3)	11.95-12.91	1.38	(1.15, 1.66)	1.29	(0.97, 1.71)
	12.92-13.75	1.28	(1.09, 1.50)	1.27	(1.00, 1.62)
	13.76-14.68	1.13	(0.96, 1.32)	1.09	(0.86, 1.39)
	≥14.68	1.34	(1.14, 1.56)	1.06	(0.80, 1.40)

Data Source: Smith 2013

[†] Analysis includes years 1993 - 2007

[‡] Analysis includes years 1999 - 2007

*Adjusted for age, gender, race, and year of report

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CHAPTER 2

RESEARCH METHODS

Study Design

The primary aim of the proposed large, population-based case-control study, nested within the Kaiser Permanente Northern California (KPNC) cohort membership, was to evaluate the hypothesis that individual-level estimates of concentrations of the six criteria air pollutants, SO_2, NO_2, CO, O_3, PM_{10} and $PM_{2.5}$, are associated with incident pulmonary TB. A secondary aim was to examine the association between active smoking and TB within the same study population.

A nested case-control study approach utilizing existing data from Northern California (CA) was utilized to address the study aims. The study population, drawn from the existing KNPC membership cohort, and includes 2380 incident cases of pulmonary TB (PTB) and 4738 matched controls without TB. Exposure assessment is based on individual air pollution concentrations, which are estimates derived using spatial and temporal analyses. Identification of the study population, electronic abstraction of the corresponding medical record data for key covariates, and individual estimates of air pollution concentrations are being conducted by Dr. Stephen Van Den Eeden and colleagues at KNPC, utilizing the protocol funded by American Lung Association (PI: Genee Smith); a de-identified data set

was made available for the proposed dissertation. Logistic regression was used to examine both the relationship between individual-level pollutant concentrations and PTB risk (aim 1), as well as, the association between ever smoking, as recorded in the electronic clinical data, and risk of PTB (aim 2).

Northern CA was an excellent geographic location in which to conduct this study. TB rates in CA are among the highest in the nation. The KNPC has a large electronic clinical database (ECD), which provides comprehensive medical and demographic data with which to identify and characterize a large number of TB cases and matched controls. Extensive air pollution data were available with considerable exposure spatial variability, based on California Air Resources Board (CARB) data of mean pollutant values and/or days exceeding standards (**Table 2.1**). The proposed investigation was the first to yield results from an observational analytic epidemiologic study on whether TB is associated with individual-level air pollution concentrations.

Study Population

All eligible participants were drawn from KPNC, which provides integrated comprehensive care to approximately 3.3 million people, about 25% of the total population in the geographic areas served (**Figure 2.1**). KPNC members closely approximate the general population ethnically, racially, and socioeconomically; however, they are somewhat more educated and under-represent income extremes (**Figure 2.2**). Patient information on TB and other factors is electronically available from the KPNC ECD, which houses linkages to

multiple databases and disease registries which are constantly updated and frequently validated via chart review.

Subject Eligibility Criteria

Based on KPNC data, an average of nearly 300 incident TB cases per year occur within KPNC. This study included adult incident cases of PTB newly diagnosed among KPNC members between January 1, 1996 and December 31, 2010, inclusive. Controls were drawn from the KPNC cohort and matched to cases (2:1) by age, gender, and race/ethnicity. Patients with a known history of past TB disease were excluded from the initial study population. Over the study period, KPNC documented a total of 4750 cases of incident PTB in their ECD. However, this study was restricted to adults (21 years of age and older) with a required the length of membership enrollment in KPNC study participants of at least two years (to screen out recent immigrants). This resulting population included 2380 cases and 4738 controls for analysis.

For each participant, electronic medical record data were abstracted, including diagnosis date, age, gender, race/ethnicity, length of KPNC membership, smoking status, alcohol related hospitalization (as a proxy for alcohol abuse), history of TB drug use, HIV status, co-morbidity (e.g., diabetes, COPD, renal dialysis), and current/historical residential addresses. The latter was geocoded and linked to available U.S. Census data to obtain contextual indicators of socioeconomic status (SES) not routinely collected at KPNC (e.g., education, income, percent foreign born) at the block-level.

Case Definition

TB was defined as a positive TB culture or a prescription for at least 30 days of four or more anti-tuberculosis medications isoniazid, rifampin, ethambutol, and pyrazinamide (the general regimen for TB treatment). At Kaiser, PTB is ascertained among patients with cough, clinical uncertainty (pneumonia, TB, other), chest x-ray that is on reading "suspicious for TB", and in-house sputum smear microscopy, and when positive, the TB diagnosis is made. ECD were reviewed for each case of TB to ascertain site of disease, and only cases of pulmonary TB were included in the study data. EMR were also used to verify the date of diagnosis (defined as the date of the collection of the specimen for sputum smear microscopy, or, if necessary, the date of the start of treatment).

Exposure Assessment

Air Pollution Exposure

Monthly averages for criteria air pollutants particulate matter with an aerodynamic diameter less than 2.5 μm ($PM_{2.5}$), PM with an aerodynamic diameter less than 10 μm (PM_{10}), sulfur dioxide (SO_2), nitrogen dioxide (NO_2), ozone (O_3), and carbon monoxide (CO) were developed by the California Air Resources Board (CARB) staff from January 1994 to December 2010. Exposure to monthly pollutant averages were chosen over maximum concentrations as they were more likely to be indicative of personal exposure and reflect relevant long term exposure previously explored in animal models. Though lead (Pb) is also a criteria pollutant, this was not included in the analysis as ambient levels have

dramatically decreased since 1973 when lead began to be phased out of motor vehicle gasoline (U.S. EPA); this exclusion is common for epidemiologic investigations on air pollution.

CARB manages the collection of real-time measurements of ambient air pollutants through the California state and local air monitoring network (http://www.arb.ca.gov/aqd/netrpt/netrpt.htm). These fixed-site monitors (**Table 2.2, Figures 2.3-2.8**) were used to estimate levels of $PM_{2.5}$, PM_{10}, SO_2, NO_2, O_3, and CO are part of exposure. Though the CA state and local air monitoring network has $PM_{2.5}$ data, these measurements have only been routinely collected in California since 1999. Because initial $PM_{2.5}$ monitoring sites throughout the study area were limited, monitoring data were supplemented with data from the Interagency Monitoring of Protected Visual Environments (IMPROVE). Analogous to the other pollutant measurements, the $PM_{2.5}$ network measurements from the IMPROVE network were also created with monitors using Federal Reference Methods, however they include continuous and every 6 day monitors. These measurements were all treated the same in calculating monthly averages, however months having less than three CARB calculated monthly average ambient concentrations for all monitoring sites in California from, 1994 to 2010.

Monthly values of ambient concentrations were delivered to KPNC for spatial linkage of study participant addresses. All participants' addresses through 2010 had been previously geocoded as KPNC has database of geocoded addresses for all KPNC members. These estimates were based upon available patient data on the date of diagnosis and previous 24 months, the most etiologically relevant exposure window (Raviglione 1995), including all

new addresses for participants relocating within California from January 1994 through December 2010. If a change in address was not found in the electronic clinical database, this was assumed to have remained the same throughout the entire study period. Under the assumption that pollutant measurements at monitors closest to the subjects' residences have more influence on pollutant concentrations at each residence than those farther away, exposure estimates were assigned based on the monitor closest to the residence. The distance from each case and control residence to the closest air pollution monitor with available data each month was calculated separately for every pollutant (distributions shown in appendix **Table A9**). To account for the relative importance of each monitor assignment, the distance from the closest monitor was used to weight each exposure value.

Cigarette Smoking Exposure

The primary covariate of interest for aim 2, to evaluate whether there is a positive association between cigarette smoking and incident PTB disease, and to test whether smoking is a confounder and/or effect modifier of the association between air pollution levels and TB is smoking. Data on this covariate was obtained by abstracting data in the KPNC electronic clinical databases for study subjects. However, in the Kaiser ECD smoking exposure has been recorded in several different forms with inconsistent definitions over the study period, complicating the derivation a smoking variable. The variable containing the most detailed information on smoking status, which is ideal for addressing aim 2, is a covariate labeled as 'Tobacco', which was most heavily recorded in the EMR in more recent years (2008-2010). Additionally, of all the smoking variables used over the years at Kaiser,

'Tobacco' appears to have the least amount of missing data. **Table A1** shows the amount of missing data by year of diagnosis for the 'Tobacco' variable.

The proportion of smokers among the 2008-2010 cohort is nearly identical regardless of the smoking definition used, as expected. However, for the entire cohort (where the earlier cohort had a large amount of missing data on smoking for any one variable including "Tobacco"), the newly derived variable reduces the amount of missing data substantially (**Table A2**). Although the prevalence of non-smokers appears to be less among those diagnosed with PTB prior to 2008, it is consistent with the smoking prevalence data published by the California state health department (http://www.cdph.ca.gov/Pages/NR11-031.aspx; Accessed June 30, 2012), which shows a steady decline in the proportion of smokers during this time period.

To obtain more complete data on smoking status of the entire cohort of study participants for aim 1, a new smoking variable looking back over the entire study period, 1996-2010, was constructed from all other smoking variables in the EMR. If smoking was indicated anywhere in records, the subject was considered an 'Ever' smoker, otherwise the subject was considered a 'Never' smoker. Only those indicating smoking on the date of diagnosis were considered 'Current' smokers. 'Ever' smokers without any indication of smoking on the date of diagnosis were categorized as 'Past' smokers. Because the most comprehensive and complete smoking data (labeled 'Tobacco') occurred in the clinical databases during the time period between 2008-2010, estimates of smoking dose (cigarettes per day) and duration (years of smoking) were also available for many 'Ever' smokers.

Additional Covariates

Potential confounders and effect measure modifiers for the two study aims were identified through review of the relevant biologic and epidemiologic literature (Yancey 2008). These other covariates, also obtained from KPNC clinical databases, and include age (continuous), gender (male/female), race/ethnicity (categorical), U.S. Census-derived income, education, and neighborhood percentage of foreign birth, length of KPNC membership (continuous), TB drug use (categorical), alcohol hospitalization (yes/no), HIV status (yes/no), and other comorbidities including (diabetes, COPD, renal dialysis, (each yes/no)).

KPNC administrative databases house demographic information for nearly 8 million past and current members. This includes members' names, birthdates, gender, address, etc. Approximately 80% of race/ethnicity data has been captured in the original database. An algorithm was created by Kaiser researchers that assigns race values based on name, location, and other various indicators present in the Kaiser database to individuals missing race/ethnicity. As a result, few observations were missing information on demographic factors.

The immigrant population is generally at a higher risk for PTB, unfortunately, information on immigration status/place of birth was not available in this population. However, mandatory screening confirms that all immigrants should be free of PTB upon entry into the U.S (Liu 2009). In addition, the two-year Kaiser membership requirement for subject eligibility ensures that recent immigrants (those at the highest risk) are screened out of the study. Data on percent of foreign born individuals living within a census block were

used to explore the possibility that an environment/neighborhood with a large foreign born population may affect risk of PTB.

Kaiser also has an existing HIV registry and diabetes registry. These records are very well maintained and kept up to date. Information on these covariates were available for every patient record (i.e. none missing).

Exposure Measurement Concerns

Outdoor Air Pollution

Exposure assessment is frequently the most complex portion of an environmental epidemiologic study. Air pollution exposure in itself has many challenges, which this investigation attempts to overcome. Exposure assessment becomes particularly difficult when assessing chronic exposures, as is the case for this investigation. This study attempted to circumvent common problems of assessing chronic environmental exposures, by examining exposure over time, ranging from 0-24 months. Although the relevant exposure window for influencing the susceptibility to ambient air pollution is unknown, murine models have shown that exposure to air pollution may lead to increased susceptibility for up to six months following the exposure (Hiramatsu 2005). However, whether this same timeline is transferrable to humans remains unknown. It is possible that environmental exposures may result in increased risk on the day of exposure, the following week, or even months after exposure. Polynomial distributed lag models, which assume the effect of exposure on outcome is distributed across time and that there may be immediate as well as delayed effects

(Almon 1965), were initially explored as a method to determine the most relevant time windows of exposure (**Supplement A1**). However, while individual-level exposure estimates varied over time, the ranking of individuals (required due to the non-linearity of pollutant data) did not change substantially over the 0-24 month lag period assessed (see Appendix **Figure A4**). Instead, average exposure during the 24 months prior to diagnosis/entry into the study was analyzed using logistic regression.

Another question that arose in the exposure assessment of ambient air pollution is the ability of a single geocoded residential address, which is linked to a single stationary monitor, to estimate individual level exposure during the proposed etiologic time frame of 24 months. This particular concern may be less of an issue for this study because coverage from Kaiser Permanente is generally offered and kept through employment, so individuals in this study population are more likely to be residentially stable as well. For the proposed study, the residential addresses were updated from each visit. Although the exact date of address change was unknown, updating addresses with each visit is likely to introduce much less measurement error than an assumption of the same residence over several years or decades, which is a common practice in studies of environmental exposures and cancer, for example (Pope 2002, Laden 2006).

Indoor Air Pollution

An additional issue that often comes along with exposure measurement is that most individuals spend a great deal of their time indoors, yet indoor sources of air pollutants are often difficult or impractical to assess. No data on passive smoking was available for this

investigation, but given there has been little research on the effects of passive smoking on TB, it is not certain that this exposure is a concern for my study question.

There was also no information available on fireplace use and other indoor sources of air pollution. However, this particular concern may be less of an issue for this study, given that the indoor pollutants shown to cause increased TB such as burning biomass, and other indoor cooking and heating fuels such as coal (Fullerton 2008, Lin 2007, Mishra 1999) are mostly non-existent in the U.S., particularly in Northern California (US EIA 2009). Moreover, most other epidemiological studies of air pollution, including reproductive and cardiovascular studies, do not have this level of information available for analyses yet still offer valuable findings to the literature. Additional information on indoor air pollution would be desirable, but certainly not necessary to address the study question.

Statistical Analysis

All statistical analyses were performed using SAS version 9.3 (SAS Institute, Cary, NC).

Conditional vs. Unconditional Logistic Regression

In studies where cases and controls are matched on certain factors the problem arises of how to account for non-independence in the matching. The inclusion of dummy coded variables in logistic regression often has problems when the number of degrees of freedom is close to the total degrees of freedom available. In matched case-control studies, conditional logistic regression can be used to circumvent this issue and condition parameters out of the

analysis and each case and matching controls form a stratum. When either case or control has missing information, or is excluded, information is lost for the entire stratum. To prevent the loss of information and provide more precise estimates, unconditional logistic regression, adjusting for all of the matching factors in the analysis, is often used to analyze the matched data. In this study, both conditional and unconditional logistic regression was used in determining associations that air pollution and smoking have on PTB risk.

Aim 1

The use of splines to fit linear trends in the data was considered, however, the numerous pollutants in this analysis may each require a varying number of knots at different locations. Additionally, splines are not always easily interpreted. Because pollutant variables were non-linear, these exposures were grouped into quintiles according to their population distribution, and exposure levels below the 20th percentile were used as the referent category for each pollutant (see Appendix **Figure A5**). When categorizing continuous covariates ensuring homogeneity of variance across levels of exposure is often becomes concern in conducting data analysis. However in logistic regression the variance is always varies with the probability of success.

Conditional logistic regression analysis was used, adjusting for all matching factors (age, gender, and race/ethnicity), to estimate the odds ratios (OR) and 95% confidence intervals (CI) for the association between PTB and each average individual-level air pollutant concentration (SO_2, NO_2, CO, O_3, PM_{10} and $PM_{2.5}$) for the 24 months prior to diagnosis.

Potential effect modification (by cigarette smoking, alcohol hospitalizations, diabetes, COPD, HIV status, renal dialysis, and percent foreign born in census block) was assessed

using likelihood ratio tests (Hosmer 1989) with and without inclusion of multiplicative interaction terms in the logistic regression model ($p<0.10$). Terms considered statistically significant were included in stratified analyses; only cigarette smoking met these criteria and thus only these results are shown.

Confounding was assessed using a greater than 10% change in estimate criterion (Greenland 1989) for several risk factors that could potentially confound the relationship between air pollution and PTB: median household income, percent foreign born in census block, education in residential census block, alcohol hospitalization, diabetes, and HIV status. Using this criterion, no factors were found to confound the PTB-air pollution associations and were therefore not included in the final analysis. Final models only included the matching factors (age, gender, and race/ethnicity).

A multi-pollutant analysis for air pollution and PTB association was also considered using a forward model building approach (see Appendix **Tables A14-A16**). This began with a simple model of two pollutants showing no association with PTB. Additional pollutants were entered in the model, one at a time, in order of no association, positive association, and negative association. At each stage of modeling, effect estimates were inspected for any changes in association.

Aim 2

Splines were used to visualize changes in data and detect whether or not the continuous variables, years of smoking and packs per day, are linearly related to the log-odds of PTB. No consistent patterns indicated in splines, indicating nonlinearity (see Appendix **Figure A3**). Taking into consideration the public health message that will need to be

communicated from study results, it was also important to manually examine the data, as splines may result in cut points that are not easily translated (see Appendix **Tables A4-A5**). I explored several categorical structures for each smoking variable, settling on ≤5, 6-30, and >30 for years of smoking, and ≤½ and >½ for packs per day. These groupings were chosen because they represented the risk profile correctly while using the fewest number of classification levels possible. Because of the completeness of data, all cut-points were created using years 2008-2010.

Odds ratios (ORs) and 95% confidence intervals (CIs) for the association between smoking status and PTB, among cases and controls selected from KPNC between 1996-2010 were calculated using unconditional and conditional logistic regression adjusted for all matching factors (age, gender, and race/ethnicity) (Hansson 2008). Separate models were constructed to estimate effects associated with varying definitions of smoking status (ever vs. never smoking, and current/past vs. never smoking) over the entire 1996-2010 study period (n = 2380 cases and 4738 controls) and restricted to 2008-2010 (n = 734 cases and 1462 controls). In addition, models were constructed to consider dose (cigarettes smoked per day) and duration (years of smoking) as determined from data recorded in the ECD from 2008-2010.

Potential effect modification was assessed using likelihood ratio tests (Hosmer 1989) with and without inclusion of multiplicative interaction terms in the logistic regression model, and those terms with p-values <0.10 were considered statistically significant. When interaction was present, ORs for PTB were calculated stratified by indicators of exposure. Covariates considered as potential effect modifiers include: percent foreign born in residential census block, COPD, alcohol hospitalizations, Crohn's disease, renal dialysis,

immunological prescriptions, diabetes, and HIV status. Potential confounding was assessed using a greater than 10% change in estimate criterion (Greenland 1989) for the following covariates: median household income, percent foreign born in census block, education in residential census block, alcohol hospitalization, diabetes, and HIV status. No covariates assessed were determined to confound the association; thus all final models only included as covariates the matching factors (age, gender, and race/ethnicity).

Study Power

In specific aim 1, I proposed to estimate an association between incident PTB and estimated individual-level air pollutant concentrations. For varying study power, given a sample size of 7118 cases and controls, I estimated the minimum detectable ORs for the association between PTB and air pollutant concentrations, each evaluated as (Quartile 4 vs. Quartile 1) for 2008-2010 and 1996-2010 using nQuery Advisor (version 7). The following assumptions were made: a two-sided test, the case-to-control ratio is 1:2, and a significance level of 0.05. The minimum detectable OR for the air pollution-PTB association with all subjects combined, smokers and non-smokers, is 1.27 at 80% power.

In specific aim 2, I proposed to estimate the OR and 95% CI for the association between incident pulmonary TB and cigarette smoking and test whether smoking is a confounder and/or effect modifier of the association between air pollution levels and TB. Assuming a smoking prevalence of 15% (CDPH 2009), the minimum detectable OR for the cigarette smoking-PTB association is 1.36 at 80% power.

In the Stratified Analysis, where power for the air pollutant-PTB association with smokers and non-smokers is considered separately, the sample size will be reduced depending upon the availability of monitor concentrations and smoking data. Using data from 2006-2010, the minimum detectable ORs (Quartile 4 vs. Quartile 1) for the air pollutant-PTB association at 80% power are 1.91 and 1.29, for smokers and non-smokers, respectively.

In sum, although power will be slightly reduced for different exposure prevalences than what we illustrate here, and when adjustments are made for confounding, study power is expected to be adequate to evaluate the proposed aims (see **Table 2.3**).

Summary

TB is a contagious and often deadly disease that internationally affects millions of people each year. Although it is known that this disease is caused by the bacterial agent *Mycobacteriun tuberculosis*, the reasons for increased susceptibility are not fully understood. I propose that increased concentrations of air pollutants result in a decrease in immune function and ability to ward off TB disease. Further, I hypothesize that adults residing in geographic areas with relatively higher air pollution concentrations are at increased susceptibility of developing PTB disease.

My study objective was to conduct a case-control study nested within the Northern CA Kaiser cohort population to determine whether PTB risk is associated with estimated individual-level air pollution concentrations, considering a range of lagged exposure times

(from zero to 24 months) and controlling for smoking and other potential confounders. I also explored in stratified analyses whether smoking, and other susceptibility factors, modulate the association between air pollution and pulmonary TB. The results from this study are the first to be generated from an analytic epidemiologic, population-based study on this novel hypothesis. Results from this U.S.-based study will be a critical step in helping to determine whether the burden of TB worldwide could be reduced with a reduction in air pollution levels worldwide.

Chapter 2 Figures and Tables

Table 2.1 Mean Annual $PM_{2.5}$ level ($\mu g/m^3$) and No. of days exceeding NAAQS 8-hr Ozone Standard, Northern California Kaiser counties, 2006

County	$PM_{2.5}$	O_3	County	$PM_{2.5}$	O_3
Alameda	11.3	10	San Francisco	10.7	0
Colusa	9.8	1	San Joaquin	15.3	23
Contra Costa	10.4	14	San Mateo	10.9	0
Fresno	17.2	74	Santa Clara	11.6	12
Lake	8.3	0	Santa Cruz	9.9	0
Madera	8.9	15	Sierra	6.5	0
Marin	9.1	0	Solano	14.2	9
Merced	16.7	30	Sonoma	9.6	0
Napa	11.5	0	Stanislaus	16.3	25
Nevada	7.9	63	Tulare	14.8	104
Placer	13.3	62	Yolo	9.3	15
Sacramento	14.2	46			

Figure 2.1 Counties covered by Kaiser Permanente Northern California

Figure 2.2 Comparability of KPNC Members to the General CA population

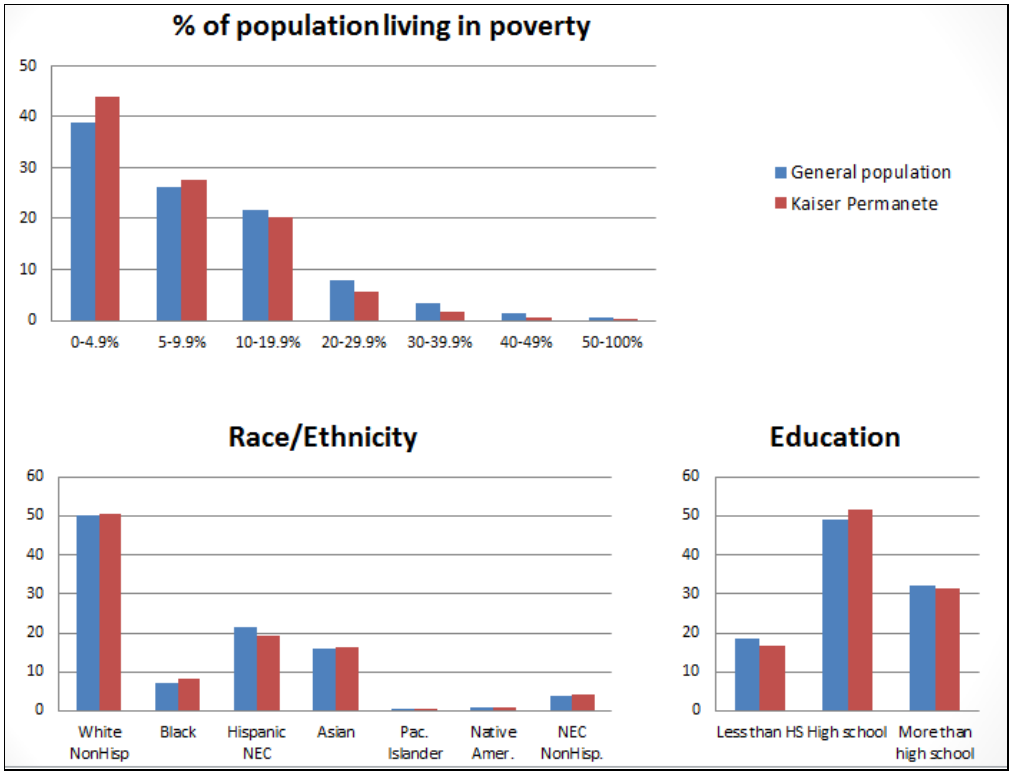

Table 2.2 Air pollution monitors available over the study period

Pollutant	Minumum	Maximum	Mean
CO	78	92	84
NO_2	92	113	104
O_3	149	182	168
$PM_{2.5}$	56	82	77
PM_{10}	135	167	150
SO_2	31	52	38

Data Source: Lipsett 2011

Figure 2.3 Available $PM_{2.5}$ pollutant monitors in California

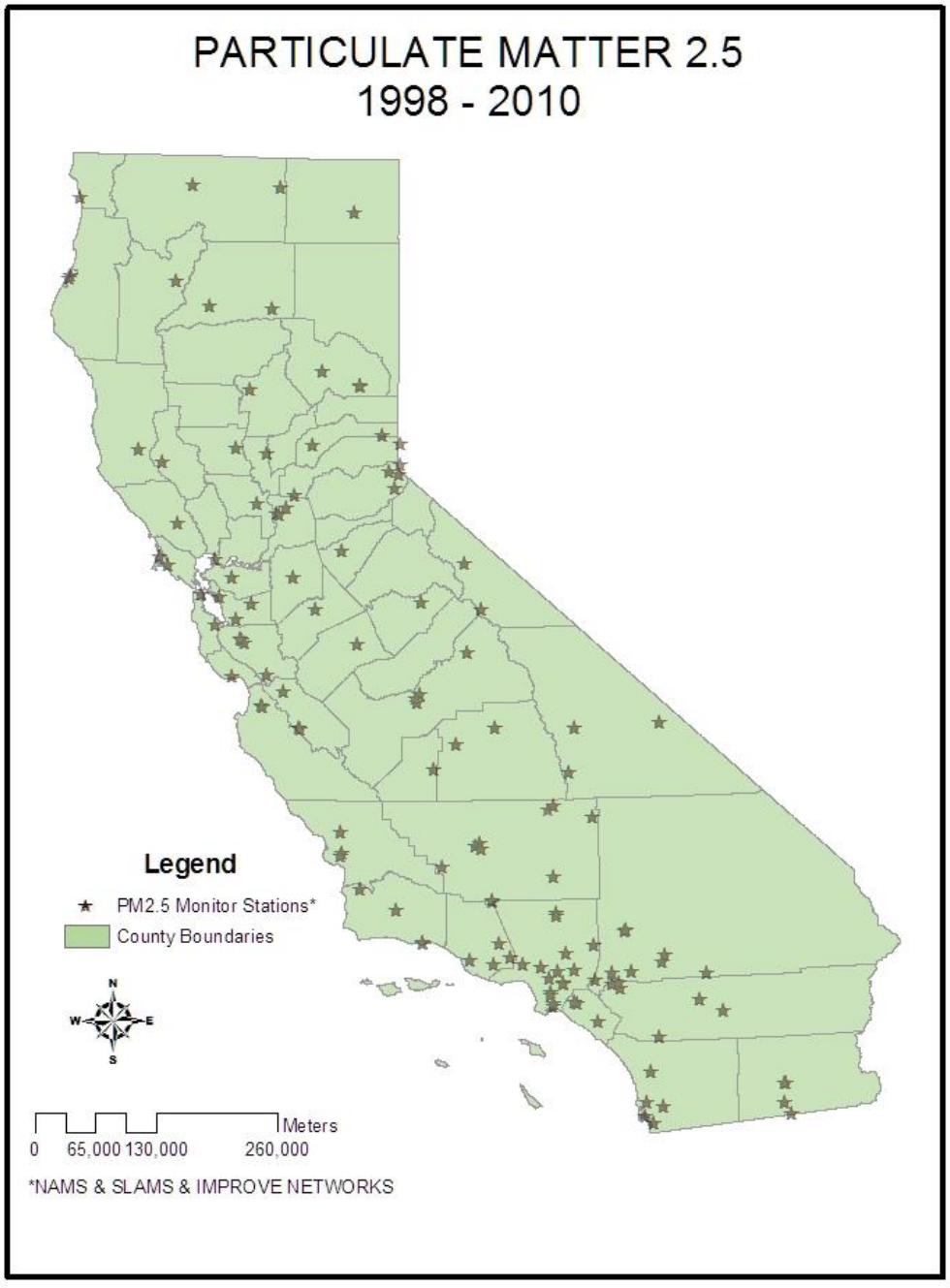

Figure 2.4 Available PM_{10} pollutant monitors in California

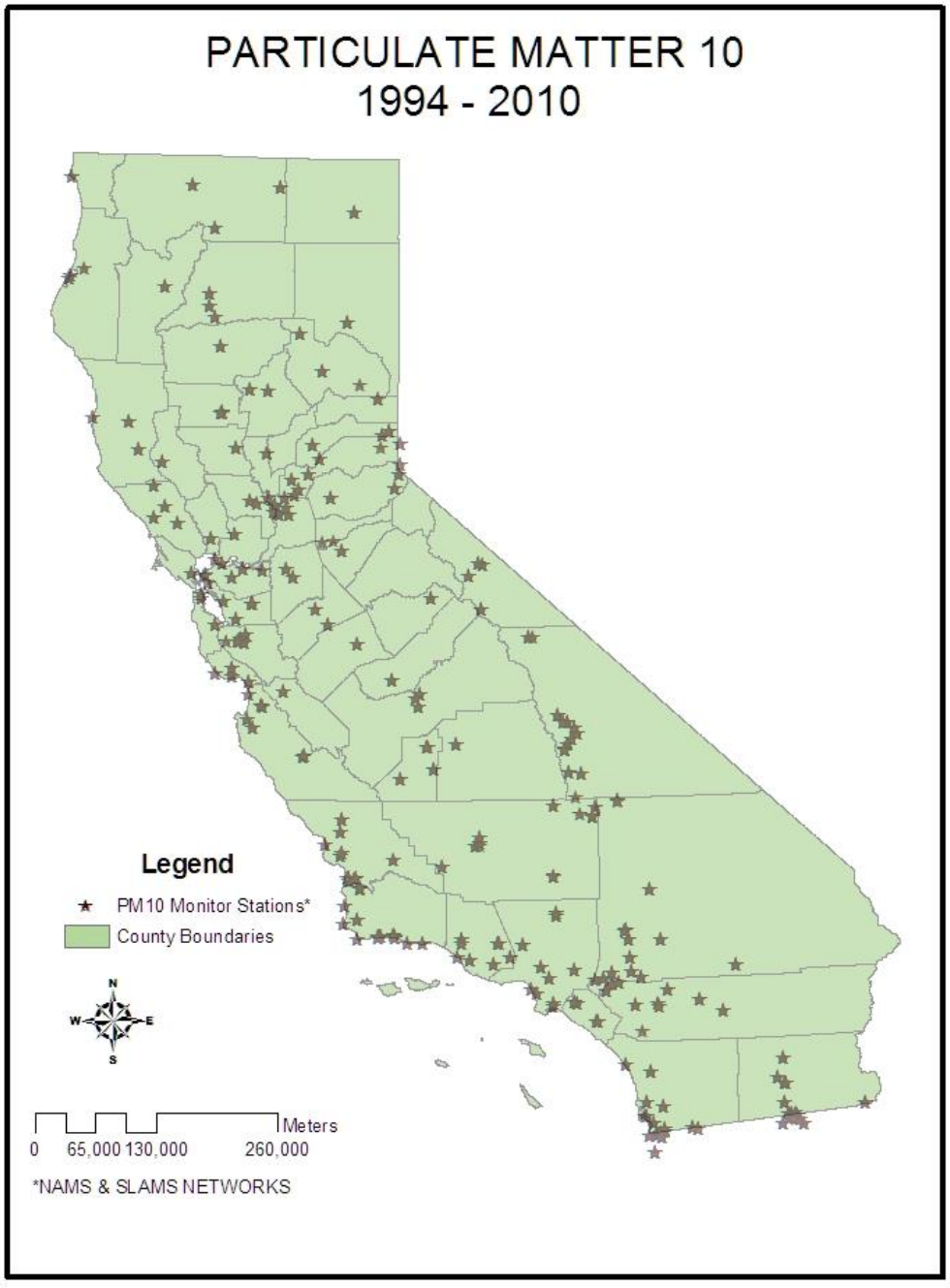

Figure 2.5 Available SO_2 pollutant monitors in California

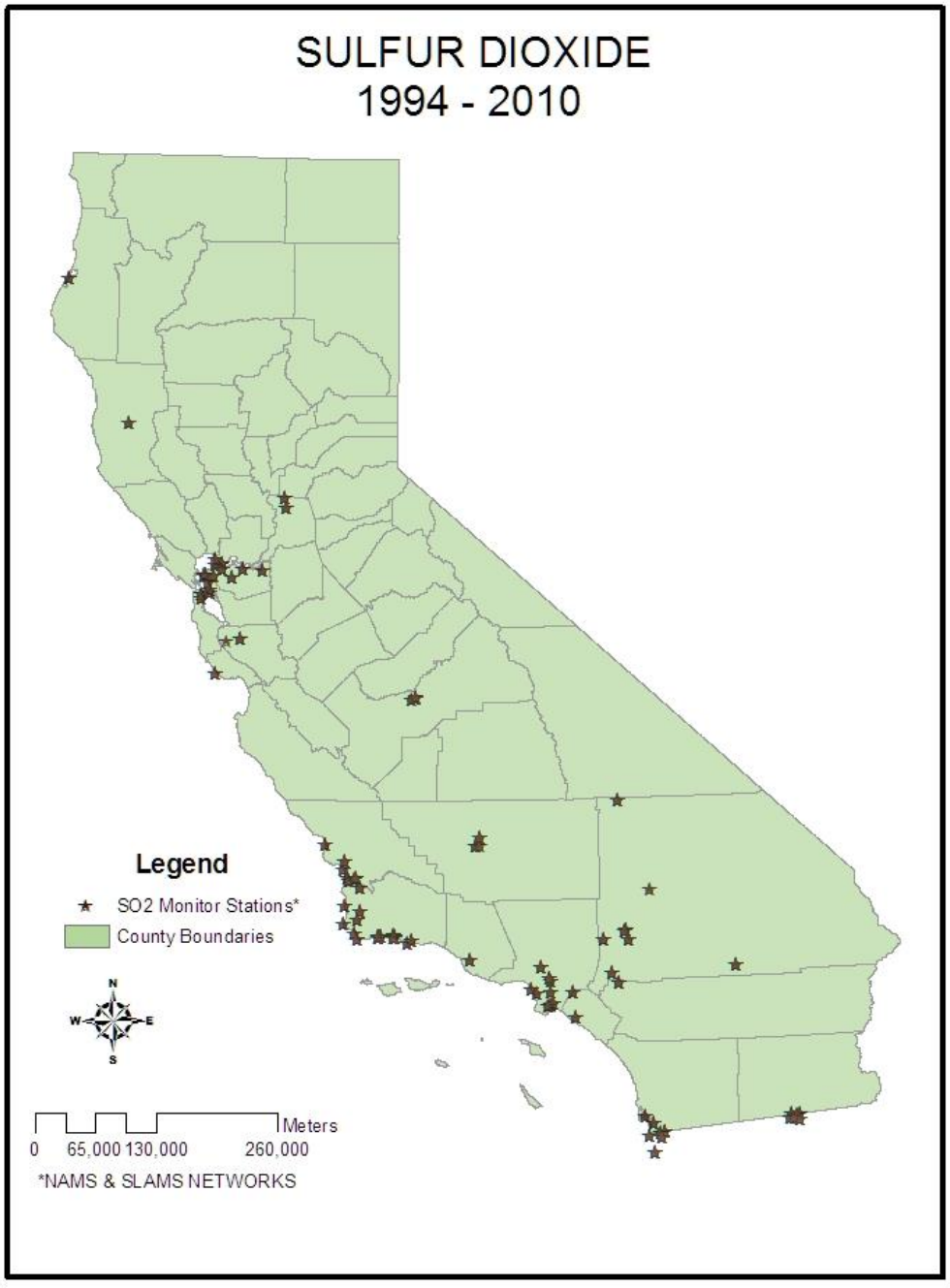

Figure 2.6 Available NO_2 pollutant monitors in California

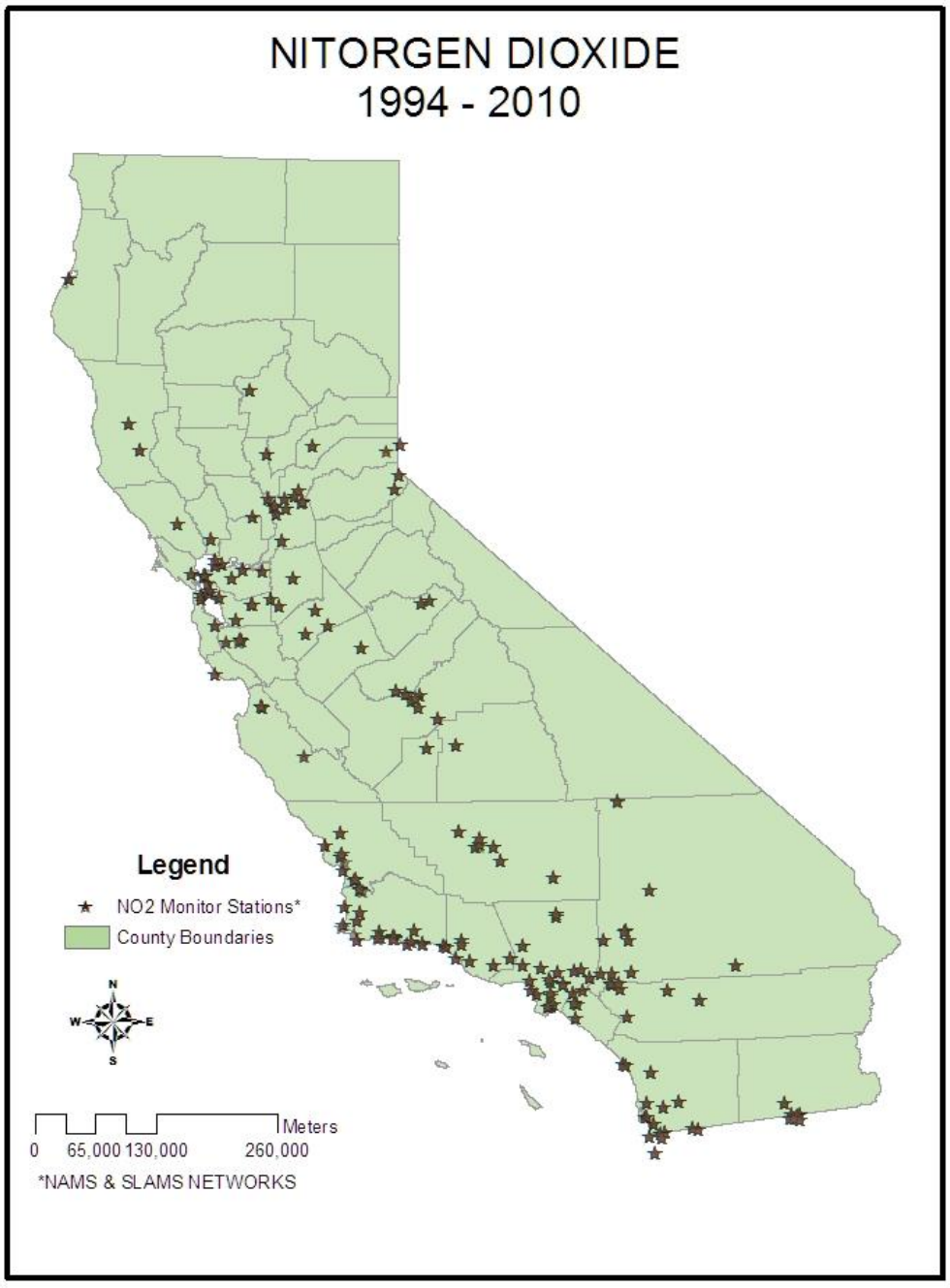

Figure 2.7 Available O_3 pollutant monitors in California

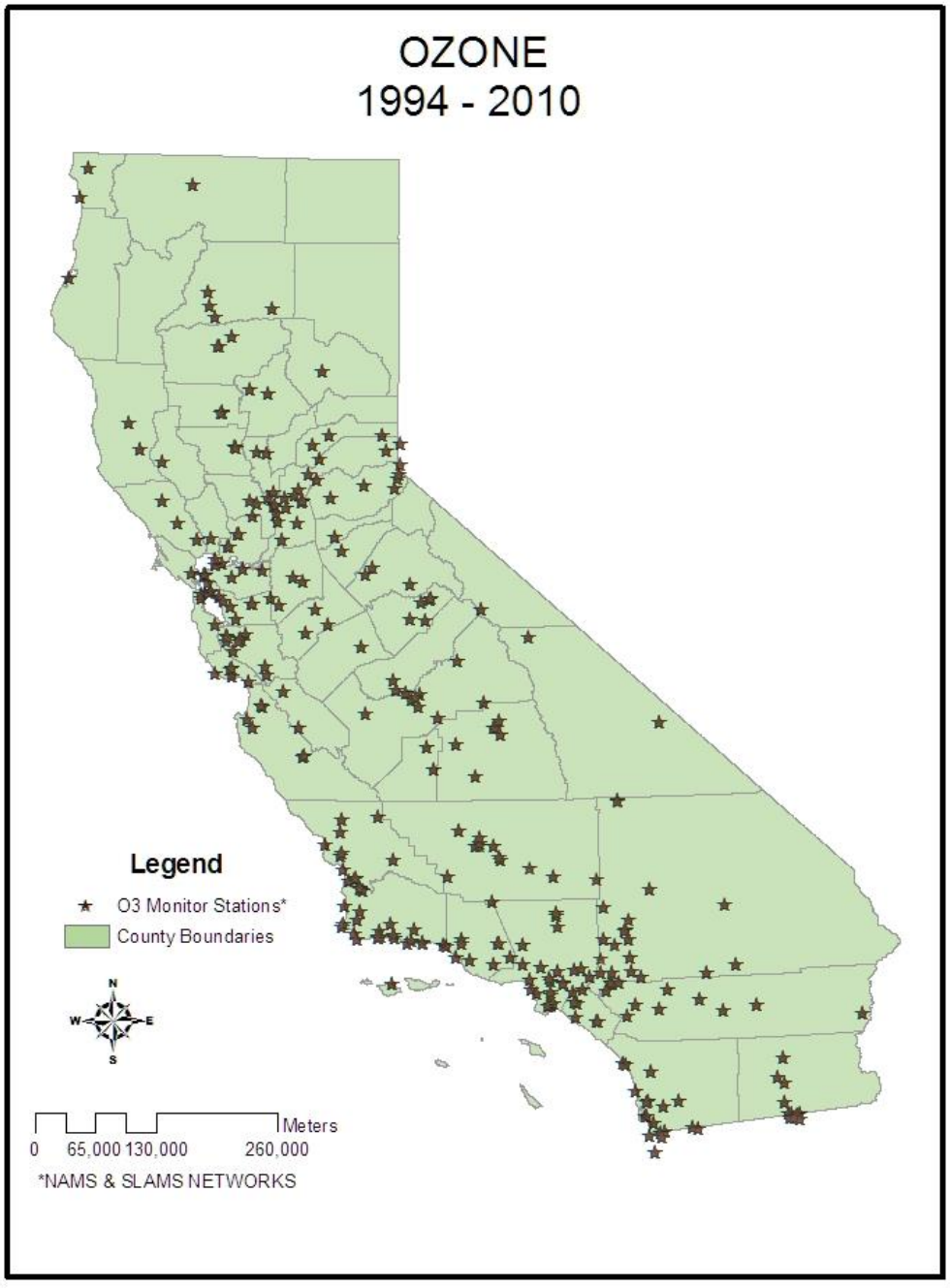

Figure 2.8 Available CO pollutant monitors in California

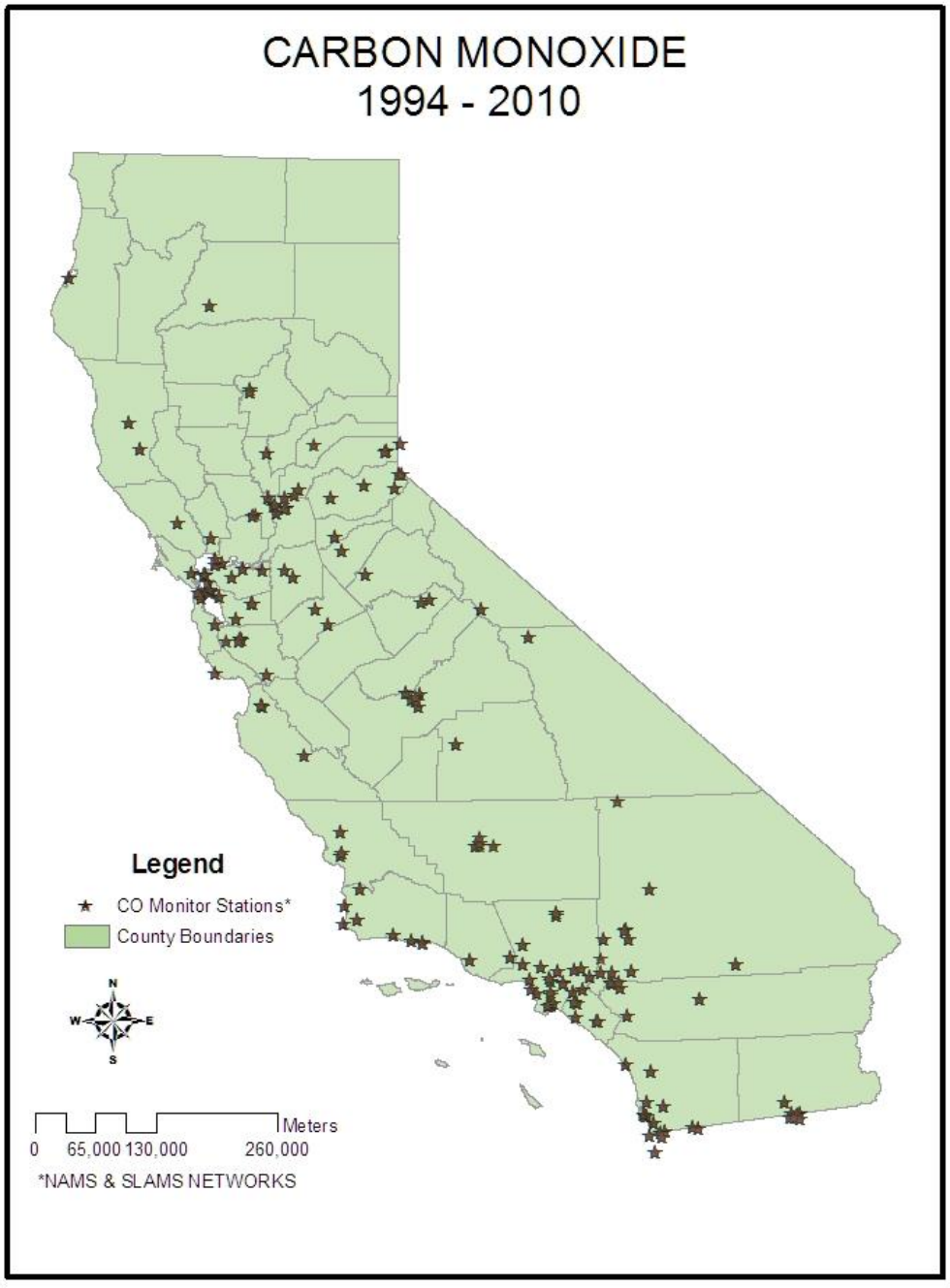

Table 2.3 Minimum detectable odds ratios (Quartile 4 vs. Quartile 1) for the air pollution-PTB association and smoking-PTB association

	2008-2010		**2006-2010**	
Power (%)	80	90	80	90
Air Pollution → PTB	1.33	1.39	1.27	1.32
Non-smokers	1.35	1.42	1.29	1.35
Smokers only	2.29	2.59	1.91	2.11
Smoking → PTB	1.46	1.55	1.36	1.42

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CHAPTER 3

CIGARETTE SMOKING AND PULMONARY TUBERCULOSIS IN NORTHERN CALIFORNIA[1]

Overview

A positive association between smoking and increased risk of tuberculosis disease is well-documented for populations outside the United States (U.S.). However, it is unclear whether smoking increases risk of tuberculosis in the U.S., where both smoking prevalence and disease rates are much lower than in the countries where previous studies have been conducted. To explore the tuberculosis-smoking association in a general U.S. population, we conducted a nested case-control study among members of Kaiser Permanente Northern California (KPNC). We identified all newly diagnosed cases of active pulmonary tuberculosis (PTB) disease between 1996-2010. Each of the 2380 cases were individually matched to two controls by age, gender, and race/ethnicity. Using adjusted logistic regression, increased PTB risk was observed among ever smokers (odds ratio (OR)=1.25; 95% confidence interval (CI): 1.00, 1.55), as well as current (OR=1.17; 95% CI:0.84, 1.62) and past (OR=1.29; 95% CI: 1.00, 1.66) smokers, compared to never smokers. Increased

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[1] Smith GS, Van Den Eeden SK, Baxter R, Shan J, Van Rie A, Herring AH, Richardson DB, Emch M, and Gammon MD. (2013). Cigarette smoking and pulmonary tuberculosis in northern California. [*Submitted for Review*]

intensity and duration of smoking were also positively associated with PTB risk. Our findings support the hypothesis that smoking is a risk factor for PTB in a general U.S. population, underscoring the importance of tobacco cessation and prevention programs in eliminating tuberculosis.

Introduction

Cigarette smoking is the number one cause of preventable deaths in the United States (U.S.), accounting for 443,000 (or 1 out of every 5) deaths each year (CDC 2011a). Approximately one in five adults (18+ years) in the U.S. smokes cigarettes. In contrast to the decreasing prevalence of smoking in the U.S. (CDC 2011b), rates continue to increase outside the U.S., particularly in regions with a high prevalence of tuberculosis (TB). For example, China and India rank among the top countries in the number of smokers as well as the largest number of active TB cases (1.4 million and 1.9 million, respectively) (Hassmiller 2006, Chan 2010).

TB is caused by Mycobacterium tuberculosis (Brosch 2002). As consistently demonstrated in studies conducted outside of the U.S. (Kolappan 2002, Tekkel 2002, Leung 2003, Miguez-Burbano 2003, Liendhart 2005, Gordon 2006, Shetty 2006, Chan 2010), smoking appears to increase a person's risk of developing TB. It remains unclear whether smoking increases the risk of TB in the U.S., where the prevalence of smoking and the rates of TB are much lower than many of the countries in which these previous smoking-TB studies have been conducted. Further, most of the previous studies conducted outside the U.S. focused on latent tuberculosis infection, rather than on active TB disease (Lin 2007).

Two previous U.S-based studies have explored the association between active TB disease and smoking among high risk populations. A case-control study of TB risk factors conducted from 1988-1990 in King County, Washington found an association increased risk for active tuberculosis among smokers where 45% of the clinic's patients were initially seen for immigration screening (Buskin 1994). In a 2003 case-control study, a positive association was observed between tobacco use and TB, but the study included only human immunodeficiency virus (HIV)-positive individuals on antiretroviral therapy (Miguez-Burbano 2003). Thus, the findings from these studies are notable, but difficult to generalize to the U.S. population as a whole.

To examine the hypothesis that cigarette smoking is associated with active pulmonary tuberculosis (PTB) disease risk in a general U.S. population, we conducted a case-control study nested within the large and diverse population of Kaiser Permanente Northern California (KPNC). The population-based study design will help to increase the generalizability of our findings to the American population.

Methods

Institutional Review Board approval for this nested case-control study was obtained from all participating institutions.

Study Population

All eligible participants were drawn from the KPNC membership, which provides integrated comprehensive care to approximately 3.3 million people (about 25% of the total

population in the geographic areas served). All incident cases of PTB diagnosed among adult KPNC members from January 1996 through December 2010, were included in the investigation. TB was defined as a positive TB culture or a prescription for at least 30 days of four or more anti-tuberculosis medications including isoniazid, rifampin, ethambutol, and pyrazinamide (CDC 2003). Electronic clinical databases were reviewed for each case of TB to ascertain site of disease, and only cases of pulmonary TB were included in this analysis. Controls were drawn from KPNC among members free of TB on the date of diagnosis of the index case for each control, and matched to cases (2:1) by age, gender, and race/ethnicity (Rothman 1998). To exclude recent immigrants from the study, individuals in the initial study population who had not been KPNC members for at least two years prior to the case date of diagnosis (n=3577) were dropped from the analysis. A total of 2380 cases of active PTB disease and 4738 tuberculosis-free controls were included in the study.

Exposure Assessment

Cigarette smoking status was obtained for study subjects by abstracting data from the electronic clinical databases, which had been collected and recorded at various visits over the length of membership at KPNC. Over time, increasing details on the subjects' smoking status were recorded in the KPNC databases; therefore, the association of smoking and PTB was explored for two different times during the study period (1996-2010 and 2008-2010). To examine the smoking-PTB relationship over the entire study period, 1996-2010, a derived variable was constructed from all smoking variables ascertained from the clinical databases. If smoking was indicated anywhere in records, the subject was considered an 'Ever' smoker; otherwise the subject was considered a 'Never' smoker. Only those indicating smoking on

the date of diagnosis were considered 'Current' smokers. Because the most comprehensive and complete smoking data occurred in the clinical databases between 2008-2010, we were able to categorize subjects as 'Current' and 'Past' smokers, as well as estimate smoking dose (cigarettes per day) and duration (years of smoking) during this period. Although we were unable to explore dose and duration for the 1996-2010 study period for the entire study population, reliance on a standard "Derived" smoking variable to categorize subjects as ever (current/past)/never smokers yields a much larger sample size with increased exposure variability.

Covariates

Selected covariates, including age, gender, race/ethnicity, and length of membership, were similarly obtained from electronic KPNC databases. In addition, outpatient, inpatient and pharmacy databases were used to determine covariate data on alcohol-related hospitalizations, history of TB drug use, immunological prescriptions (immune compromising medications known to increase TB risk), and other co-morbidities (e.g., chronic obstructive pulmonary disease (COPD), renal dialysis). HIV and diabetes mellitus II (diabetes) status were obtained from KPNC registries of these conditions. To obtain contextual indicators of socioeconomic status (SES) not routinely collected at KPNC (e.g., education, income, and percent foreign born within each subjects neighborhood at the time of diagnosis/selection), we constructed census-block level covariates, and current/historical residential addresses were linked to year 2000 U.S. Census data.

Statistical Analysis

Odds ratios (ORs) and 95% confidence intervals (CIs) for the association between

smoking status and PTB, among cases and controls selected from KPNC between 1996-2010, were calculated using unconditional logistic regression adjusted for all matching factors (age, gender, and race/ethnicity) (Hansson 2008). We used separate models to estimate effects associated with varying definitions of smoking status (ever vs. never smoking, and current/past vs. never smoking) over the entire 1996-2010 study period (n = 2380 cases and 4738 controls) and restricted to 2008-2010 (n = 734 cases and 1462 controls). In addition, we constructed models to consider dose (cigarettes smoked per day) and duration (years of smoking) as determined from data recorded in the clinical databases from 2008-2010.

Potential effect modification was assessed using likelihood ratio tests (Hosmer 1989) with and without inclusion of multiplicative interaction terms in the logistic regression model, and those terms with p-values <0.10 were considered statistically significant. When interaction was present, we calculated ORs for PTB stratified by indicators of exposure. Covariates considered as potential effect modifiers include: percent foreign born in residential census block, COPD, alcohol hospitalizations, Crohn's disease, renal dialysis, immunological prescriptions, diabetes, and HIV status. Only percent foreign born in the residential census block met our criteria for effect modification, and thus stratified results are shown for this covariate only.

Potential confounding was assessed using a greater than 10% change in estimate criterion (Greenland 1989) for the following covariates: median household income, percent foreign born in census block, education in residential census block, alcohol hospitalization, diabetes, and HIV status. No covariates assessed were determined to confound the association; thus all final models only included as covariates the matching factors (age, gender, and race/ethnicity).

Conditional logistic regression was also performed and yielded virtually identical results; therefore only results from unconditional logistic regression are shown. All statistical analyses were performed using SAS software (version 9.3; SAS Institute, Cary, NC).

Results

The distribution of select characteristics of cases of PTB disease and controls selected from the KPNC membership (**Table 3.1**) are similar for the study time periods, 1996-2010 and 2008-2010. As expected, the distributions for the two time periods reflect the KPNC population, as well as the population residing in the surrounding Bay area and environs served by KPNC (Gordon 2006). The mean age of study participants for the entire 1996-2010 study period was 50.5 years, ranging from 21 to 98 years of age; and the mean length of membership in KPNC was 10.3 years, ranging from 2 to 31 years. Since gender was a matching factor, 50% of both cases and controls were female. The study sample includes a high proportion of Hispanics and Asians, which is reflective of the general population at risk for TB in the U.S. and the heterogeneous population residing in this geographic area. The distribution of TB risk factors by case-control status are similar for the two time periods, but there are marked differences between cases and controls, with cases being more likely to have been diagnosed with COPD, HIV, and diabetes.

Smoking within our KPNC study population declined over time, from 14.2% in 1996 to 11.0% in 2010, which is consistent with the declining smoking prevalence reported by the California State Health Department (CDPH 2010). **Table 3.2** shows that the adjusted effect

estimates were elevated for the associations between PTB and smoking status and smoking patterns for the 2008-2010 period, when greater smoking detail was available. An adjusted OR for ever smoking relative to never smoking was 1.25 (95% CI: 1.00, 1.55). Once ever smokers were partitioned into current and past smokers, relative to non-smokers, the adjusted OR was 1.29 (95% CI: 1.00, 1.66) for past smoking and 1.17 (95% CI: 0.84, 1.62) for current smoking. Among past smokers, time since smoking cessation varied by case status, with 17% of cases quitting in the two years prior to the date of diagnosis, compared to only 4.0% of controls quitting in this same window.

Due to the manner in which smoking was collected in the electronic clinical databases, data is available on dose and duration for approximately only 40% of all ever smokers of the 2008-2010 KPNC study population, with more than twice as many past smokers missing the more detailed information than current smokers. As shown in **Table 3.2**, among the 2008-2010 KPNC study population, PTB risk increased with years of smoking; however, the increase does not appear to occur until 6 or more years of smoking. Among those with more than 30 years duration of smoking, the risk of developing PTB was more than two times greater than among never smokers. Among ever smokers, as compared with never smokers, those smoking up to one-half pack per day were at 1.47 (95% CI: 0.97, 2.21) times the odds of PTB, which increased to 1.78 (95% CI: 1.18, 2.69) for those smoking more than a half pack a day.

When we considered the full KPNC study population (1996-2010), for whom we created a standard "Derived" ever/never smoking variable, the effect estimates for the association between smoking status and PTB disease are more precise and modestly higher in magnitude. As shown in **Table 3.3**, the OR for ever smoking was 1.24 (95% CI: 1.10, 1.39)

relative to non-smokers, and, the ORs for current and past smoking were 1.17 (95% CI: 1.00, 1.37) and 1.31 (95% CI: 1.13, 1.52) respectively.

Effect modification of the association between smoking and PTB among the 1996-2010 KPNC study population was assessed by stratifying on the percent of the population that is foreign born in the subject's residential U.S. census block. As shown in **Table 3.4**, ever smoking was associated with a slight, non-significant 1.13 (95% CI: 0.82, 1.56) increase in the odds of PTB among those living in a census block with the fewest (up to 15%) foreign born individuals. In contrast, among those living in a census block with 15% to 30% and more than 30% foreign born individuals, ever smokers had an increased risk compared to never smokers [1.46 (95% CI: 1.07, 1.98) and 1.48 (95% CI: 1.11, 1.99), respectively].

Discussion

Among the population-based membership of a large and diverse integrated health system in northern California, we observed clear positive associations between smoking and active PTB disease. Ever smokers were at an increased risk of PTB when compared to never smokers. Past smokers were at a higher risk of developing PTB than current smokers. However, this latter observation may result from the fact that individuals often quit smoking as a result of the symptoms brought on by pulmonary TB which causes inflammation (Willemse 2004). There was also a significant elevation observed in the risk of PTB with increasing dose (packs per day) and duration (packs per year) of smoking; for example, the OR was increased by 78% for a half pack or more of cigarettes per day, and was doubled among those smoking greater than 30 years. The effect estimates observed for the

association between PTB and smoking in this Northern California population are consistent with a meta-analysis conducted by Bates et al. of studies conducted primarily outside the U.S., which estimated an elevated risk of PTB from ever smokers at 2.27 (95% CI: 1.90, 2.71) when compared to never smokers (Bates 2007).

Foreign born individuals are at a disproportionately increased risk of TB in the U.S. (CDC 2011a). However, information on immigration status/place of birth was not available for each individual study participant in this population. Instead, we explored effect modification of the association between smoking status and TB by the percent of the census block that was foreign born in the for each subject's neighborhood (at the time of diagnosis with active PTB for cases, or time of selection into the study for controls). We found evidence in support of a statistical interaction between the percent of foreign born individuals within a census block and smoking status. Our results are not entirely surprising as certain immigrant populations are born in countries with a high burden of active TB. Thus residence in such an area with a high immigrant population would most likely increase the risk of exposure to M. tuberculosis as residents are more likely to be foreign and therefore have been exposed in the past, as well as being more likely to be exposed while living in this area. However, mandatory screening confirms that all immigrants should be free of active TB upon entry into the U.S. (Liu 2009). In addition, the two-year KPNC membership requirement for subject eligibility used in our study helps to ensure that recent immigrants, who are at increased risk of reactivation, are unlikely to be included in the study.

Two previous studies (Buskin 1994, Miguez-Burbano 2003) conducted in the U.S. targeted very specific high-risk individuals (clinic with high proportion of immigrants and people living with HIV) and are thus not readily generalizable to the general U.S.

population. Our investigation takes advantage of a large cohort of subjects with uniform access to health care, which minimizes potential issues of selection bias. The KPNC membership has been previously demonstrated to be very similar to residents in the surrounding geographic area, and representative of the population at large, with the exception of its lower percentage of White non-Hispanics, individuals in the income extremes, and smokers (Gordon 2006). Additional advantages of this study include the large case-control nested within a cohort study design, which greatly enhances clear interpretation of the study findings, uniform access to health care, which minimizes potential issues of selection bias, and the availability of detailed clinical data, including risk factors for PTB, such as COPD, alcohol abuse, diabetes, renal dialysis and immunosuppressive conditions including HIV (Liu 2009, CDC 2011a).

Although this study was able to include assessment of a personal history of smoking, information on environmental tobacco smoke (ETS) exposure was not available. However, in California, stringent ETS exposure regulations (CDPH 2009) have minimized the impact of this potential exposure outside the home. Further, whether ETS at levels found commonly in U.S. homes is a risk factor for PTB is currently unknown.

Smoking status was recorded in several different forms in the KPNC electronic database over the study period, restricting the availability of detailed data on smoking patterns to the years 2008-2010. We explicitly designed our analytic strategy in consideration of this potential limitation. We first considered those with the detailed data from those selected for the 2008-2010 cohort, and then estimated effects among the entire 1996-2010 cohort. This strategy was facilitated by the strong similarities of the subjects within these two study time periods. The analysis from 1996-2010, using the "Derived"

ever/never smoking variable, yields nearly the same results seen as seen in 2008-2010, years with the most comprehensive smoking data, and strengthens the case for an association between smoking and PTB in the U.S. Although more than twice as many past smokers were missing data on dose and duration than current smokers in our study, this will have negligible influence on observed effects given the point estimate of the effect of smoking on risk of PTB is not significantly different among past smokers in this study population.

In conclusion, our nested case-control study is the first evaluation on whether cigarette smoking is associated with active PTB disease in a general U.S. population sample. Our findings of a positive link between smoking and PTB, and a positive association with dose and duration of smoking, in a well-characterized population suggest that, as is observed in non-U.S. based populations, smoking is a risk factor for PTB in the U.S. Unfortunately, smoking data are often not requested or recorded in present TB surveillance systems within the U.S. (CDC 2012). Inclusion of such information would foster research to identify effective strategies to reduce the risk of developing active PTB disease among Americans. Furthermore, knowledge of the increased risk of TB associated with smoking underscores the importance that smoking prevention and cessation programs can play in the goal of eliminating TB in the U.S.

Chapter 3 Figures and Tables

Table 3.1 Distribution of select characteristics of cases of pulmonary tuberculosis (PTB) disease and controls by year of diagnosis for cases and year of selection for controls between 1996-2010 and 2008-2010, Kaiser Permanente Northern California (KPNC)

	1996-2010		2008-2010	
Characteristic	**Cases**	**Controls**	**Cases**	**Controls**
From ECD				
All	2380 (100)	4738 (100)	734 (100)	1462 (100)
Gender				
Male	1180 (49.6)	2364 (49.9)	350 (47.7)	708 (48.4)
Female	1200 (50.4)	2374 (50.1)	384 (52.3)	754 (51.6)
Age				
21-34	527 (22.1)	1036 (21.9)	155 (21.1)	305 (20.9)
35-49	667 (28.0)	1318 (27.8)	230 (31.3)	453 (31.0)
50-64	647 (27.2)	1297 (27.4)	210 (28.6)	419 (28.7)
65+	539 (22.7)	1087 (22.9)	139 (18.9)	285 (19.5)
Race/Ethnicity				
White	418 (17.6)	835 (17.6)	112 (15.3)	225 (15.4)
Black	188 (7.9)	372 (7.9)	52 (7.1)	104 (7.1)
Asian	924 (38.8)	1838 (38.8)	304 (41.4)	603 (41.2)
Hispanic	464 (19.5)	929 (19.6)	132 (18.0)	269 (18.4)
Other	153 (6.4)	307 (6.5)	44 (6.0)	90 (6.2)
Unknown	233 (9.8)	457 (9.7)	90 (12.3)	171 (11.7)
Length of Membership				
2-5 years	793 (33.3)	1547 (32.7)	247 (33.7)	471 (32.2)
5-10 years	648 (27.2)	1257 (26.5)	191 (26.0)	395 (27.0)
10-15 years	343 (14.4)	656 (13.9)	112 (15.3)	204 (14.0)
15+ years	596 (25.0)	1278 (27.0)	184 (25.1)	392 (26.8)
Census Block Derived Data				
Median Household Income				
Median	$60,747	$61,414	$61,101	$61,709
(IQR*)	($46k-$77k)	($46k-78k)	($46k-76k)	($46k-$78k)
Percent Foreign Born				
Median	26.3	21.9	26.4	22.3
(IQR*)	(15.0-41.0)	(12.5-37.6)	(14.6-41.0)	(12.9-36.8)
> High School Education				
Median	62.9	64.1	61.9	63.4
(IQR*)	(49.4-75.1)	(50.4-76.8)	(49.7-74.4)	(50.1-76.5)

*Abbreviations: ECD = electronic clinical data; IQR = interquartile range; COPD = chronic obstructive pulmonary disease; and HIV+ = human immunodeficiency virus positive.

Table 3.1 continued Distribution of select characteristics of cases of pulmonary tuberculosis (PTB) disease and controls by year of diagnosis for cases and year of selection for controls between 1996-2010 and 2008-2010, Kaiser Permanente Northern California (KPNC)

Characteristics	1996-2010		2008-2010	
	Cases	Controls	Cases	Controls
From Billing Codes				
Tuberculosis Risk Factors				
None	1334 (56.1)	3185 (67.2)	405 (55.2)	920 (62.9)
COPD*	881 (37.0)	1329 (28.1)	273 (37.2)	444 (30.4)
Alcohol Hospitalization	107 (4.5)	173 (3.7)	33 (4.5)	72 (4.9)
Renal Dialysis	149 (6.3)	155 (3.3)	61 (8.3)	94 (6.4)
Immunological Prescriptions	0	0	0	0
Diabetes	465 (19.5)	567 (12.0)	140 (19.1)	202 (13.8)
HIV +*	50 (2.1)	13 (0.3)	11 (1.5)	4 (0.3)

*Abbreviations: ECD = electronic clinical data; IQR = interquartile range; COPD = chronic obstructive pulmonary disease; and HIV+ = human immunodeficiency virus positive.

Table 3.2 Adjusted* odds ratios (OR) and 95% confidence intervals (CI) for the associations between smoking status, duration and dose, and pulmonary tuberculosis (PTB) disease, among cases diagnosed and controls selected between 2008-2010, Kaiser Permanente Northern California (KPNC)

	Cases n=734	**Controls n=1462**	**OR***	**95% CI**	
Smoking Status					
Never	487	961	ref		
Ever	239	398	1.25	1.00	1.55
Past	160	255	1.29	1.00	1.66
Current	79	143	1.17	0.84	1.62
Years of Smoking					
Never Smoker	487	961	ref		
≤ 5 years	15	31	0.95	0.47	1.84
6-30 years	64	91	1.50	1.04	2.17
> 30 years	25	27	2.16	1.19	3.90
Packs per Day					
Never Smoker	487	961	ref		
≤ ½ pack	51	72	1.47	0.97	2.21
> ½ pack	53	65	1.78	1.18	2.69

*Adjusted for age, gender, and race/ethnicity

Table 3.3 Adjusted* odds ratios (OR) and 95% confidence intervals (CI) for the association between smoking status and pulmonary tuberculosis (PTB) disease, among cases diagnosed and controls selected between 1996-2010, Kaiser Permanente Northern California (KPNC)

	Cases n=2380	Controls n=4738	OR*	95% CI	
Smoking Status					
Never	1624	3418	ref		
Ever	756	1320	1.24	1.10	1.39
Past	406	674	1.31	1.13	1.52
Current	350	646	1.17	1.00	1.37

*Adjusted for age, gender, and race/ethnicity

Table 3.4 Adjusted* odds ratios (OR) and 95% confidence intervals (CI) for the association between "Derived" smoking status and pulmonary tuberculosis (PTB) disease, stratified by the percent of the population that is foreign born in the subject's residential U.S. census block at the time of subject diagnosis/selection+, among cases diagnosed and controls selected between 1996-2010, Kaiser Permanente Northern California (KPNC)

% Foreign Born in census block	"Derived" Smoking Status	Cases n=2260	Controls n=4474	OR*	95% CI	
Up to 15%	Never	361	1016	ref		
	Ever	203	467	1.13	0.82	1.56
15-30%	Never	503	1039	ref		
	Ever	228	403	1.46	1.07	1.98
Over 30%	Never	670	1168	ref		
	Ever	295	381	1.48	1.11	1.99

*Adjusted for age, gender, and race/ethnicity; + P-value =.079 for the interaction on a multiplicative scale

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CHAPTER 4

AIR POLLUTION AND PULMONARY TUBERCULOSIS AMONG A POPULATION-BASED SAMPLE OF NORTHERN CALIFORNIA RESIDENTS[2]

Overview

Ecologic analyses and animal experiments suggest a positive association between air pollution and tuberculosis. No previous epidemiologic studies have examined this hypothesis using individual-level data. We evaluated the association between ambient air pollutants and active pulmonary tuberculosis (PTB). A nested case-control study was conducted among members of Kaiser Permanente Northern California. All cases of active PTB diagnosed from 1996-2010 (n=2309) were matched to two controls (n=4604) by age, gender and race/ethnicity. Average individual-level concentrations of carbon monoxide (CO), nitrogen dioxide (NO_2), sulfur dioxide (SO_2), ozone (O_3), and particulate matter with aerodynamic diameter <2.5μm ($PM_{2.5}$) and 10μm (PM_{10}) for two years prior to diagnosis/entry into the study were estimated based on home address. In single-pollutant, adjusted conditional logistic regression models, the odds ratio (95% confidence interval) was 1.50 (1.15, 1.95) for the association between the highest CO quintile (vs. lowest); corresponding estimates were

[2] Smith GS, Van Den Eeden SK, Garcia C, Baxter R, Shan J, Van Rie A, Herring AH, Richardson DB, Emch M, and Gammon MD. (2013). Air pollution and pulmonary tuberculosis among a population-based sample of northern California residents. [*To Be Submitted*]

higher among never (1.68 (1.26, 2.24)) than ever smokers (1.19 (0.74, 1.92)). In contrast, NO_2–PTB associations were higher among ever (1.81 (1.13, 2.91)) than in never smokers (1.29 (0.97, 1.71). O_3 was inversely associated with PTB for both smokers and nonsmokers. No consistent patterns were observed for other pollutants. Findings from multi-pollutant models were similar. Among a population-based sample of Northern California residents, exposure to ambient CO was positively associated with PTB. This association requires confirmation in other U.S.-based and international populations.

Background

Air pollution is a substantial cause of morbidity and mortality worldwide, resulting in major public health impacts and millions of dollars lost each year (Raviglione 1995, WHO 2010). Recent ecologic studies conducted in several countries, including the United States (U.S.), suggest that ambient air pollution may contribute to an increased risk of tuberculosis (TB) (Shilova 2004, Iwai 2005, Tremblay 2007) or pulmonary TB (PTB) (Smith 2013). A cross-sectional study in Japan reported a correlation between total suspended particles in air and TB (Iwai 2005). While exploring seasonal fluctuations of TB incidence in an ecologic analysis in Russia, Shilova et al. (2004) found that along with climatic factors, atmospheric pollutants (including NO, CO, and SO_2) were associated with TB incidence In a recent case-case comparison based on 196 hospital-based patients in Los Angeles, a correlation was observed between small particle-size particulate matter ($PM_{2.5}$) and TB (Jassal 2012). However, large epidemiologic studies that consider a wider variety of individual-level air

pollutant exposures in a population-based study sample that is more generalizable to the general population are lacking.

Mycobacterium tuberculosis is the causative agent of TB. The immune system is most often able to contain this infection; however weakened immunity, caused by HIV, diabetes, and host of other factors, can cause TB to reactivate (Raviglione 1995, WHO 2010). Biologically, air pollutants could be involved in the reactivation of TB through altering macrophage function, thereby increasing susceptibility to developing active TB. Tumor necrosis factor (TNF)-α and interferon-gamma (IFN-γ) play a central role in containing and inhibiting the growth of mycobacteria (Flad 1995, Döffinger 2004, Fremond 2005), but levels of these have been shown to decrease in animal experiments with pollutant exposures (Hirmatsu 2005, Saito 2002a,b). Long-term exposure to diesel exhaust has been shown to decrease the expression levels of interleukin-10, interferon-gamma, and inducible NO synthase mRNAs reduce TNF-α and IFN-γ production and increase the mycobacterial load in the mice (Hirmatsu 2005, Saito 2002a,b). Further, the inhibition of TNF-α by drugs in clinical trial has been linked to TB reactivation, implying that the association in mice may be applicable to humans (Jacobs 2007).

Objectives

This study aims to investigate whether exposure to criteria air pollutants (sulfur dioxide (SO_2), nitrogen dioxide (NO_2), carbon monoxide (CO), ozone (O_3), particulate matter with aerodynamic diameter of 10 μm or less (PM_{10}) and particulate matter with aerodynamic diameter of 2.5 μm or less ($PM_{2.5}$)) are associated with increased risk of active PTB in a well-

defined population of northern California residents.

Methods

Study Population

We conducted a nested case-control study of the association between air pollution and PTB disease among the members of Kaiser Permanente Northern California (KPNC), an integrated healthcare delivery system providing care to 3.3 million residents in the greater San Francisco, Oakland, San Jose, Sacramento and Fresno areas. KPNC serves approximately 25-30% of the total population in the geographic areas served. Cases included all newly diagnosed active PTB among adult KPNC members. Case identification included individuals with a new clinical diagnosis and prescription for at least 30 days of four or more anti-tuberculosis medications including isoniazid, rifampin, ethambutol, and pyrazinamide or a positive TB culture, between January 1996 and December 2010. For cases, diagnosis date, use of anti-TB drugs and relevant laboratory assays were abstracted from the KPNC clinical data bases. Controls, selected from KPNC members free of TB on the index date of diagnosis of the case, were matched individually to cases (2:1) by age, gender, and race/ethnicity. All cases and controls were KPNC members for a minimum of 2 years prior to entry into the study.

Exposure Assessment

Exposures estimates for each individual were constructed using average ambient concentrations of $PM_{2.5}$, PM_{10}, O_3, NO_2, CO, SO_2 from all relevant monitors operating in

California in the 24 months prior to diagnosis/entry into the study. Based on the assumption that air pollution acts to increase susceptibility to develop PTB upon exposure to *Mycobacterium tuberculosis*, we a priori posited that the etiologically-relevant exposure window was the period within 24 months prior to of initial exposure (Raviglione 1995, WHO 2010). Pollutant concentration surfaces were generated using monitoring stations from California's State and Local Air Monitoring Network (http://www.arb.ca.gov/aqd/netrpt/netrpt.htm) with at least 75% completeness in each month. The state and local agencies began monitoring $PM_{2.5}$ in1999. The Interagency Monitoring of Protected Visual Environments (IMPROVE) Network was included to supplement $PM_{2.5}$ measurements; since this IMPROVE network provides additional coverage of $PM_{2.5}$ in less populated areas of the state.

To assess individual-level air pollution exposure, geocoded patient addresses (current and up to two years prior to diagnosis) were assigned the pollutant concentration of the closest available monitor. For each study participant, monthly exposures for each residence of record were averaged to define an aggregate approximation of exposures at each residence of record during the 24 months prior to diagnosis/entry into study.

To assess individual-level air pollution exposure, geocoded patient addresses (current and up to two years prior to diagnosis) were assigned the pollutant concentration of the closest monitor. For each study participant, the cumulative exposure was estimated by accumulating monthly exposures for each residence of record during the 24 months prior to index date.

Potential individual-level confounders and effect measure modifiers were ascertained from electronic clinical databases of KPNC, including data on age, gender, race/ethnicity, length of KPNC membership, cigarette smoking, alcohol hospitalization, HIV status, co-morbidity (e.g., diabetes, COPD, renal dialysis), and residential address history. To examine the potential role of smoking on the association between air pollution and PTB, a variable was constructed from all other smoking information recorded in the KPNC electronic clinical database; if smoking was indicated anywhere in records, the subject was considered an ever smoker, otherwise the subject was considered a never smoker. Additionally, U.S. Census block level variables (e.g., education, median household income, and percent foreign born) were created as indicators of socioeconomic status for factors not routinely collected at KPNC.

Statistical Analysis

We used conditional logistic regression analysis, adjusting for all matching factors (age, gender, and race/ethnicity), to estimate the odds ratios (OR) and 95% confidence intervals (CI) for the association between PTB and each average air pollutant concentration (SO_2, NO_2, CO, O_3, PM_{10} and $PM_{2.5}$) assigned to each individual for the 24 months prior to diagnosis date/date of study entry. We grouped pollutant exposures into quintiles according to their population distribution, and exposure levels below the 20th percentile were used as the referent category for each pollutant. Monthly air pollutant averages were considered, but results did not differ from the 24-month averages, and thus the results for the 24-month averages are shown.

Potential effect modification (by cigarette smoking, alcohol hospitalizations, diabetes, COPD, HIV status, renal dialysis, and percent foreign born in census block) was assessed using likelihood ratio tests (Hosmer 1989) with and without inclusion of multiplicative interaction terms in the logistic regression model ($p<0.10$). Terms considered statistically significant were included in stratified analyses; only cigarette smoking met these criteria and thus only these results are shown.

Confounding was assessed using a greater than 10% change in estimate criterion (Greenland 1989) for several risk factors that could potentially confound the relationship between air pollution and PTB: length of KPNC membership, median household income, percent foreign born in census block, education in residential census block, alcohol hospitalization, diabetes, and HIV status. Using this criterion, no factors were found to confound the air pollution-PTB associations and were therefore not included in the final analysis. Final models only included the matching factors (age, gender, and race/ethnicity).

A multi-pollutant analysis for air pollution and PTB association was also considered using a forward model building approach. All analyses were conducted using SAS (version 9.3; SAS Institute, Cary, NC).

Results

Characteristics of PTB cases and matched controls nested within the KPNC membership are summarized in **Table 4.1**. We identified 2309 cases of PTB, and 4604 controls matched 2:1 to cases by age, gender, and race/ethnicity, which were drawn from the

existing KNPC membership during the study period, 1996-2010. Less than a third (2028) of the participants were classified as ever smokers. The study population was split equally by gender, though a larger percentage of males were considered ever smokers than females (68.5% vs. 31.5% among cases and 63.9% vs. 36.1% among controls). As expected, never smokers were younger in age than ever smokers. The higher proportion of Asians and Hispanics within this study is consistent with the racial/ethnic distribution of residents in this geographic area, as well as the at-risk population for TB in the U.S. (CDC 2011a).

As shown in **Table 4.2**, average ambient air pollution concentrations in the 24 months prior to the diagnosis date varied greatly for participants. Mean air pollution concentrations were the same among never smokers and ever smokers for all pollutants, with the exception of CO. Ambient CO concentrations measured in the 24 months prior to PTB diagnosis/entry into the study were slightly higher for never smokers than ever smokers. Because the number and the location of available monitors within the network varied throughout the study period, the number of cases and controls with available pollutant data also varied over the study. As shown in **Table 4.3**, Spearman's correlation air-pollutant averages in the 24 months prior to date of diagnosis showed only moderate correlations between pollutants. The strongest correlation observed between ambient averages of air pollutants was seen for PM_{10} and $PM_{2.5}$ (r=0.61).

Figure 4.1 presents the odds ratios (ORs) and 95% confidence intervals (CI) for the associations between air pollution, in quintiles, and PTB in the 24 months prior to diagnosis date/study entry, among all cases and matched controls. All effect estimates are adjusted for the matching factors of age, gender and race/ethnicity. There was no evidence of association between each of the criteria pollutants $PM_{2.5}$, PM_{10}, and SO_2 and PTB. Examination of the

air pollution-PTB associations stratified on smoking status revealed that any weak associations observed for many of these pollutants were attenuated once we considered the effects of smoking (**Figure 4.2**).

As shown in **Figures 4.1-4.2,** and **Table 4.4**, CO and NO_2 were positively associated with PTB in this population-based sample; however the confidence intervals for these estimates were wide. The highest effect estimates among all subjects (regardless of smoking status) for the association of ambient air pollution and PTB were observed for CO (OR=1.50 (95% CI: 1.15, 1.95)), as shown in **Table 4**. Additionally, as measured concentrations of CO exposure increased, the associated odds of PTB increased. This dose-response pattern persisted among both never and ever smokers; however, the effect estimates for the highest quintiles of 8hr CO exposure, compared to the lowest, were more pronounced among never smokers (OR=1.68 (95% CI: 1.26, 2.24)) then ever smokers (OR=1.19 (95% CI: 0.74, 1.92).

NO_2 was also positively associated with PTB in this population-based sample, with an apparent dose-response pattern between exposure and PTB odds. However, once stratified on smoking status, the dose-response pattern was less pronounced. Among ever smokers the magnitude of the association between NO_2 and PTB was more pronounced (OR=1.81 (1.13, 2.92) for the highest vs. the lowest quintile of exposure), than among never smokers (OR=1.29 (0.97, 1.71).

An unexpected inverse association was observed for 8h O_3 and PTB, with all exposures above the lowest quintile resulting in decreases in the effect estimates. Those in the highest quintile of O_3 exposure had a considerable decrease in risk of PTB (OR= 0.66 (0.55, 0.79)) when compared to those in the lowest quintile. The association between O_3 and PTB did not differ upon stratification by smoking.

Multi-pollutant analyses were conducted to assess the association of combined exposure to several air pollutants on odds of PTB (data not shown). However, the inclusion of additional pollutants into the model had little influence on the estimated associations for PTB and the air pollutants in this study, including the effect estimates for CO and NO_2 exposure concentrations. Stratified analyses of the multi-pollutant model estimates by cigarette smoking status yielded results similar to the single pollutant analysis.

Discussion

In this sample of 6,914 northern California residents, which is the first large population-based study to assess the potential associations between individual-level estimates of the criteria air pollutants and PTB, increased exposure to CO concentrations in the 24 months prior to diagnosis, compared to those with the lowest, were observed to be associated with a 50% elevation in the odds of developing PTB. When the analysis was stratified by smoking status, these increased estimates remained for CO, even among nonsmokers, although confidence intervals were wide. Stratified analysis showed slightly stronger effects among ever smokers, suggesting that a saturation of CO exposure may occur in ever smokers preventing any additional impact from air pollution from further increasing the risk of PTB in this group of individuals. Positive associations were also noted for NO_2, although the magnitude of the association was stronger in nonsmokers than in smokers. In addition, an unexpected inverse association was observed for O_3. No consistent associations were observed among the other pollutants studied, including $PM_{2.5}$, PM_{10}, and SO_2.

The associations observed in CO are consistent with other available evidence on this

issue. Exposure to ambient CO in the U.S. in this setting is primarily a marker of exposure to combustion products and secondary vehicular traffic (U.S. EPA 2010), and experimental studies show that diesel exhaust affects immune processes that inhibit TB in mice (Saito 2002a, Saito 2002b, Hiramatsu 2005). This also is consistent with the hypothesis that coal consumption may be linked to TB since both have followed similar trends in the U.S., Canada, and China (Tremblay 2007); CO is a known pollutant from coal firing power plants.

Risk of PTB was also associated with exposure to NO_2. This gaseous pollutant produced primarily from combustion sources, such as motor vehicle exhaust, and electric generating units. Individuals living near busy roads are particularly vulnerable to NO_2 pollution and related health effects. Although individual pollutant exposure depends predominantly on local outdoor concentrations, indoor pollution such as smoking and using gas appliances may alter exposure levels (U.S. EPA 2008).

While Nitric oxide (NO), oxygen (O_2), and CO are normally important for host defense against TB recent studies have demonstrated the ability of *M. tuberculosis* sense the presence of NO and CO and alter gene expression to bypass CO toxicity (Zacharia 2012). Previous studies have reported unusual differences in concentrations of O_2, NO, and CO in organs, tissues, and cells models (Sherman 2001, Ohno 2003, Voskuil 2008) and it has been hypothesized that while optimal levels of NO and CO keep TB bacteria inactive, extreme amounts of CO, and also NO, could result in abnormal immune responses (Kumar 2008).

In this study, an inverse association was seen with O_3 and PTB, with all exposures above the lowest quintile resulting in decreases in PTB risk for never smokers. This finding is supported by the fact that for years UV light, of which O_3 is a byproduct, has been used to kill TB bacteria (Escombe 2009). An experimental study exploring this association found

that exposure to increased, but non-toxic levels of O_3 (similar to the levels observed in our population) resulted in reduced numbers of TB bacilli (Belianin 2004).

We observed no consistent association between $PM_{2.5}$ and PTB, which is in contrast to the one previous case-case comparison (Jassal 2012). The 2012 study by Jassal and colleagues reported a positive association between $PM_{2.5}$ and TB, but the finding is based on less than 200 patients in a single Los Angeles hospital. In contrast, our results are based on data from a nested case-control study conducted among 6913 subjects that are representative of the Northern California population from which they are drawn. Further, our case definition of active PTB was carefully defined, and compared to a control population that did not have active TB. Thus, our findings are more applicable to the general population. Finally, our study considered a larger number of criteria air pollutants. Thus, additional carefully conducted studies addressing this issue are needed.

The method used to assess individual-level air pollution exposure in our study is an improvement from previously conducted ecologic studies. However, assigning individual average exposures by relying on the ambient air monitoring likely introduces some exposure misclassification. This issue may be particularly relevant to CO assessments, because pollutant measurements may differ within a specific geographic area due to climatic variations. Further, measurement error might have occurred as a result of variations in residential ventilation systems, levels of physical activity, or time spent outdoors, away from home or traveling by car outside of their residential area. The latter is important as considerable CO exposure can be experienced while driving or riding in vehicles (U.S. EPA 2010). However, these potential sources of error are common to many epidemiologic studies focused on assessing the health effects associated with air pollution, which have also used

our approach to estimate individual-level ambient concentrations (Brauer 2008, Brunekreef 2009, Lipsett 2011). These errors are usually assumed to be nondifferential, given that exposure estimates are constructed without knowledge of case-control status; hence this type of exposure misclassification is likely to lead to underestimation of the effect estimate.

To examine the potential role of smoking on the association between air pollution subjects were considered never smokers if there was no indication of smoking anywhere in records. This method of categorizing smokers may have resulted in underestimation of smoking prevalence in this study population if smoking status is underreported in the clinical data. This underestimation could lead to a spurious result when smoking status was included in the analyses if the error was differential by air pollution exposure status. However, assignment of smoking status was made blinded to case status and air pollution exposure status, thus differential misclassification of smoking seems unlikely.

Recent immigration status, a factor that puts individuals at disproportionately increased risk of TB in the U.S. (CDC 2011a), was not available for cases and controls in this population. We instead, explored used percent of the census block that was foreign born as a confounder. Use of this this proxy is inexact; however, mandatory screening confirms that all immigrants should be free of active TB upon entry into the U.S. (Liu 2009). Furthermore, to ensure that recent immigrants are unlikely to be included in the study, we implemented a two-year KPNC membership requirement for subject eligibility.

This nested case-control study conducted within a large well defined population has several advantages. The KPNC membership from which cases and controls were drawn is very similar to residents in the surrounding geographic area, and is generally representative

of the population-at-large, with the following exceptions: the KPNC includes a somewhat better educated membership than the surrounding geographic population, as well as fewer individuals in the income extremes, and smokers (Gordon 2006). The uniform access to health care in this population minimizes the potential for selection bias, and the availability of detailed clinical data, allowed us to explore the potential confounding effects of numerous risk factors associated with TB including COPD, alcohol abuse, diabetes, renal dialysis and immunosuppressive conditions including HIV (CDC 2011a, Liu 2009). Finally, the availability of the criteria air pollution data permitted us to construct ambient exposure concentrations for all KPNC study participants for the 24-month period prior to a diagnosis of PTB, which is considered the critical window between exposure to mycobacteria and development of active PTB (Raviglione 1995, WHO 2010).

The persistence of ambient air pollution remains a major public health problem, as millions worldwide die each year from causes directly related to air pollution (WHO 2011). While levels of air pollution have continually dropped in the U.S. and other developed countries in recent years, levels in developing countries remain high and are even increasing in many areas (WHO 2011). Many of the same countries with high levels of air pollution are also burdened with the highest levels of TB and increasing prevalence of cigarette smoking (Hassmiller 2006, Cohen 2007).

In conclusion, the results from our nested case-control study showed a positive association between ambient concentrations of CO and risk of PTB among a population-based sample of residents in Northern California, however confidence intervals were wide. This is the first large, population-based analytic epidemiologic study with individual-level estimates of air pollution concentrations conducted in the U.S., thus future studies in places

similar to the U.S. (low TB rates and air pollution levels) using a larger sample size are needed to confirm our findings. Studies are also needed in countries outside the U.S. which experience higher rates of TB and increased exposure to air pollution. Given that large number of people worldwide infected with *Mycobacterium tuberculosis* and exposed to high air pollution concentrations, any association between air pollution and TB is of considerable public health importance, as attention to the impact of air quality may contribute to global TB control.

Table 4.1 Distribution of matching characteristics of pulmonary tuberculosis disease (PTB) cases and matched controls nested within the Kaiser Permanente Northern California (KPNC) membership, 1996-2010.

	All		Ever Smokers		Never Smokers	
	Cases n=2309	**Controls n=4604**	**Cases n=737**	**Controls n=1291**	**Cases n=1572**	**Controls n=3313**
Gender						
Male	1144 (49.6%)	2291 (49.8%)	505 (68.5%)	825 (63.9%)	639 (40.7%)	1466 (44.3%)
Female	1165 (50.4%)	2313 (50.2%)	232 (31.5%)	466 (36.1%)	933 (59.4%)	1847 (55.6%)
Age						
21-34	512 (22.1%)	1000 (21.7%)	94 (12.8%)	202 (15.7%)	418 (26.6%)	798 (24.1%)
35-49	648 (28.1%)	1271 (28.1%)	178 (24.1%)	305 (23.6%)	470 (29.9%)	966 (29.2%)
50-64	631 (27.3%)	1270 (27.6%)	268 (36.4%)	418 (32.3%)	363 (23.1%)	852 (25.7%)
65+	518 (22.4%)	1063 (23.1%)	197 (26.7%)	366 (28.4%)	321 (20.4%)	697 (21.0%)
Race/Ethnicity						
Non-Hispanic White	402 (17.4%)	811 (17.6%)	175 (23.7%)	288 (22.3%)	227 (14.4%)	523 (15.8%)
Black	184 (8.0%)	356 (7.7%)	79 (10.7%)	135 (10.5%)	105 (6.7%)	221 (6.7%)
Asian	894 (38.7%)	1801 (39.1%)	222 (30.1%)	371 (28.7%)	672 (42.7%)	1430 (43.2%)
Hispanic	451 (19.5%)	912 (19.8%)	142 (19.3%)	263 (20.4%)	309 (19.7%)	649 (19.6%)
Other	150 (6.5%)	298 (6.4%)	56 (7.6%)	124 (9.6%)	94 (6.0%)	174 (0.5%)
Unknown	228 (9.9%)	426 (9.3%)	63 (8.5%)	110 (8.5%)	165 (10.5%)	316 (9.5%)

Table 4.2 Mean and percentile distribution of estimates of average ambient criteria air pollution concentrations cumulated over the 24-month period prior to the pulmonary tuberculosis (PTB) diagnosis/entry into study for cases/matched controls nested within the 1996-2010 KPNC membership with available pollutant monitoring data.

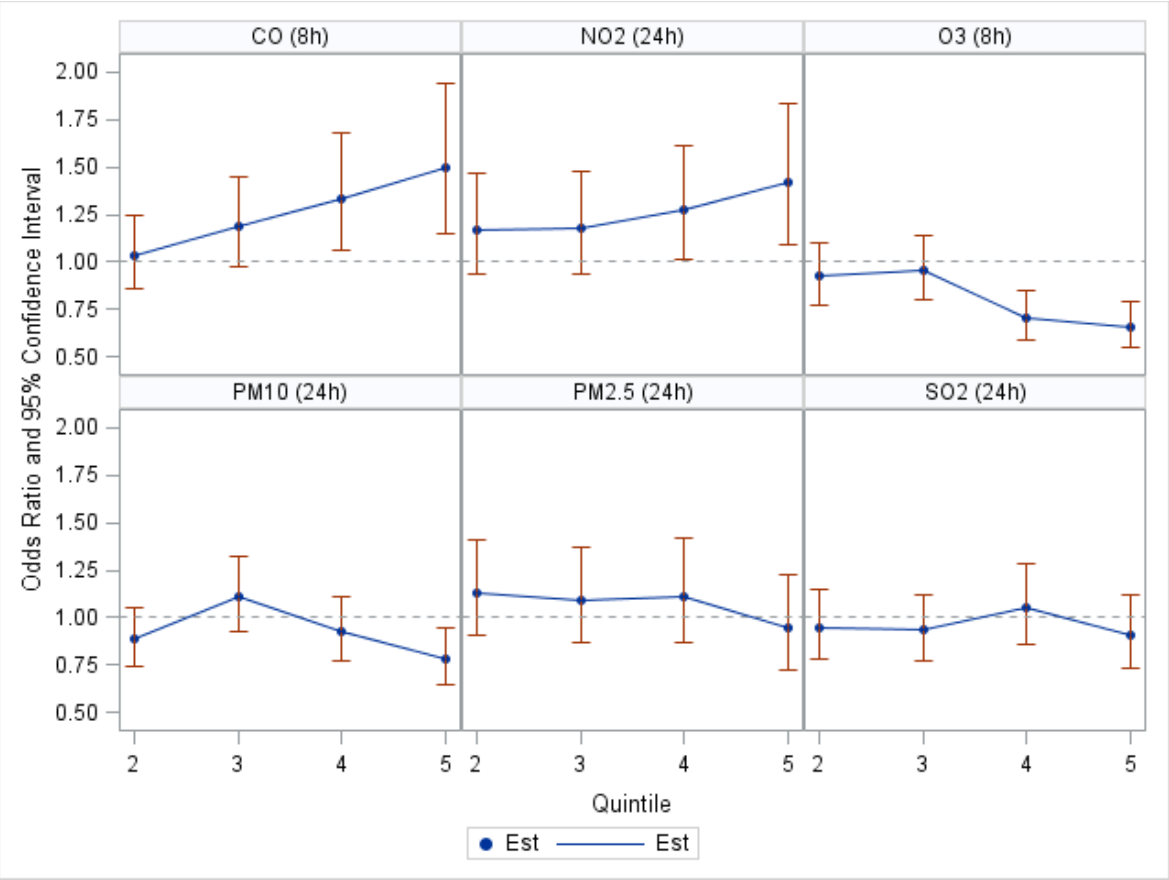
	N (%) with Pollutant Data				Percentile Distribution				
Air Pollutant	Cases	Controls	Mean	Min	20th	40th	60th	80th	Max
24h $PM_{2.5}$ ($\mu g/m^3$)	1842 (79.8)	3661 (79.5)	9.9013	0.1408	8.5600	9.1840	10.3324	11.6602	26.4783
24h PM_{10} ($\mu g/m^3$)	2309 (100.0)	4604 (100.0)	21.7027	9.1000	18.3896	19.8733	21.6487	24.4700	56.6082
24h SO_2 (ppm)	2248 (97.4)	4439 (96.4)	0.0013	0.0001	0.0009	0.0011	0.0013	0.0018	0.0039
24h NO_2 (ppm)	2309 (100.0)	4601 (99.9)	0.0138	0.0003	0.0098	0.0133	0.0151	0.0177	0.0390
8h O_3 (ppm)	2309 (100.0)	4604 (100.0)	0.0329	0.0178	0.0279	0.0301	0.0330	0.0378	0.0670
8h CO (ppm)	2309 (100.0)	4598 (99.9)	0.8420	0.0983	0.5481	0.6793	0.8760	1.1114	3.0572

Table 4.3 Spearman correlation coefficients* for the estimates of cumulative ambient criteria air pollutant concentrations, 0-24 months, among pulmonary tuberculosis (PTB) cases and matched controls nested within the 1995-2010 KPNC membership.

	All					
Air Pollutant	24h $PM_{2.5}$	24h PM_{10}	24hr SO_2	24h NO_2	8h O_3	8h CO
24h $PM_{2.5}$	1	0.61	0.12	0.28	0.25	0.35
24h PM_{10}		1	0.09	0.33	0.09	0.42
24h SO_2			1	0.19	-0.24	0.30
24h NO_2				1	-0.33	0.23
8h O_3					1	-0.28
8h CO						1

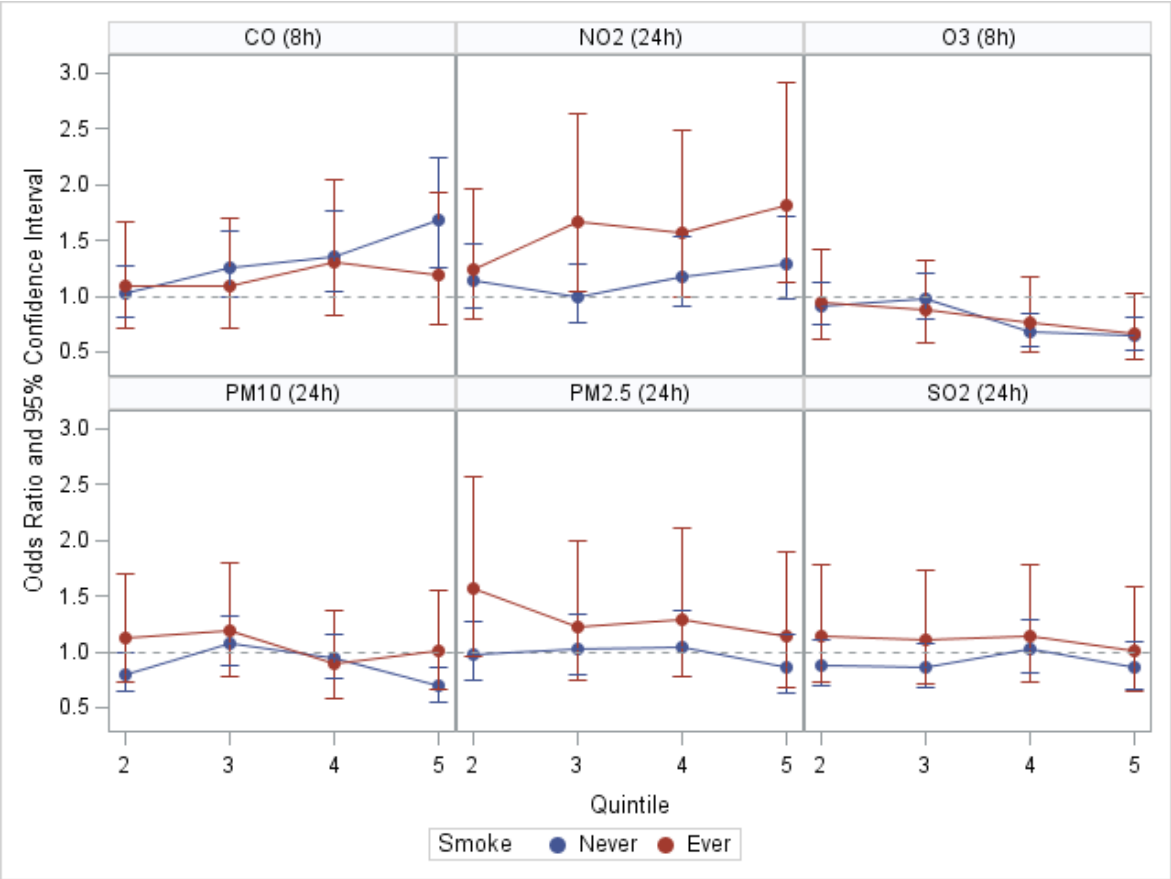
* All coefficients statistically significant ($p<0.05$)

Figure 4.1 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile in the estimates of ambient criteria air pollutants concentrations within the 24 months prior to diagnosis date, among all cases and matched controls nested within the 1996-2010 KPNC membership.



*Adjusted for the matching factors (age, gender, and race/ethnicity).

Figure 4.2 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile in estimates of ambient criteria air pollutants concentrations within the 24 months prior to diagnosis date, among all cases and matched controls nested within the 1996-2010 KPNC membership stratified by smoking status (ever vs. never smokers).



*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table 4.4 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among all cases and matched controls nested within the 1996-2010 KPNC membership, and stratified by smoking status (ever vs. never smokers).

Air Pollutant	Quintile	All	Never Smokers	Ever Smokers
$PM_{2.5}$	1	ref	ref	ref
	2	1.13 (0.90, 1.41)	0.98 (0.75, 1.27)	1.57 (0.95, 2.58)
	3	1.09 (0.87, 1.37)	1.03 (0.79, 1.34)	1.22 (0.75, 2.00)
	4	1.11 (0.87, 1.42)	1.04 (0.78, 1.37)	1.28 (0.78, 2.11)
	5	0.94 (0.73, 1.23)	0.85 (0.64, 1.15)	1.13 (0.68, 1.89)
PM_{10}	1	ref	ref	ref
	2	0.89 (0.74, 1.06)	0.80 (0.65, 0.99)	1.12 (0.73, 1.70)
	3	1.11 (0.93, 1.32)	1.07 (0.87, 1.32)	1.19 (0.78, 1.80)
	4	0.93 (0.77, 1.11)	0.94 (0.76, 1.16)	0.90 (0.59, 1.37)
	5	0.78 (0.65, 0.94)	0.69 (0.55, 0.87)	1.01 (0.66, 1.56)
SO_2	1	ref	ref	ref
	2	0.95 (0.79, 1.15)	0.88 (0.70, 1.10)	1.15 (0.74, 1.79)
	3	0.93 (0.78, 1.12)	0.86 (0.69, 1.08)	1.11 (0.72, 1.73)
	4	1.05 (0.86, 1.29)	1.02 (0.80, 1.29)	1.14 (0.73, 1.79)
	5	0.90 (0.73, 1.12)	0.86 (0.67, 1.10)	1.01 (0.64, 1.58)
NO_2	1	ref	ref	ref
	2	1.17 (0.93, 1.46)	1.15 (0.89, 1.48)	1.24 (0.79, 1.97)
	3	1.17 (0.93, 1.48)	1.00 (0.77, 1.29)	1.66 (1.05, 2.63)
	4	1.27 (1.01, 1.61)	1.18 (0.91, 1.53)	1.57 (0.99, 2.49)
	5	1.42 (1.10, 1.84)	1.29 (0.97, 1.71)	1.81 (1.13, 2.92)
O_3	1	ref	ref	ref
	2	0.92 (0.78, 1.10)	0.91 (0.74, 1.12)	0.94 (0.62, 1.43)
	3	0.95 (0.80, 1.14)	0.98 (0.80, 1.21)	0.88 (0.58, 1.33)
	4	0.71 (0.59, 0.85)	0.68 (0.55, 0.85)	0.76 (0.50, 1.17)
	5	0.66 (0.55, 0.79)	0.65 (0.52, 0.81)	0.66 (0.43, 1.02)
CO	1	ref	ref	ref
	2	1.04 (0.86, 1.24)	1.02 (0.82, 1.27)	1.08 (0.70, 1.66)
	3	1.19 (0.98, 1.45)	1.25 (0.99, 1.58)	1.10 (0.71, 1.70)
	4	1.33 (1.06, 1.68)	1.36 (1.05, 1.77)	1.30 (0.83, 2.05)
	5	1.50 (1.15, 1.95)	1.68 (1.26, 2.24)	1.19 (0.74, 1.92)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

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CHAPTER 5

DISCUSSION

Rationale

Ambient air pollution is a major cause for health concern as exposure to high levels result in morbidity and mortality throughout the world. Conditions resulting from high concentrations of ambient air pollutants range from minor irritations like coughing, to life threatening conditions such as asthma, birth defects, and cardiovascular disease (U.S. EPA 2010). Well documented cases even exist in which air pollution is associated with lowered immune response and increase risk for adverse health outcomes, including asthma, allergies, and respiratory viruses. This progression indicates the potential for this relationship to exist in other health conditions, even infectious diseases. Recent existing literature has resulted in speculation over whether air pollution, by inhibiting certain immune processes, may result in an increased risk of tuberculosis (TB).

Approach

This is the first large population-based analytic epidemiologic study to evaluate the hypothesis that ambient levels of air pollution are associated with pulmonary TB (PTB). To

evaluate the hypothesis in a U.S. population, estimates of air pollution concentrations obtained from the California Air Resources Board were used in combination with data from the Kaiser Permanente Northern California (KPNC) on PTB and cigarette smoking. Using a case-control study design nested within members of KPNC, electronic clinical databases (ECD) were used to identify cases diagnosed with PTB, from 1996-2010, and controls free of TB, matched 2:1 by age, gender, and race/ethnicity. Cases and controls were linked, by geocoded residential address, to monthly estimates of criteria air pollutants at the closest available monitor. Individual level air pollution estimates for each of the 24 months prior to diagnosis/entry into study, were averaged to define a cumulative estimate indicative of exposure during the period where risk of conversion to active TB is highest. Smoking and other TB risk factors, housed within ECD, were used to explore confounding/effect modification in the analysis. Data on smoking status were inconsistently recorded over the study period; therefore a derived smoking variable was produced as a combination of the various forms of smoking data present through the years of the study period. Logistic regression analyses were used to estimate the odds ratio and 95% confidence interval association between PTB and smoking as well as the association between PTB and each estimated individual-level air pollutant concentration in single and multi-pollutant models.

Findings

Cigarette Smoking and PTB

This is the first study to formally evaluate whether cigarette smoking is associated with active PTB disease in a population-based sample that is generalizable to the American

population. Results from this study show increased risk of TB for ever smokers (OR=1.25 (1.00, 1.55)), corroborating smoking as a risk factor for TB in U.S. The smoking-PTB association observed is biologically plausible since nicotine present in cigarette smoke is known to prevent the production of TB confining macrophages in the lungs (Davies 2006). In this study, the association between smoking and PTB risk also increased with packs per day (dose) and years of smoking (duration). It is not surprising that smoking is substantiated by our study findings as numerous studies in other countries have observed similar results, including the existence of a dose-response pattern in relation to TB risk (Lin 2007).

Statistical interaction was observed between the percent of foreign born individuals within a census block and smoking status with ever smoking being associated with those living in a census block with up to 15% foreign born individuals having an odds ratio of 1.13 (95% CI: 0.82, 1.56) compared to an odds ratio of 1.46 (95% CI: 1.07, 1.98) for those living in a census block with 15% and more foreign born individuals. These results are not entirely surprising as certain immigrant populations are born in countries with a high burden of active TB. Persons residing in census blocks with higher percentages of foreign born individuals are also expected to spend more time traveling back to their place of birth, or to other localities having a high burden of TB, than those that reside in census blocks with a relatively low percentage of foreign born individuals. Thus residence in such an area with a high immigrant population would most likely increase the risk of exposure to *M. tuberculosis* as residents are more likely to be foreign and therefore have been exposed in the past, as well as being more likely to be exposed while living in this area and traveling to countries with high rates of TB.

Unfortunately, smoking data are often not requested or recorded in present surveillance systems within the U.S. (CDC 2011). Inclusion of such information would

foster research to identify effective strategies to reduce the risk of developing active PTB disease among Americans. This awareness of the smoking-TB relationship is imperative to those that remain in close contact to others with TB and necessary to inform intervention programs aimed at reducing TB transmission. Knowledge that smoking is a risk factor for TB in the U.S., in addition to other countries, underscores the importance for smoking cessation programs.

Air Pollution and PTB

This investigation aimed to estimate the magnitude of the association between long-term, 24-month exposure to air pollution active PTB in a U.S. population. This study demonstrated a 50% increase in the risk of PTB for those exposed to the highest concentrations of CO compared to those with the lowest. Stratified analysis showed similar effects among never smokers and ever smokers, suggesting that a saturation of CO exposure may occur in ever smokers preventing any additional impact from air pollution from further increasing the risk of PTB in this group of individuals. The associations seen in this study between long-term exposure to ambient CO and elevated risks of PTB supports findings from ecologic studies, as well as, animal experiments (Saito 2002a, Saito 2002b, Shilova 2004, Hiramatsu 2005, Smith 2013). This study also substantiates historical statistics that show the consumption of coal, a large source of CO exposure in countries outside the U.S., follow the same trends as rates of TB in the U.S., Canada, and China (Tremblay 2007). Considering the goal to eliminate TB in the U.S., an increased risk of TB stemming from air pollution is troublesome, particularly in U.S. metropolitan areas, where on-road vehicle exhaust accounts for up to 75% of all CO emissions (U.S. EPA 2010). Moreover, given the high air pollution

levels and TB rates internationally, this potential association has the ability to impact a millions of people worldwide, putting those susceptible to TB at higher risk for activation of the disease.

NO_2, which also resulted in an increased risk of PTB among smokers, is a gaseous pollutant produced primarily from combustion sources, such as motor vehicle exhaust, and electric generating units. Individuals living near busy roads are particularly vulnerable to NO_2 pollution and related health effects. Though individual exposure depends predominantly on local outdoor concentrations, indoor pollution such as smoking and using gas appliances can alter exposure levels (U.S. EPA 2008).

Several gases including nitric oxide (NO), oxygen (O_2), and CO are critical in defense against TB. These gases form a hypoxic environment within a granuloma to prevent the replication of TB and contain the infection. However previous studies have reported unusual differences in concentrations of O_2, NO, and CO in organs, tissues, and cells models (Sherman 2001, Ohno 2003, Voskuil 2008) causing investigators to hypothesize that while optimal levels of NO and CO keep TB bacteria inactive, extreme amounts of CO, and also NO, could result in abnormal immune responses (Kumar 2008). *The* ability to alter gene expression and bypass CO toxicity in response to varying levels of CO has recently been demonstrated in *M. tuberculosis* (Zacharia 2012), further supporting the hypothesis that increased levels of CO and NO_2 may actually increase risk for the development of active TB.

In this study, an inverse association was seen with O_3 and PTB, with all exposures above the lowest quintile resulting in decreases in PTB risk for never smokers. Although this seems counterintuitive, for years UV light, of which O_3 is a byproduct, has been used to kill TB bacteria (Escombe 2009). An experimental study exploring this association found that

exposure to increased, but non-toxic levels of O_3 (similar to the levels observed in our population) resulted in reduced numbers of TB bacilli (Belianin 2004).

No consistent associations were observed among any of the other pollutants considered in our study, including $PM_{2.5}$, PM_{10} and SO_2. Any modest trends observed between air pollution and PTB were stronger for participants that were ever smokers compared to never smokers. Though potential did exist for a synergistic effect to occur, the attenuation of the apparent associations between the other criteria air pollutants and PTB risk upon stratification by smoking status is not unexpected given that inhalation of tobacco smoke has been shown to elicit similar responses in the lung as air pollution (Holt 1977, Xu 1998).

Overall, results from this study are suggestive of a relationship between long-term, individual level estimates air pollution and risk of TB and underscores the need to confirm our findings regarding the impact that this modifiable ubiquitous exposure may have on TB risk.

Limitations

As in all epidemiologic studies, interpretation of the study results will need to take into consideration some concerns regarding the proposed study approach. Study drawbacks are primarily centered on the proposed exposure assessment methods, which is a common feature of population-based environmental health studies, particularly those that are well-powered.

The core limitation in this study is the inability to assess personal exposure to air pollutants. In this study, ambient air pollution monitors were used to assign estimates of individual-level average exposures. This approach of assigning pollutant concentrations at the closest monitor, while frequently utilized in epidemiologic studies on air pollution (Brauer 2008, Brunekreef 2009, Lipsett 2011), may have resulted in exposure misclassification. No data were available on numerous factors that may have altered personal exposure levels of ambient pollutants including indoor exposures, time spent indoors, residential ventilation systems, time spent at work or away from home, time spent traveling/commuting outside of their residential area and levels of physical activity. Because exposure concentrations are estimated blind to case-control status, this type of exposure misclassification is more likely to lead to underestimation of the effect estimate, rather than to identification of spurious effects (Shy 1997).

This study initially planned to use polynomial distributed lag models to identify a possible critical time window of exposure in the relationship between air pollution and PTB; however the ability to make this assessment was limited by nature of the data available. While individual-level exposure estimates varied over time, the ranking of individuals (required due to the non-linearity of pollutant data) did not change substantially over the 0-24 month lag period assessed. This analysis instead uses the 24-month averages of pollutant concentrations based on previous research suggesting most cases of PTB disease occur within two years of initial exposure to *Mycobacterium tuberculosis* (WHO 2010).

The measurement of smoking status in this study may also have resulted in exposure misclassification. Smoking status was obtained for each study participant using electronic clinical data. The questions used to ascertain information on smoking status from KPNC

members varied over the study period so smoking status was recorded in several different forms in the KPNC electronic clinical database. To address this issue, a variable was constructed from all other smoking information recorded in the KPNC electronic clinical database; if smoking was indicated anywhere in records, the subject was considered an ever smoker, otherwise the subject was considered a never smoker. It is possible that this method of categorizing smokers may have resulted in the misclassification of smoking status. However, the analysis from 1996-2010, using the derived smoking variable, yielded nearly identical results as the analysis from 2008-2010, which contained the most comprehensive smoking data. These results strengthen the case for an association between smoking and PTB in the U.S.

Although the study analysis was able to account for personal history of cigarette smoking, a likely confounder and/or effect modifier of the air pollution-PTB association, assessment of exposure to indoor environmental tobacco smoke (ETS) was not available. However, in the state of California, stringent ETS exposure regulations (CDPH 2009) have minimized the impact of this potential exposure outside the home. Further, whether ETS at levels found commonly in U.S. homes is a risk factor for PTB is currently unknown. Thus, although a lack of information on ETS would be a potentially serious drawback if the proposed study was conducted outside of the U.S., where indoor air pollution from cooking fires is a serious public health problem, the impact of this missing information should be substantially less for a study conducted in the U.S.

Risk factors for PTB, which could have potentially influenced the study results, were recorded in the KPNC records and available through electronic clinical or administrative databases. However, the prevalence of COPD in this study appears to be higher than

expected in the general population, but may better reflect the experience of a cohort in a prepaid health delivery system. In a study of nontuberculous mycobacteria prevalence in four large health care systems with an electronic medical record in the U.S. approximately 1/3 of cases had COPD (Prevots 2010). A different investigation carried out in the Netherlands determined the 40-year risk of developing COPD for those age 55 and older ranges from 16-25% (van Durme 2009). Likewise, a retrospective longitudinal cohort study conducted in Canada using population-based health administrative data from approximately 13 million individuals found that over a lifetime there was a 1 in 4 risk of being diagnosed with COPD (Gershon 2011). So, the rates of COPD found within this study of slightly older adults do not appear to be unrealistic.

Strengths

There were also several advantages to this large population-based study. This study uses data from the KPNC membership, which was optimal for our study approach for several reasons, as summarized below.

The large number of study participants arising from KPNC population resulted in sufficient study power to detect an association between air pollution and PTB. Access to this study population also facilitated conducting this analysis as a case-control study nested within a cohort of KPNC members, which greatly enhances clear interpretation of the study findings. The uniform access to health care through KPNC healthcare minimizes the potential for selection bias through self-selection into the study population. Furthermore, all adult cases of PTB, among KPNC member of two or more years, were included in the study.

The KPNC membership serves 3.2 million in the surrounding geographic area. Cases and controls identified for this study are very similar to residents in the surrounding area, and are thus representative of the population at large, with the following exceptions: a lower percentage of White non-Hispanics, individuals in the income extremes, and smokers (Gordon 2006). The use of this study population enables these results to be characterized to a much larger population.

One of the reasons KPNC, an integrated healthcare delivery system, was chosen as a base for the study population was its unique electronic clinical database, which houses health information on all of its members. This includes records of patient data linked to pharmacy prescriptions, laboratory results, HIV and diabetes registries, pathology reports, and more. The availability of this detailed clinical data, allowed us to explore numerous risk factors associated with TB including COPD, alcohol abuse, diabetes, renal dialysis and immunosuppressive conditions including HIV (CDC 2011, Liu 2009). Using this ECD available from KPNC permitted an objective assessment of TB and confounders, with only a minimal possibility of recall bias.

Lastly, the availability of the criteria air pollution data from the California Air Resources Board, which is based on a tightly placed surveillance monitors, has optimized this proposed approach and improved the ability to detect modest effect estimates. These data permitted us to construct ambient exposure concentrations for all KPNC study participants for the 24-month period prior to a diagnosis of PTB, which is considered the critical window between exposure to mycobacteria and development of active PTB (WHO 2010). Using exposure concentrations from the closest monitor to assign exposure concentrations to cases and controls is a widely accepted technique, previously used and validated (Kiinziil 1997).

This method of exposure assessment is a vast improvement from previously conducted ecologic studies.

Future directions

While these results are biologically plausible and supported by both ecological and experimental studies, additional epidemiologic investigations are needed to confirm the association between air pollution and TB observed in this study. The air pollution-TB relationship should be explored in places similar to the U.S., with lower rates of TB and lower air pollution levels, but using a larger sample size, as well as in countries outside the U.S. that have higher rates of TB and increased exposure to air pollution. Studies with improved methods have the ability to add our knowledge of air pollution as a TB risk factor and may even be able to shed light on whether some of the unexpected findings that occurred in this study, such as an inverse O_3-PTB, actually exist or are spurious. Future studies that have the ability to assess the impact of personal pollutant exposures, both indoor and outdoor, as well as personal smoking habits and environmental tobacco smoke, can lead to a better understanding of which exposures are primary in driving the association with TB and any interactions that may occur among them. Additional matters to consider in the association between air pollution and TB include place of birth, seasonal variability, and critical windows of pollutant exposure. Continuation of this research is imperative as TB affects millions each year and additional studies can result in improved understanding of effects associated with TB risk.

Conclusions

In summary, this study showed an increased risk of PTB among smokers and among individuals exposed to high concentrations of CO. Risk associated with each of these exposures followed a dose-response pattern. Results from this investigation highlight the importance of understanding risk factors for TB and have the potential for far reaching public health implications. Considering the persistent exposure to both smoking and AP in regions where TB poses a major health risk, it is essential to delineate the role of these environmental factors in the etiology and epidemiology of TB.

APPENDIX

Figure A1 Directed acyclic graph for the association between air pollution and pulmonary tuberculosis (PTB).

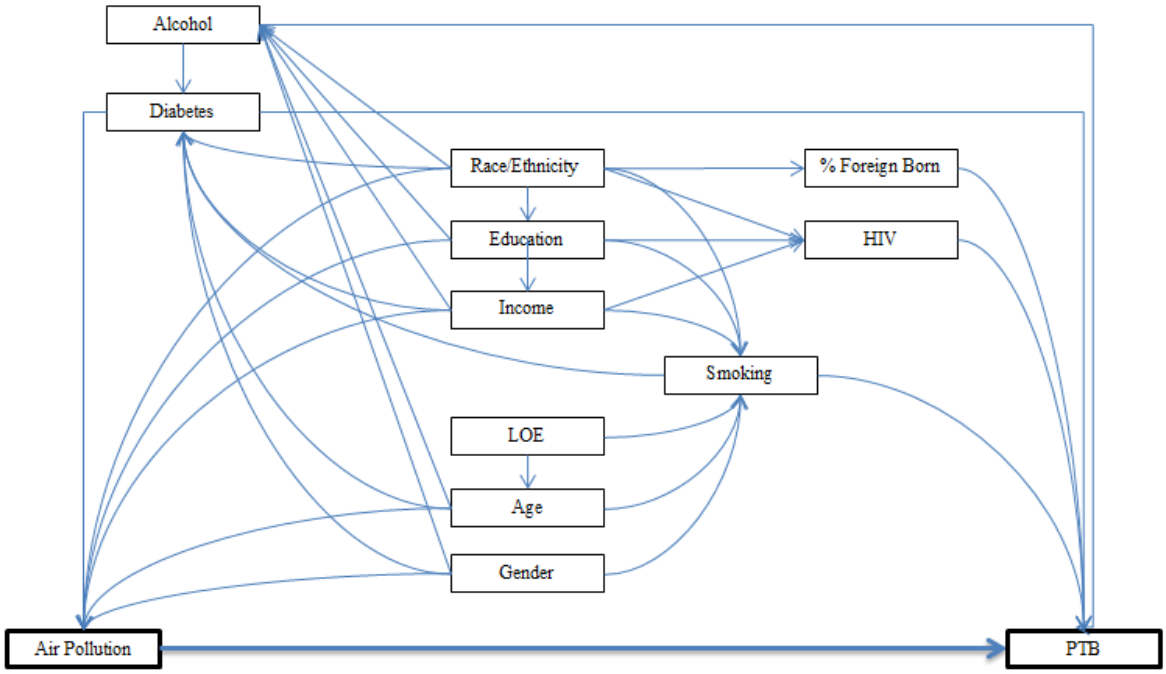


Figure A2 Directed acyclic graph for the association between smoking status and pulmonary tuberculosis (PTB).

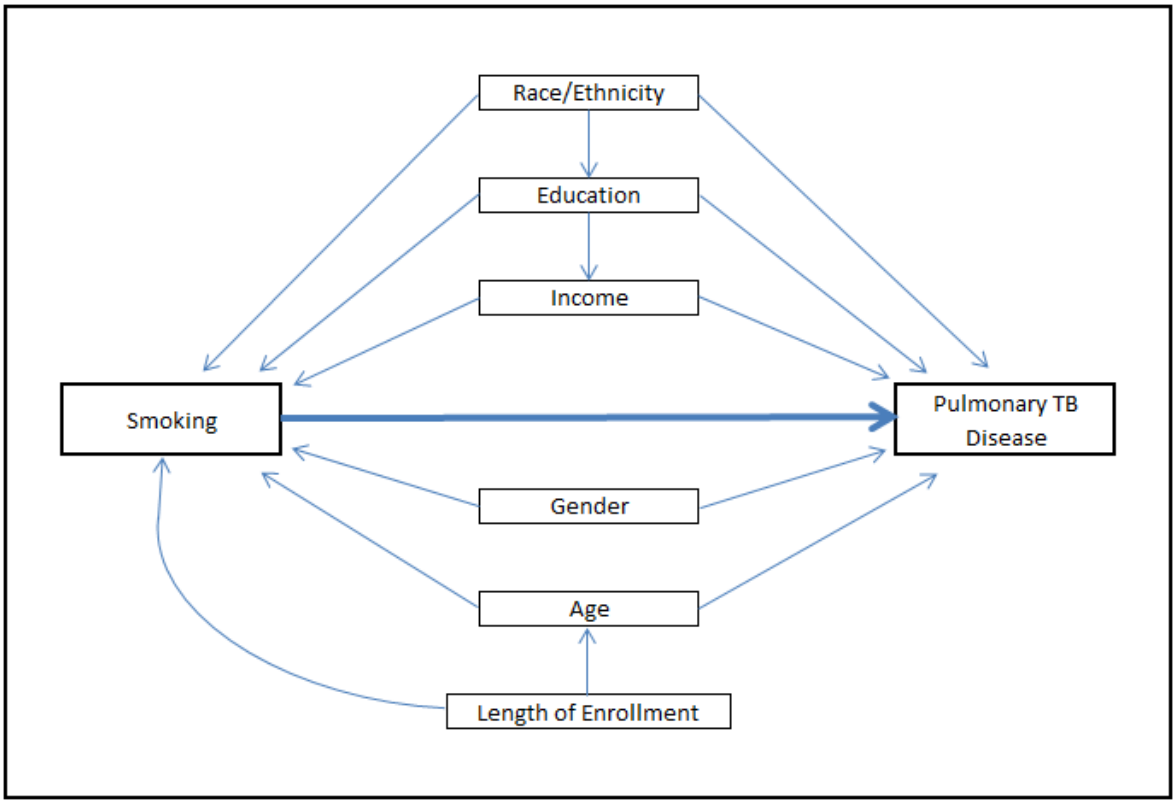


Table A1 Number and percent of study participants with "Tobacco" smoking variable data available over the entire study period (1996-2010), KPNC.

Year	Numbers of Participants with data on "Tobacco"	Percent of Participants with data on "Tobacco"
1996	5	0.6
1997	11	2.8
1998	14	3.6
1999	16	5.7
2000	14	5.0
2001	26	8.7
2002	63	16.2
2003	61	16.4
2004	80	18.6
2005	72	19.1
2006	159	43.4
2007	385	70.5
2008	559	89.4
2009	666	94.3
2010	845	97.7
Total	2976	41.8

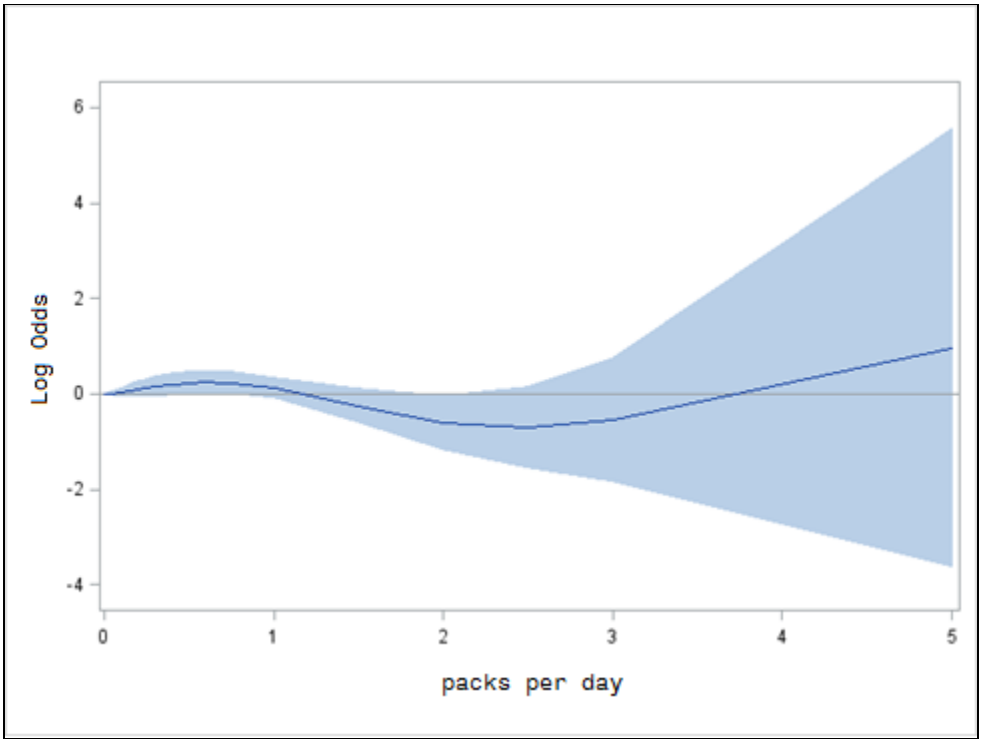
Table A2 Distribution of Smoking Exposure for initial and derived variables for cases diagnosed and controls selected between 1996-2010 and 2008-2010, KPNC.

	1996-2010		2008-2010	
	Cases (n=2380)	**Controls (n=4738)**	**Cases (n=734)**	**Controls (n=1462)**
Derived Smoking				
Never	1264 (62.6)	2742 (67.5)	484 (66.0)	998 (70.4)
Ever	756 (37.4)	1320 (32.5)	249 (34.0)	420 (29.6)
Past	406 (20.1)	674 (16.6)	166 (22.7)	271 (19.1)
Current	350 (17.3)	646 (15.9)	83 (11.3)	149 (10.5)
	Missing = 1036		Missing = 45	
"Tobacco" Smoking				
Never	629 (55.6)	1165 (61.6)	487 (67.1)	961 (70.7)
Ever	503 (44.4)	725 (38.4)	239 (32.9)	398 (29.3)
Past	258 (22.8)	343 (18.2)	160 (22.0)	255 (18.8)
Current	245 (21.6)	382 (20.2)	79 (10.9)	143 (10.5)
	Missing = 4096		Missing = 111	
"Tobacco" Years of Smoking				
Never	629 (83.5)	1165 (86.9)	487 (82.4)	961 (86.6)
less than 10 years	18 (2.4)	41 (3.1)	15 (2.5)	36 (3.2)
10+ years	106 (14.1)	135 (10.1)	89 (15.1)	113 (10.2)
	Missing = 5024		Missing = 495	
"Tobacco" Packs per Day				
Never	629 (83.6)	1165 (87.8)	487 (82.4)	961 (87.5)
less than 1 pack	109 (14.6)	140 (10.6)	92 (15.6)	118 (10.7)
1+ packs	14 (1.9)	22 (1.7)	12 (2.0)	19 (1.7)
	Missing = 5039		Missing = 507	

Table A3 Missing data on smoking dose (packs per day) and duration (years of smoking) among cases and matched controls within the KPNC cohort, 2008-2010.

	Cases n=734	**Controls n=1462**	**Total n=2196**
"Tobacco" Packs per Day			
Never Smoker	487 (67.1%)	961(70.7%)	1448(69.4%)
Ever Smoker with packs per day	104 (14.3%)	137 (10.1%)	241 (11.6%)
Ever Smoker missing packs per day	135 (18.6%)	261 (19.2%)	396 (19.0%)
Past Smoker	95 (70.4%)	182 (69.7%)	277 (69.9%)
Current Smoker	40 (29.6%)	79 (30.3%)	119 (30.1%)
"Tobacco" Years of Smoking			
Never Smoker	487 (67.1%)	961 (70.7%)	1448
Ever Smoker with years of smoking	104 (14.3%)	149 (11.0%)	253 (12.1%)
Ever Smoker missing years of smoking	135 (18.6%)	249 (18.3%)	384 (18.4%)
Past Smoker	95 (70.4%)	164 (65.9%)	259 (67.4%)
Current Smoker	40 (29.6%)	85 (34.1%)	125 (32.6%)

Figure A3 Spline plots of association between (a) packs of cigarettes smoked per day and PTB (b) years of smoking and PTB.



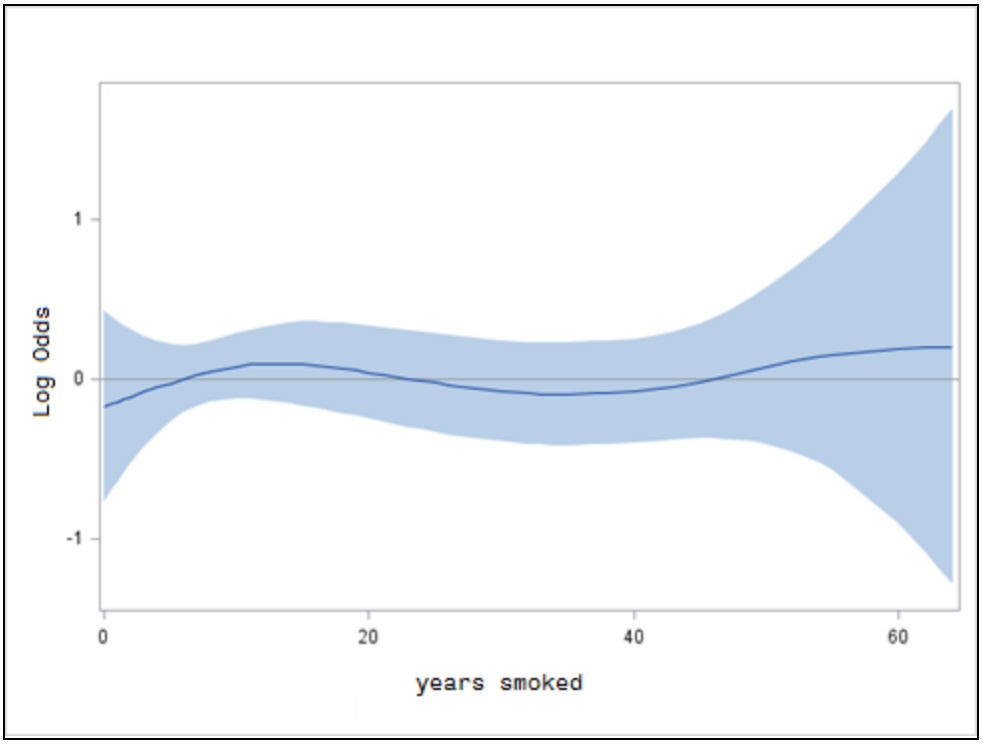


Table A4 Development of cut points for packs per day smoking variable for the association between air pollutants and PTB.

Packs Per Day	N	OR
0.04	1	Numbers too small to estimate
0.1	9	
0.2	12	
0.25	4	
0.3	3	
0.4	3	
0.5	91	
0.6	1	
0.7	9	
0.75	1	
1	76	
1.5	15	
2	11	
2.5	1	
3	3	
5	1	
>0 - <0.25	22	0.94 (0.38, 2.33)
0.25 - <0.5	10	2.59 (0.68, 9.85)
0.5	91	1.59 (1.00, 2.51)
>0.5 - 0.75	11	4.94 (1.26, 19.39)
1	76	1.67 (1.02, 2.74)
1.5	15	1.79 (0.61, 5.28)
>1.5	16	1.07 (0.36, 3.17)
>0 - <0.5	32	1.28 (0.61, 2.68)
0.5	91	1.58 (1.00, 2.5)
>0.5 - 0.75	11	4.94 (1.26, 19.39)
1	76	1.67 (1.02, 2.74)
>1	31	1.37 (0.63, 2.97)
>0 - 0.5	123	1.49 (1.00, 2.23)
>0.5 - 0.75	11	4.94 (1.26, 19.38)
>=1	107	1.61 (1.04, 2.48)
≤0.5	123	1.47 (0.97, 2.21)
>0.5	118	1.78 (1.18, 2.69)

Table A5 Development of cut points for years of smoking variable for the association between air pollutants and PTB.

Years of Smoking	N	OR
0-4	24	Numbers too small to estimate
5	22	
6-9	5	
10	28	
11-14	9	
15	21	
16-19	4	
20	38	
21-24	6	
25	12	
26-29	4	
30	29	
31-34	6	
35	4	
36-39	2	
40	16	
41-44	2	
45	8	
46-49	4	
50	3	
>50	6	
>0 – 4	24	1.1 (0.48, 2.54)
5	22	0.85 (0.29, 2.43)
6-10	33	1.43 (0.71, 2.87)
11-15	30	1.37 (0.63, 2.99)
16-20	42	1.23 (0.62, 2.41)
21-25	18	2.8 (1.08, 7.24)
26-30	33	1.34 (0.61, 2.93)
31-35	10	1.8 (0.47, 6.89)
36-40	18	2.13 (0.8, 5.65)
41-45	10	0.57 (0.12, 2.73)
>45	13	6.09 (1.86, 20.01)
>0 - 5	46	0.95 (0.47, 1.84)
6-15	63	1.48 (0.86, 2.54)
16-30	92	1.52 (0.96, 2.42)
31+	52	2.16 (1.2, 3.89)
>0 - 5	46	0.95 (0.47, 1.84)
6-30	155	1.50 (1.04, 2.17)
31+	52	2.16 (1.19, 3.90)

Table A6 Unconditional logistic regression estimated adjusted* odds ratios (OR) and 95% confidence intervals (CI) for the association between "Derived" smoking status and pulmonary tuberculosis disease, among cases diagnosed and controls selected between 1996-2010, KPNC

(a) Missing data

	Cases n=2020	**Controls n=4062**	**OR***	**95% CI**	
"Derived" Smoking Status					
Never	1264	2742	ref		
Ever	756	1320	1.35	1.19	1.53
Past	406	674	1.43	1.23	1.67
Current	350	646	1.26	1.08	1.48

*Adjusted for age, gender, and race/ethnicity

(a) No missing data

	Cases n=2380	**Controls n=4738**	**OR***	**95% CI**	
Smoking Status					
Never	1624	3418	ref		
Ever	756	1320	1.24	1.10	1.39
Past	406	674	1.31	1.13	1.52
Current	350	646	1.17	1.00	1.37

*Adjusted for age, gender, and race/ethnicity

Table A7 Conditional logistic regression estimated adjusted* odds ratios (OR) and 95% confidence intervals (CI) for the associations between "Tobacco" smoking status, duration and dose, and pulmonary tuberculosis disease, among cases diagnosed and controls selected between 2008-2010, KPNC

	Cases n=734	**Controls n=1462**	**OR***	**95% CI**	
"Tobacco" Smoking Status					
Never	487	961	ref		
Ever	239	398	1.27	1.02	1.59
Past	160	255	1.34	1.04	1.74
Current	79	143	1.15	0.82	1.61
"Tobacco" Years of Smoking					
Never Smoker	487	961	ref		
≤ 5 years	15	31	0.91	0.43	1.91
6-30 years	64	91	1.34	0.89	2.00
> 30 years	25	27	2.35	1.19	4.63
"Tobacco" Packs per Day					
Never Smoker	487	961	ref		
≤ ½ pack	51	72	1.30	0.83	2.04
> ½ pack	53	65	1.81	1.14	2.90

Table A8 Conditional logistic regression estimated adjusted* odds ratios (OR) and 95% confidence intervals (CI) for the association between "Derived" smoking status and pulmonary tuberculosis disease, among cases diagnosed and controls selected between 1996-2010, KPNC

	Cases n=2380	**Controls n=4738**	**OR***	**95% CI**	
"Derived" Smoking Status					
Never	1624	3418	ref		
Ever	756	1320	1.34	1.07	1.66
Past	406	674	1.37	1.07	1.75
Current	350	646	1.26	0.91	1.75

Supplement A1 Polynomial distributed lag analysis of association between air pollution and PTB.

Lag models assume that the effect of an input variable X on an output Y is distributed over time. If you change the value of X at time *t*, Y will experience some immediate effect at time *t*, and it will also experience a delayed effect. In polynomial distributed lag models betas are assumed to lie on a polynomial curve.

$$y = \alpha + \beta_0 x_t + \beta_1 x_{t-1} + \beta_2 x_{t-2} + \beta_3 x_{t-3} + \beta_4 x_{t-4} + \beta_5 x_{t-5} + \beta_6 x_{t-6}$$

$$\text{where } \beta_i = f(i) = \gamma_0 + i\gamma_1 + i^2\gamma_2 + i^3\gamma_3 + \ldots + i^r\gamma_r$$

A logistic regression model with a polynomial distributed lag of 6, with 5 degrees used to compare the results to estimates found using logistic regression at lag 0:

$$\log(\text{odds}) = \alpha + \beta_0 x_t + \beta_1 x_{t-1} + \beta_2 x_{t-2} + \beta_3 x_{t-3} + \beta_4 x_{t-4} + \beta_5 x_{t-5} + \beta_6 x_{t-6}$$

where:

$$\beta_0 = f(0) = \gamma_0$$

$$\beta_1 = f(1) = \gamma_0 + \gamma_1 + \gamma_2 + \gamma_3 + \gamma_4 + \gamma_5$$

$$\beta_2 = f(2) = \gamma_0 + 2\gamma_1 + 2^2\gamma_2 + 2^3\gamma_3 + 2^4\gamma_4 + 2^5\gamma_5$$

$$= \gamma_0 + 2\gamma_1 + 4\gamma_2 + 8\gamma_3 + 16\gamma_4 + 32\gamma_5$$

$$\beta_3 = f(3) = \gamma_0 + 3\gamma_1 + 3^2\gamma_2 + 3^3\gamma_3 + 3^4\gamma_4 + 3^5\gamma_5$$

$$= \gamma_0 + 3\gamma_1 + 9\gamma_2 + 27\gamma_3 + 81\gamma_4 + 243\gamma_5$$

$$\beta_4 = f(4) = \gamma_0 + 4\gamma_1 + 4^2\gamma_2 + 4^3\gamma_3 + 4^4\gamma_4 + 4^5\gamma_5$$

$$= \gamma_0 + 4\gamma_1 + 16\gamma_2 + 64\gamma_3 + 256\gamma_4 + 1024\gamma_5$$

$$\beta_5 = f(5) = \gamma_0 + 5\gamma_1 + 5^2\gamma_2 + 5^3\gamma_3 + 5^4\gamma_4 + 5^5\gamma_5$$

$$= \gamma_0 + 5\gamma_1 + 25\gamma_2 + 125\gamma_3 + 625\gamma_4 + 3125\gamma_5$$

$$\beta_6 = f(6) = \gamma_0 + 6\gamma_1 + 6^2\gamma_2 + 6^3\gamma_3 + 6^4\gamma_4 + 6^5\gamma_5$$

$$= \gamma_0 + 6\gamma_1 + 36\gamma_2 + 216\gamma_3 + 1296\gamma_4 + 7776\gamma_5$$

So the PDL equation becomes:

$$\log(\text{odds}) = \alpha + \gamma_0 \overbrace{(x_t + x_{t-1} + x_{t-2} + x_{t-3} + x_{t-4} + x_{t-5} + x_{t-6})}^{z_{t0}}$$
$$+ \gamma_1 \overbrace{(x_{t-1} + 2x_{t-2} + 3x_{t-3} + 4x_{t-4} + 5x_{t-5} + 6x_{t-6})}^{z_{t1}}$$
$$+ \gamma_1 \overbrace{(x_{t-1} + 4x_{t-2} + 9x_{t-3} + 16x_{t-4} + 25x_{t-5} + 36x_{t-6})}^{z_{t2}}$$
$$+ \gamma_1 \overbrace{(x_{t-1} + 8x_{t-2} + 27x_{t-3} + 64x_{t-4} + 125x_{t-5} + 216x_{t-6})}^{z_{t3}}$$
$$+ \gamma_1 \overbrace{(x_{t-1} + 16x_{t-2} + 81x_{t-3} + 256x_{t-4} + 625x_{t-5} + 1296x_{t-6})}^{z_{t4}}$$

$$= \alpha + \gamma_0 z_{t0} + \gamma_1 z_{t1} + \gamma_2 z_{t2} + \gamma_3 z_{t3} + \gamma_4 z_{t4} + \gamma_5 z_{t5}$$

Figure A4 Distribution of mean monthly air pollution for cases and controls in KPNC cohort across 24 months prior (lags 0-24) to PTB diagnosis/entry into study, 1994-2010.

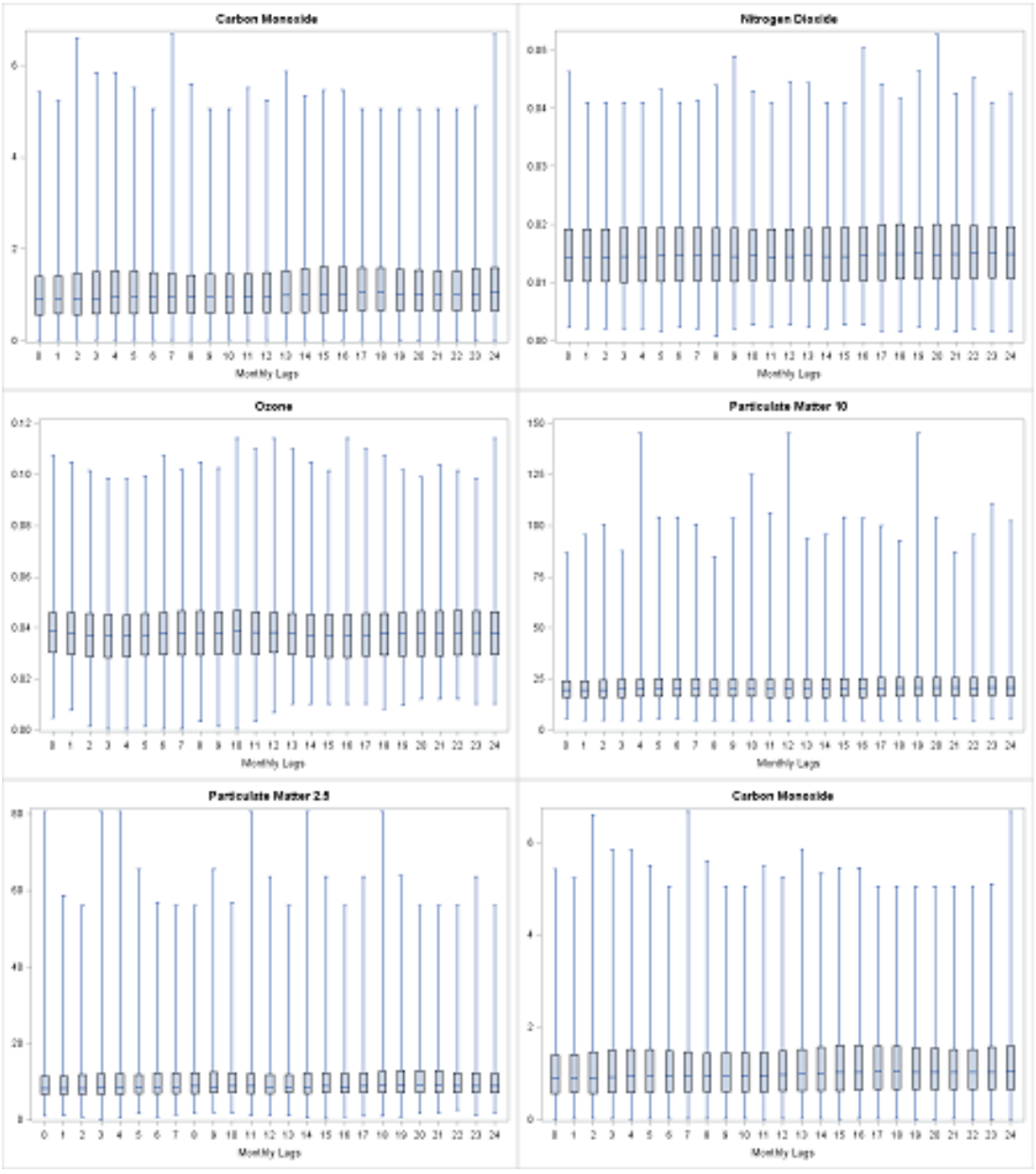


Figure A5. Exploring use of quintiles as a categorization for non-linear air pollution exposure variables: Odds ratios and 95% confidence intervals for the associations of air pollution (lag 6 months) and PTB comparing quintiles of exposure to lowest level quintile (Q1) as a reference group.

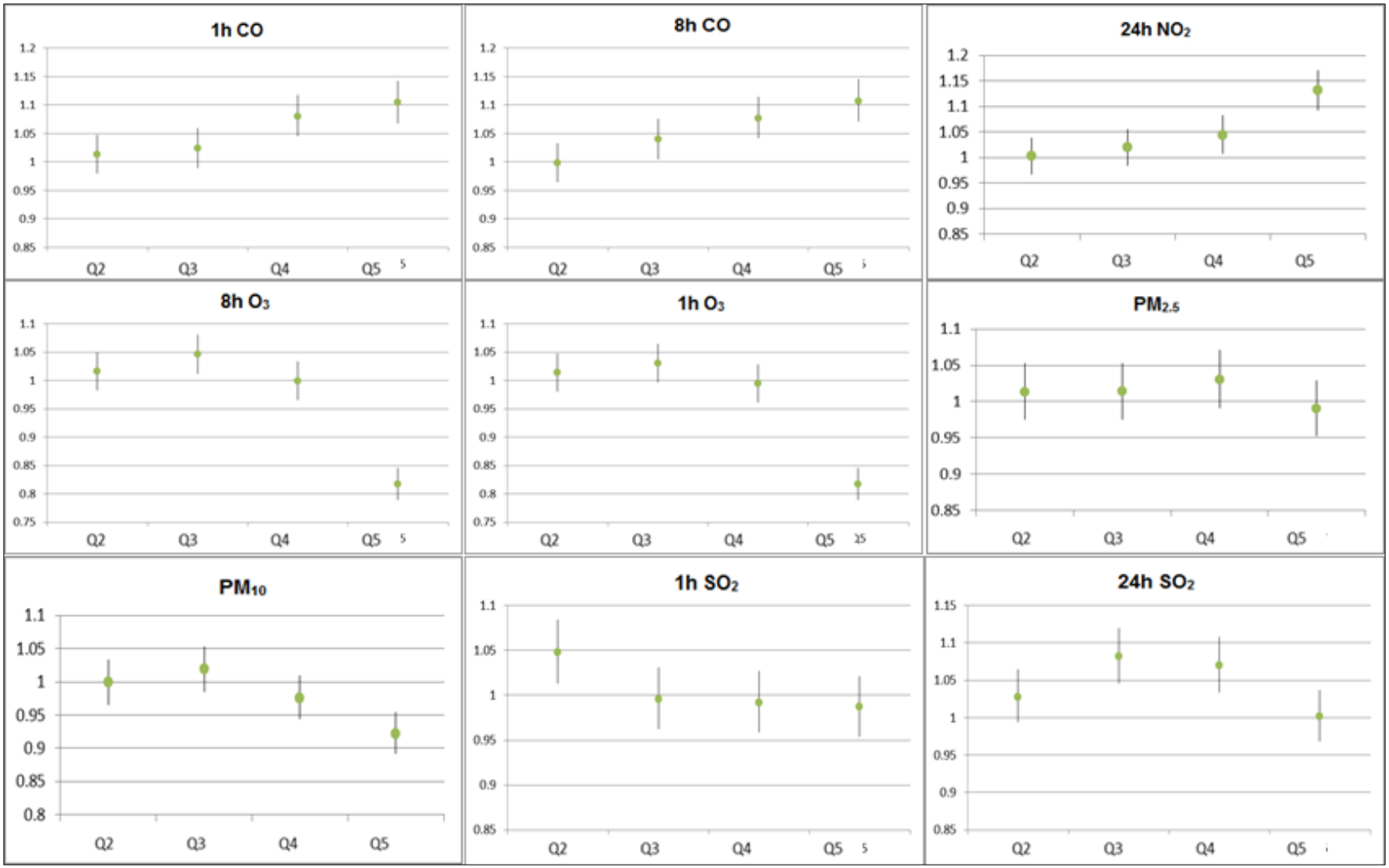


Table A9 Distance (mi.) of residence from closest ambient pollutant monitors for the KPNC cohort, 1996-2010.

Variable	N	Mean	Std. Dev.	Minimum	Maximum
$PM_{2.5}$ ($\mu g/m^3$)	4923	7.63	5.91	0.15	48.70
PM_{10} ($\mu g/m^3$)	6913	6.05	4.24	0.11	35.35
1h SO_2 (ppm)	6591	14.90	10.71	0.14	49.50
24h SO_2 (ppm)	6582	14.93	10.74	0.14	49.50
24h NO_2 (ppm)	5763	6.45	4.78	0.11	45.91
1h O_3 (ppm)	6912	4.66	3.12	0.05	36.74
8h O_3(ppm)	6912	4.66	3.12	0.05	36.74
8h CO (ppm)	6905	6.23	4.70	0.05	43.15

Table A10 Mean and percentile distribution of estimates of average ambient criteria air pollution concentrations cumulated over the 24-month period prior to the pulmonary tuberculosis (PTB) diagnosis/entry into study for cases/matched controls nested within the 1996-2010 KPNC membership with available pollutant monitoring data.

	N (%) with Pollutant Data				Percentile Distribution				
Air Pollutant	Cases	Controls	Mean	Min	20th	40th	60th	80th	Max
24h $PM_{2.5}$ ($\mu g/m^3$)	1842 (79.8)	3661 (79.5)	9.9013	0.1408	8.5600	9.1840	10.3324	11.6602	26.4783
24h PM_{10} ($\mu g/m^3$)	2309 (100.0)	4604 (100.0)	21.7027	9.1000	18.3896	19.8733	21.6487	24.4700	56.6082
1h SO_2 (ppm)	2248 (97.4)	4439 (96.4)	0.0032	0.0001	0.0021	0.0029	0.0034	0.0042	0.0109
24h SO_2 (ppm)	2248 (97.4)	4439 (96.4)	0.0013	0.0001	0.0009	0.0011	0.0013	0.0018	0.0039
24h NO_2 (ppm)	2309 (100.0)	4601 (99.9)	0.0138	0.0003	0.0098	0.0133	0.0151	0.0177	0.0390
1h O_3 (ppm)	2309 (100.0)	4604 (100.0)	0.0391	0.0209	0.0327	0.0360	0.0391	0.0451	0.0750
8h O_3 (ppm)	2309 (100.0)	4604 (100.0)	0.0329	0.0178	0.0279	0.0301	0.0330	0.0378	0.0670
1h CO (ppm)	2309 (100.0)	4598 (99.9)	1.1731	0.1590	0.7361	0.9318	1.2250	1.5599	3.8802
8h CO (ppm)	2309 (100.0)	4598 (99.9)	0.8420	0.0983	0.5481	0.6793	0.8760	1.1114	3.0572

Table A11 Two tailed t-test to compare the means of ambient criteria air pollutant concentrations at the closest monitor to subject residences cumulated over the 24-month period prior to the pulmonary tuberculosis (PTB) diagnosis/entry into study for never and ever smokers nested within the 1996-2010 KPNC membership.

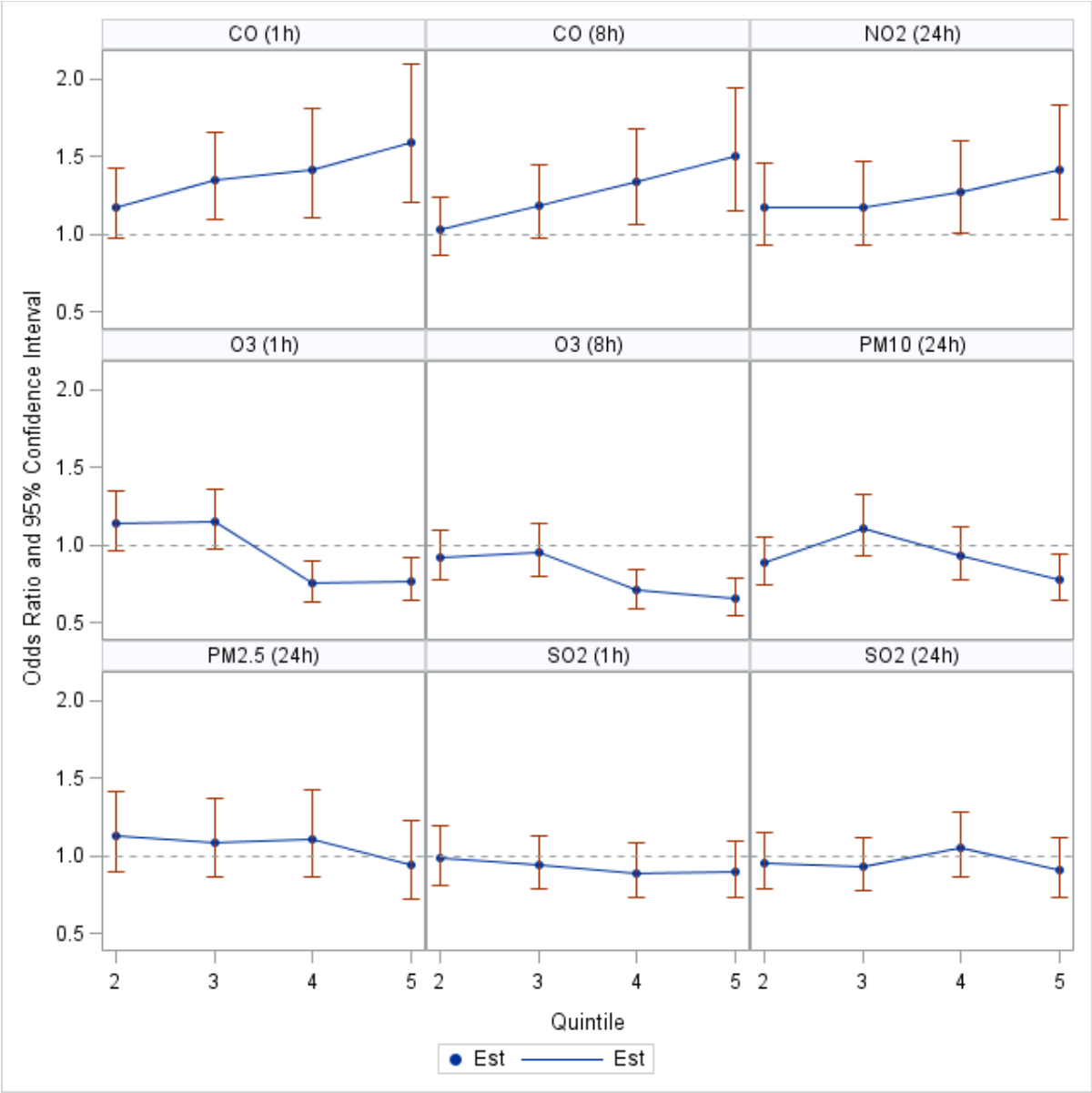
	Mean		
Air Pollutant	**Never**	**Ever**	**t**
24h $PM_{2.5}$ ($\mu g/m^3$)	9.8883	9.9313	0.6062
24h PM_{10} ($\mu g/m^3$)	21.7734	21.5324	0.0626
1h SO_2 (ppm)	0.0032	0.0032	0.6862
24h SO2 (ppm)	0.0013	0.0013	0.709
24h NO_2 (ppm)	0.0138	0.0137	0.621
1h O_3 (ppm)	0.0391	0.0391	0.9576
8h O3(ppm)	0.0329	0.0329	0.7703
1h CO (ppm)	1.1889	1.1352	<.0001
8h CO (ppm)	0.8526	0.8166	<.0001

Table A12 Spearman correlation coefficients[†] for ambient criteria air pollutant concentrations at the closest monitor to subject residences, 0-24 months, among pulmonary tuberculosis (PTB) cases and matched controls nested within the 1995-2010 KPNC membership.

All									
Air Pollutant	$PM_{2.5}$	PM_{10}	1h SO_2	24h SO_2	24h NO_2	1h O_3	8h O_3	1h CO	8h CO
$PM_{2.5}$	1.00	0.61	0.04[†]	0.12	0.28	0.31	0.25	0.33	0.35
PM_{10}		1.00	-0.01[†]	0.09	0.33	0.16	0.09	0.41	0.42
1h SO_2			1.00	0.90	0.04	-0.26	-0.23	0.23	0.20
24h SO_2				1.00	0.19	-0.24	-0.24	0.33	0.30
24h NO_2					1.00	-0.28	-0.33	0.22	0.23
1h O_3						1.00	0.98	-0.16	-0.17
8h O_3							1.00	-0.27	-0.28
1h CO								1.00	0.99
8h CO									1.00
Controls									
Air Pollutant	$PM_{2.5}$	PM_{10}	1h SO_2	24h SO_2	24h NO_2	1h O_3	8h O_3	1h CO	8h CO
$PM_{2.5}$	1.00	0.63	0.04[†]	0.12	0.28	0.32	0.26	0.33	0.34
PM_{10}		1.00	-0.01[†]	0.10	0.30	0.19	0.12	0.41	0.42
1h SO_2			1.00	0.90	0.05	-0.26	-0.24	0.24	0.20
24h SO_2				1.00	0.20	-0.24	-0.24	0.34	0.30
24h NO_2					1.00	-0.29	-0.34	0.23	0.23
1h O_3						1.00	0.99	-0.17	-0.18
8h O_3							1.00	-0.27	-0.28
1h CO								1.00	0.99
8h CO									1.00
Cases									
Air Pollutant	$PM_{2.5}$	PM_{10}	1h SO_2	24h SO_2	24h NO_2	1h O_3	8h O_3	1h CO	8h CO
$PM_{2.5}$	1.00	0.58	0.04[†]	0.12	0.29	0.28	0.23	0.35	0.37
PM_{10}		1.00	-0.01[†]	0.08	0.37	0.11	0.03[†]	0.43	0.45
1h SO_2			1.00	0.91	0.02[†]	-0.26	-0.23	0.22	0.19
24h SO_2				1.00	0.17	-0.24	-0.24	0.32	0.29
24h NO_2					1.00	-0.27	-0.32	0.22	0.22
1h O_3						1.00	0.98	-0.15	-0.16
8h O_3							1.00	-0.28	-0.28
1h CO								1.00	0.99
8h CO									1.00

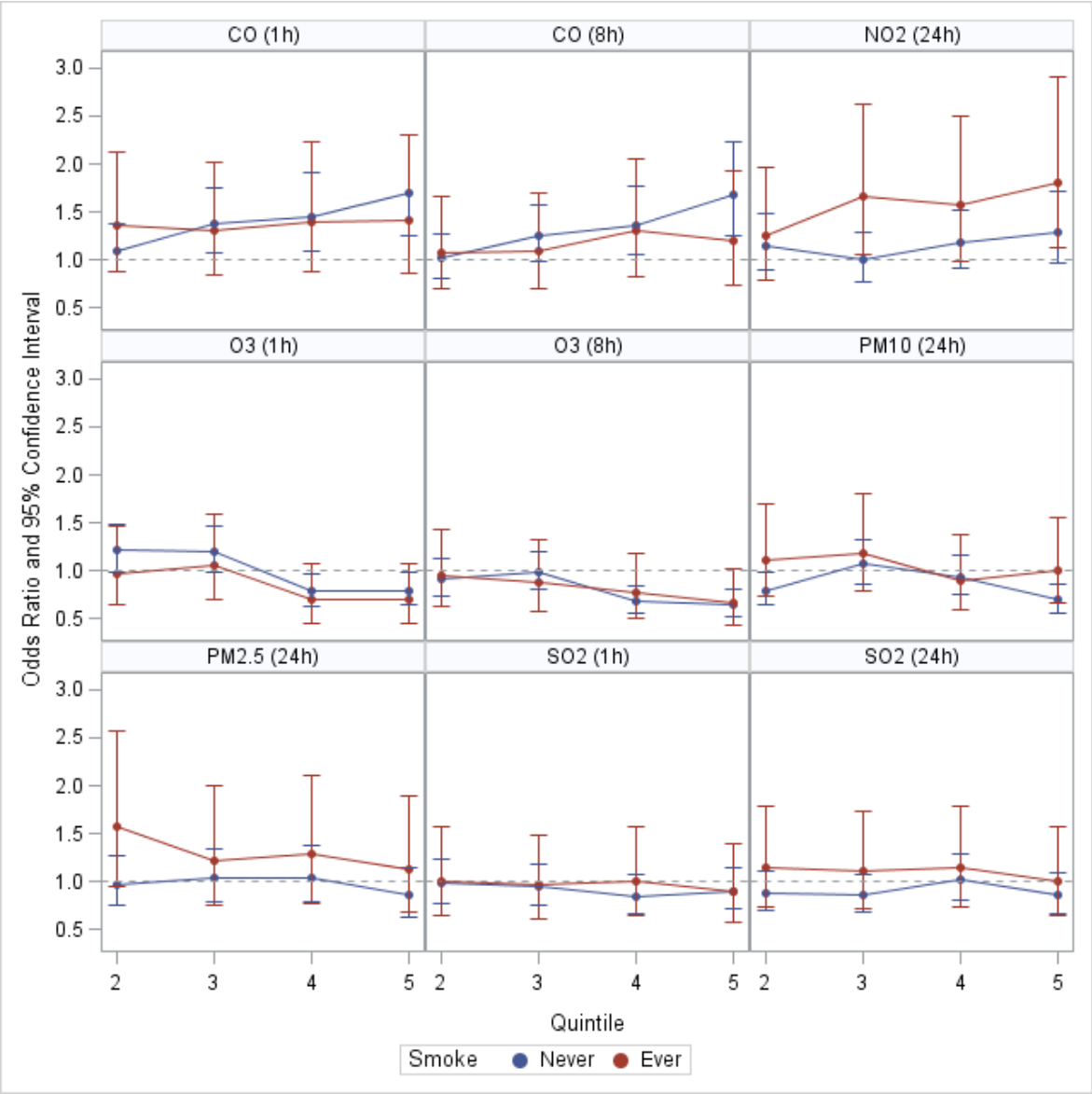
[†] Coefficients not statistically significant ($p<0.05$)

Figure A6 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations within the 24 months prior to diagnosis date, among all cases and matched controls nested within the 1996-2010 KPNC membership.



*Adjusted for the matching factors (age, gender, and race/ethnicity).

Figure A7 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations within the 24 months prior to diagnosis date, among all cases and matched controls nested within the 1996-2010 KPNC membership stratified by smoking status (ever vs. never smokers).



*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A13 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among all cases and matched controls nested within the 1996-2010 KPNC membership, and stratified by smoking status (ever vs. never smokers).

Air Pollutant	Quintile	All	Never Smokers	Ever Smokers
$PM_{2.5}$	1	ref	ref	ref
	2	1.13 (0.90, 1.41)	0.98 (0.75, 1.27)	1.57 (0.95, 2.58)
	3	1.09 (0.87, 1.37)	1.03 (0.79, 1.34)	1.22 (0.75, 2.00)
	4	1.11 (0.87, 1.42)	1.04 (0.78, 1.37)	1.28 (0.78, 2.11)
	5	0.94 (0.73, 1.23)	0.85 (0.64, 1.15)	1.13 (0.68, 1.89)
PM_{10}	1	ref	ref	ref
	2	0.89 (0.74, 1.06)	0.80 (0.65, 0.99)	1.12 (0.73, 1.70)
	3	1.11 (0.93, 1.32)	1.07 (0.87, 1.32)	1.19 (0.78, 1.80)
	4	0.93 (0.77, 1.11)	0.94 (0.76, 1.16)	0.90 (0.59, 1.37)
	5	0.78 (0.65, 0.94)	0.69 (0.55, 0.87)	1.01 (0.66, 1.56)
1h SO_2	1		ref	ref
	2	0.98 (0.81, 1.20)	0.98 (0.78, 1.23)	1.01 (0.65, 1.57)
	3	0.94 (0.79, 1.13)	0.94 (0.76, 1.17)	0.96 (0.62, 1.48)
	4	0.89 (0.73, 1.09)	0.84 (0.67, 1.07)	1.01 (0.65, 1.58)
	5	0.89 (0.73, 1.09)	0.90 (0.71, 1.14)	0.89 (0.57, 1.39)
SO_2	1	ref	ref	ref
	2	0.95 (0.79, 1.15)	0.88 (0.70, 1.10)	1.15 (0.74, 1.79)
	3	0.93 (0.78, 1.12)	0.86 (0.69, 1.08)	1.11 (0.72, 1.73)
	4	1.05 (0.86, 1.29)	1.02 (0.80, 1.29)	1.14 (0.73, 1.79)
	5	0.90 (0.73, 1.12)	0.86 (0.67, 1.10)	1.01 (0.64, 1.58)
NO_2	1	ref	ref	ref
	2	1.17 (0.93, 1.46)	1.15 (0.89, 1.48)	1.24 (0.79, 1.97)
	3	1.17 (0.93, 1.48)	1.00 (0.77, 1.29)	1.66 (1.05, 2.63)
	4	1.27 (1.01, 1.61)	1.18 (0.91, 1.53)	1.57 (0.99, 2.49)
	5	1.42 (1.10, 1.84)	1.29 (0.97, 1.71)	1.81 (1.13, 2.92)
O_3	1	ref	ref	ref
	2	1.14 (0.96, 1.35)	1.22 (0.99, 1.49)	0.98 (0.65, 1.47)
	3	1.15 (0.97, 1.37)	1.21 (0.99, 1.48)	1.05 (0.69, 1.59)
	4	0.76 (0.63, 0.90)	0.79 (0.63, 0.97)	0.70 (0.45, 1.07)
	5	0.77 (0.64, 0.91)	0.79 (0.64, 0.98)	0.70 (0.46, 1.08)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A13 continued Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among all cases and matched controls nested within the 1996-2010 KPNC membership, and stratified by smoking status (ever vs. never smokers).

Air Pollutant	Quintile	All	Never Smokers	Ever Smokers
O_3	1	ref	ref	ref
	2	0.92 (0.78, 1.10)	0.91 (0.74, 1.12)	0.94 (0.62, 1.43)
	3	0.95 (0.80, 1.14)	0.98 (0.80, 1.21)	0.88 (0.58, 1.33)
	4	0.71 (0.59, 0.85)	0.68 (0.55, 0.85)	0.76 (0.50, 1.17)
	5	0.66 (0.55, 0.79)	0.65 (0.52, 0.81)	0.66 (0.43, 1.02)
CO	1	ref	ref	ref
	2	1.18 (0.97, 1.42)	1.10 (0.87, 1.38)	1.37 (0.88, 2.13)
	3	1.35 (1.10, 1.66)	1.38 (1.08, 1.75)	1.30 (0.84, 2.02)
	4	1.42 (1.11, 1.82)	1.44 (1.09, 1.90)	1.40 (0.88, 2.24)
	5	1.59 (1.21, 2.10)	1.71 (1.26, 2.31)	1.40 (0.86, 2.30)
CO	1	ref	ref	ref
	2	1.04 (0.86, 1.24)	1.02 (0.82, 1.27)	1.08 (0.70, 1.66)
	3	1.19 (0.98, 1.45)	1.25 (0.99, 1.58)	1.10 (0.71, 1.70)
	4	1.33 (1.06, 1.68)	1.36 (1.05, 1.77)	1.30 (0.83, 2.05)
	5	1.50 (1.15, 1.95)	1.68 (1.26, 2.24)	1.19 (0.74, 1.92)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A14 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among all cases and matched controls nested within the 1996-2010 KPNC cohort.

Pollutant Quintile	Single-Pollutant Model	Multi-Pollutant Model				
		SO_2+PM_{10}	SO_2+PM_{10+} $PM_{2.5}$	SO_2+PM_{10+} $PM_{2.5}$+CO	SO_2+PM_{10+} $PM_{2.5}$+CO+NO_2	SO_2+$PM_{10+}PM_{2.5}$ +CO+NO_2+ O_3
SO_2						
1	ref	ref	ref	ref	ref	ref
2	0.95 (0.79, 1.15)	0.92 (0.76, 1.11)	0.85 (0.69, 1.05)	0.93 (0.75, 1.15)	0.89 (0.75, 1.06)	0.91 (0.74, 1.12)
3	0.93 (0.78, 1.12)	0.92 (0.77, 1.11)	0.96 (0.78, 1.17)	0.89 (0.72, 1.09)	0.87 (0.70, 1.08)	0.89 (0.73,1.09)
4	1.05 (0.86, 1.29)	1.01 (0.82, 1.24)	1.05 (0.83, 1.32)	1.00 (0.79, 1.26)	1.01 (0.79, 1.28)	1.03 (0.84, 1.23)
5	0.90 (0.73, 1.12)	0.87 (0.70, 1.08)	0.88 (0.72, 1.08)	0.86 (0.69, 1.07)	0.89 (0.75, 1.06)	0.91 (0.78, 1.06)
PM_{10}						
1	ref	ref	ref	ref	ref	ref
2	0.89 (0.74, 1.06)	0.89 (0.75, 1.07)	0.87 (0.71, 1.06)	0.82 (0.67, 1.02)	0.83 (0.67, 1.02)	0.81 (0.65, 1.00)
3	1.11 (0.93, 1.32)	1.12 (0.93, 1.35)	1.13 (0.91, 1.39)	1.01 (0.80, 1.28)	1.02 (0.81, 1.29)	1.00 (0.78, 1.28)
4	0.93 (0.77, 1.11)	0.93 (0.77, 1.12)	0.90 (0.71, 1.15)	0.91 (0.75, 1.10)	0.90 (0.74, 1.09)	0.90 (0.72, 1.13)
5	0.78 (0.65, 0.94)	0.80 (0.65, 0.98)	0.86 (0.65, 1.13)	0.72 (0.53, 0.97)	0.69 (0.51, 0.94)	0.73 (0.52, 1.01)
$PM_{2.5}$						
1	ref		ref	ref	ref	ref
2	1.13 (0.90, 1.41)		1.12 (0.88, 1.41)	1.11 (0.88, 1.41)	1.14 (0.90, 1.45)	1.17 (0.92, 1.49)
3	1.09 (0.87, 1.37)		1.04 (0.81, 1.33)	1.04 (0.82, 1.33)	1.03 (0.80, 1.33)	1.04 (0.84, 1.29)
4	1.11 (0.87, 1.42)		1.10 (0.83, 1.47)	1.12 (0.84, 1.5)	1.06 (0.78, 1.44)	1.10 (0.87, 1.39)
5	0.94 (0.73, 1.23)		0.93 (0.66, 1.31)	1.00 (0.70, 1.41)	0.99 (0.71, 1.38)	1.00 (0.83, 1.20)
NO_2						
1	ref			ref	ref	ref
2	1.17 (0.93, 1.46)			1.15 (0.89, 1.49)	1.18 (0.91, 1.53)	1.06 (0.81, 1.39)
3	1.17 (0.93, 1.48)			1.22 (0.93, 1.60)	1.25 (0.94, 1.65)	1.08 (0.80, 1.45)
4	1.27 (1.01, 1.61)			1.30 (0.98, 1.72)	1.29 (0.97, 1.72)	1.09 (0.79, 1.50)
5	1.42 (1.10, 1.84)			1.74 (1.25, 2.40)	1.61 (1.14, 2.25)	1.26 (0.85, 1.86)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A14 continued Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among all cases and matched controls nested within the 1996-2010 KPNC cohort.

Pollutant Quintile	Single-Pollutant Model	Multi-Pollutant Model				
		SO_2+PM_{10}	SO_2+PM_{10+} $PM_{2.5}$	SO_2+PM_{10+} $PM_{2.5}$+CO	SO_2+PM_{10+} $PM_{2.5}$+CO+NO_2	SO_2+$PM_{10+}$$PM_{2.5}$ +CO+NO_2+ O_3
CO						
1	ref				ref	ref
2	1.04 (0.86, 1.24)				0.94 (0.76, 1.17)	0.93 (0.75, 1.16)
3	1.19 (0.98, 1.45)				1.14 (0.90, 1.45)	1.12 (0.88, 1.43)
4	1.33 (1.06, 1.68)				1.32 (0.97, 1.79)	1.22 (0.90, 1.66)
5	1.50 (1.15, 1.95)				1.43 (0.96, 2.13)	1.21 (0.80, 1.83)
O_3						
1	ref					ref
2	0.92 (0.78, 1.10)					0.91 (0.73, 1.14)
3	0.95 (0.80, 1.14)					0.93 (0.74, 1.17)
4	0.71 (0.59, 0.85)					0.65 (0.51, 0.84)
5	0.66 (0.55, 0.79)					0.67 (0.49, 0.91)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A15 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among never smoker cases and matched controls nested within the 1996-2010 KPNC cohort.

Pollutant Quintile	**Single-Pollutant Model**	**Multi-Pollutant Model**				
		SO_2+PM_{10}	SO_2+PM_{10+} $PM_{2.5}$	SO_2+PM_{10+} $PM_{2.5}$+CO	SO_2+PM_{10+} $PM_{2.5}$+CO+NO_2	SO_2+$PM_{10+}PM_{2.5}$ +CO+NO_2+ O_3
SO_2						
1	ref	ref	ref	ref	ref	ref
2	0.98 (0.78, 1.23)	0.89 (0.72, 1.09)	0.87 (0.70, 1.08)	0.95 (0.75, 1.20)	0.92 (0.73, 1.16)	0.93 (0.74, 1.17)
3	0.94 (0.76, 1.17)	0.87 (0.74, 1.02)	0.89 (0.75, 1.07)	0.92 (0.71, 1.19)	0.87 (0.71, 1.06)	0.76 (0.60, 0.96)
4	0.84 (0.67, 1.07)	0.72 (0.57, 0.91)	0.76 (0.60, 0.96)	1.00 (0.79, 1.26)	1.01 (0.79, 1.28)	1.10 (0.83, 1.47)
5	0.90 (0.71, 1.14)	0.88 (0.68, 1.14)	0.74 (0.58, 0.94)	0.89 (0.71, 1.12)	0.89 (0.71, 1.12)	0.78 (0.62, 0.97)
PM_{10}						
1	ref	ref	ref	ref	ref	ref
2	0.80 (0.65, 0.99)	0.84 (0.67, 1.05)	0.86 (0.69, 1.07)	0.82 (0.67, 1.02)	0.83 (0.67, 1.02)	0.81 (0.65, 1.00)
3	1.07 (0.87, 1.32)	1.12 (0.93, 1.35)	1.13 (0.91, 1.39)	1.01 (0.80, 1.28)	1.02 (0.81, 1.29)	1.00 (0.78, 1.28)
4	0.94 (0.76, 1.16)	0.93 (0.77, 1.12)	0.90 (0.71, 1.15)	0.74 (0.56, 0.97)	0.80 (0.65, 0.98)	0.86 (0.65, 1.13)
5	0.69 (0.55, 0.87)	0.71 (0.54, 0.93)	0.71 (0.53, 0.96)	0.72 (0.53, 0.97)	0.69 (0.51, 0.94)	0.73 (0.52, 1.01)
$PM_{2.5}$						
1	ref		ref	ref	ref	ref
2	0.98 (0.75, 1.27)		1.00 (0.80, 1.25)	1.07 (0.85, 1.35)	1.14 (0.90, 1.45)	1.17 (0.92, 1.49)
3	1.03 (0.79, 1.34)		1.05 (0.82, 1.34)	1.03 (0.79, 1.34)	1.03 (0.80, 1.33)	1.09 (0.84, 1.42)
4	1.04 (0.78, 1.37)		1.07 (0.84, 1.36)	1.09 (0.85, 1.40)	1.06 (0.78, 1.44)	1.17 (0.85, 1.62)
5	0.85 (0.64, 1.15)		0.98 (0.79, 1.22)	0.89 (0.77, 1.03)	1.00 (0.70, 1.42)	0.99 (0.80, 1.23)
NO_2						
1	ref			ref	ref	ref
2	1.15 (0.89, 1.48)			1.07 (0.82, 1.40)	1.18 (0.91, 1.53)	1.06 (0.81, 1.39)
3	1.00 (0.77, 1.29)			1.12 (0.87, 1.44)	1.25 (0.94, 1.65)	1.08 (0.80, 1.45)
4	1.18 (0.91, 1.53)			1.20 (0.93, 1.55)	1.29 (0.97, 1.72)	1.09 (0.79, 1.50)
5	1.29 (0.97, 1.71)			1.22 (0.94, 1.58)	1.51 (1.10, 2.03)	1.26 (0.85, 1.86)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A15 continued Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among never smoker cases and matched controls nested within the 1996-2010 KPNC cohort.

Pollutant Quintile	**Single-Pollutant Model**	**Multi-Pollutant Model**				
		SO_2+PM_{10}	SO_2+PM_{10+} $PM_{2.5}$	SO_2+PM_{10+} $PM_{2.5}$+CO	SO_2+PM_{10+} $PM_{2.5}$+CO+NO_2	SO_2+$PM_{10+}$$PM_{2.5}$ +CO+NO_2+ O_3
CO						
1	ref				ref	ref
2	1.02 (0.82, 1.27)				1.09 (0.86, 1.38)	1.15 (0.97, 1.36)
3	1.25 (0.99, 1.58)				1.17 (0.97, 1.41)	1.33 (1.02 , 1.73)
4	1.36 (1.05, 1.77)				1.23 (0.88, 1.72)	1.57 (1.12, 2.20)
5	1.68 (1.26, 2.24)				1.59 (1.21, 2.09)	1.55 (1.23, 1.95)
O_3						
1	ref					ref
2	0.91 (0.74, 1.12)					0.96 (0.57, 1.62)
3	0.98 (0.80, 1.21)					0.76 (0.42, 1.40)
4	0.68 (0.55, 0.85)					0.78 (0.41, 1.45)
5	0.65 (0.52, 0.81)					0.76 (0.39, 1.49)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A16 Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among ever smoker cases and matched controls nested within the 1996-2010 KPNC cohort.

Pollutant Quintile	Single-Pollutant Model	Multi-Pollutant Model				
		SO_2+PM_{10}	SO_2+PM_{10+} $PM_{2.5}$	SO_2+PM_{10+} $PM_{2.5}$+CO	SO_2+PM_{10+} $PM_{2.5}$+CO+NO_2	SO_2+$PM_{10+}$$PM_{2.5}$ +CO+NO_2+ O_3
SO_2						
1	ref	ref	ref	ref	ref	ref
2	1.15 (0.74, 1.79)	1.05 (0.64, 1.73)	1.02 (0.59, 1.75)	1.12 (0.69, 1.82)	1.10 (0.66, 1.83)	1.13 (0.73, 1.75)
3	1.11 (0.72, 1.73)	0.91 (0.58, 1.43)	1.10 (0.67, 1.79)	1.07 (0.64, 1.79)	1.09 (0.66, 1.80)	1.10 (0.68, 1.78)
4	1.14 (0.73, 1.79)	1.07 (0.63, 1.83)	0.83 (0.46, 1.5)	1.04 (0.62, 1.74)	1.08 (0.65, 1.79)	1.10 (0.67, 1.81)
5	1.01 (0.64, 1.58)	1.16 (0.68, 1.99)	1.00 (0.65, 1.54)	0.92 (0.52, 1.63)	0.93 (0.51, 1.71)	0.97 (0.51, 1.81)
PM_{10}						
1	ref	ref	ref	ref	ref	ref
2	1.12 (0.73, 1.70)	1.16 (0.74, 1.84)	1.09 (0.67, 1.80)	0.95 (0.55, 1.64)	0.95 (0.55, 1.65)	0.91 (0.52, 1.59)
3	1.19 (0.78, 1.80)	1.56 (0.96, 2.53)	1.40 (0.79, 2.49)	1.28 (0.68, 2.42)	1.28 (0.68, 2.44)	1.16 (0.58, 2.32)
4	0.90 (0.59, 1.37)	1.07 (0.63, 1.82)	1.18 (0.61, 2.29)	0.92 (0.44, 1.95)	0.94 (0.43, 2.06)	0.86 (0.37, 1.99)
5	1.01 (0.66, 1.56)	1.05 (0.62, 1.78)	1.34 (0.66, 2.71)	1.09 (0.50, 2.39)	1.12 (0.50, 2.52)	1.10 (0.45, 2.69)
$PM_{2.5}$						
1	ref		ref	ref	ref	ref
2	1.57 (0.95, 2.58)		1.54 (0.81, 2.92)	1.77 (0.91, 3.43)	1.72 (0.85, 3.48)	1.72 (0.84, 3.53)
3	1.22 (0.75, 2.00)		1.29 (0.67, 2.50)	1.38 (0.70, 2.69)	1.34 (0.65, 2.77)	1.37 (0.65, 2.89)
4	1.28 (0.78, 2.11)		1.68 (0.76, 3.72)	1.87 (0.82, 4.23)	1.78 (0.73, 4.35)	1.83 (0.73, 4.59)
5	1.13 (0.68, 1.89)		1.07 (0.44, 2.61)	1.35 (0.53, 3.42)	1.29 (0.48, 3.46)	1.35 (0.48, 3.78)
NO_2						
1	ref			ref	ref	ref
2	1.24 (0.79, 1.97)			1.17 (0.52, 2.63)	1.21 (0.58, 2.52)	1.19 (0.57, 2.48)
3	1.66 (1.05, 2.63)			1.39 (0.63, 3.07)	1.44 (0.66, 3.14)	1.35 (0.61, 2.99)
4	1.57 (0.99, 2.49)			1.43 (0.64, 3.20)	1.41 (0.61, 3.24)	1.57 (0.62, 3.99)
5	1.81 (1.13, 2.92)			1.69 (0.83, 3.45)	1.66 (0.80, 3.43)	1.75 (0.82, 3.72)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

Table A16 continued Adjusted* odds ratios (ORs) and 95% confidence intervals (CIs) for the associations of pulmonary tuberculosis (PTB) and quintile increase in ambient criteria air pollutants concentrations, 0-24 months, among ever smoker cases and matched controls nested within the 1996-2010 KPNC cohort.

Pollutant Quintile	**Single-Pollutant Model**	**Multi-Pollutant Model**				
		SO_2+PM_{10}	SO_2+PM_{10+} $PM_{2.5}$	SO_2+PM_{10+} $PM_{2.5}$+CO	SO_2+PM_{10+} $PM_{2.5}$+CO+NO_2	SO_2+$PM_{10+}PM_{2.5}$ +CO+NO_2+ O_3
CO						
1	ref				ref	ref
2	1.08 (0.70, 1.66)				1.09 (0.59, 2.04)	1.04 (0.53, 2.04)
3	1.10 (0.71, 1.70)				1.15 (0.58, 2.27)	1.16 (0.68, 1.99)
4	1.30 (0.83, 2.05)				1.31 (0.55, 3.09)	1.35 (0.57, 3.20)
5	1.19 (0.74, 1.92)				1.34 (0.44, 4.14)	1.29 (0.56, 2.97)
O_3						
1	ref					ref
2	0.94 (0.62, 1.43)					0.95 (0.52, 1.73)
3	0.88 (0.58, 1.33)					0.98 (0.52, 1.85)
4	0.76 (0.50, 1.17)					0.68 (0.35, 1.31)
5	0.66 (0.43, 1.02)					0.97 (0.42, 2.20)

*Adjusted for the matching factors (age, gender, and race/ethnicity).

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