Biomonitoring of Wildland Firefighters: Analysis of Methoxyphenols as Viable Urinary Biomarkers of Wood Smoke Exposure

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A thesis
submitted in partial fulfillment
of the requirements for the degree of

Master of Science

University of Washington

2012

Program Authorized to Offer Degree:
Occupational & Environmental Exposure Sciences

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Abstract

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Occupational exposures to wood smoke can lead to a multitude of health effects, making it a pertinent issue of public health. Biomass in the form of wood smoke can lead to adverse health effects including respiratory impairment, asthma, cardiovascular disease, and increased mortality, especially with people who are immune-compromised. Wildland firefighters comprise a group that faces much higher exposures to wood smoke than other groups, making them a candidate for further understanding wood smoke exposure and its potential for health effects. Biomonitoring involves utilizing biomarkers of exposure as a means for quantifying the dose-response exposure relationship with other more harmful compounds. One of the compound groups that is specific to wood smoke are methoxyphenol compounds, which are formed during the pyrolysis of lignin. By measuring biomarkers of wood smoke exposure, the ambient concentration of wood smoke can be determined. This study investigated the dose-response relationship of wood smoke exposure and concentrations of urinary methoxyphenol compounds, specifically guaiacols and syringols in wildland firefighters to determine if methoxyphenol compounds could serve as suitable biomarkers of wood smoke exposure. Full-shift personal exposure monitoring of USFS wildland firefighters at the USDS Savannah River Site in South Carolina occurred during the dormant burn season of spring 2008 and

spring 2009. There were a total of 155 person-day samples collected over 32 days during the study. Urine samples were analyzed at the University of Washington for creatinine and 13 methoxyphenol compounds. The results indicated that the eight-hour TWA for CO exposure was the most representative exposure variable based on high Pearson's correlation values with other exposure variables and urinary methoxyphenols. Propylguaiacol was the most representative methoxyphenol compound for this dataset. The regression results indicate an association exists between CO and post-shift concentrations of propylguaiacol. Further, cross-shift concentrations of propylguaiacol also were associated with the eight-hour CO TWA as an exposure variable. Future studies hope to investigate subject-specific differences in urinary methoxyphenol concentrations.

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Acknowledgments:

It is a pleasure to thank those who made this thesis possible. Firstly, I would like to thank my thesis committee supervisor and adviser, Dr. Christopher Simpson for all of his mentorship and guidance through this project. Dr. Simpson's encouragement and support through the initial steps of this thesis to the final paper enabled me to develop an understanding of the subject of biomonitoring, specifically in wildland firefighters. I would also like to thank my committee members, Dr. Noah Seixas and Dr. Lianne Sheppard for their support and guidance through this process as well. Dr. Seixas and Dr. Sheppard provided expertise in the field from multiple backgrounds, which greatly aided in my success for completing this thesis. I also would like to thank the Simpson Analytical Lab for making this project possible. Without the support of the chemists and sampling crew, I would have been unable to pursue this project. Finally, I would like to thank my family and friends for their unwavering support through this endeavor.

Chapter One: Background and Significance

Introduction to Particulate Matter and Wood Smoke

Particulate matter from air pollution presents a serious issue for public health all over the world. Particulate matter for centuries has polluted cities around the globe and shortened people's lives. In recent years, epidemiologic studies have reported associations between acute and chronic exposures to particulate matter and increases in morbidity and mortality. As the global society continues to grow, the issue of particulate matter exposure and associated health effects will only persist. The effects of particulate matter depend on several factors, such as particle size distribution and the nature of the particle. Additionally, the sources play a major role. One source that continues to lead to health effects is wood smoke (Naeher et al. 2007). For wildland firefighters, occupational exposures to wood smoke occur more frequently than most groups.

Wildland firefighters can spend 4-8 months fighting fires and performing controlled burns during an active fire season. With high exposures and little to no respiratory protection, the risk for health maladies associated with occupational exposures to particulate matter in wood smoke increases. Some of these health effects include respiratory illnesses and an increased risk for cardiovascular disease, demonstrating the importance of this topic.

Currently, exposure assessment involves the use of personal sampling pumps that monitor exposures to particulate matter and the use of passive devices for CO. For firefighters, with shifts that can last from 8-36 hours, carrying a personal sampling pump can become exhausting, due to the cumbersome nature of these devices. Additionally, these sampling pumps cannot address temporal and spatial variability in the wood smoke, nor can the pumps determine individual effects in the firefighters, such as metabolism.

Biomonitoring involves measuring concentrations of known compounds specific to the exposure substance of interest, in this case wood smoke. By measuring known concentrations of metabolites of wood smoke, it is possible to determine what the exposure was for an individual. For firefighters, by measuring compounds specific to wood smoke in urine, such as methoxyphenol compounds, levoglucosan, or polycyclic aromatic hydrocarbons, the potential for determining individual exposures becomes more

feasible. Biological monitoring eliminates the need for active sampling pumps and relies instead on the collection of biological samples, such as blood, urine, or hair.

This background section will discuss these various topics in more detail to create a framework for understanding occupational exposures to wood smoke and the potential for utilizing a viable biomarker of exposure to determine these exposures.

Particulate Matter: Classification and Chemical Information

Particulate matter is a generic term to describe a complex mixture of chemicals that originate from any number of sources both natural and anthropogenic (U.S. EPA, 2011). They can include mixtures of acids (such as nitric and sulfuric), organic chemicals, metals, soil and dust particles, and allergens (U.S. EPA, 2010). $PM_{2.5}$ consists of fine particles with a median aerodynamic diameter of 2.5μm, while PM_{10} consists of particles with a median aerodynamic diameter of 10μm (U.S. EPA, 2012a). Respirable particles are 4μm in median aerodynamic diameter and are found in haze and wood-smoke, sea salt marine air, volcanoes, home heating, industry, and other combustion sources.

Particulate matter originates from primary and secondary sources. Primary forms of particulate matter refer to particles that form at the source, such as construction sites, fires, or smoke stacks (U.S. EPA, 2012a). Secondary particles form from complicated atmospheric conversions of gas phase precursors emitted from power plants, automobiles, and industry (U.S. EPA, 2012a).

Particulate Matter Standards

Due to the potential for exposure to particulate matter from myriad sources, the U.S. Environmental Protection Agency set the National Ambient Air Quality Standards (NAAQS) for particulate matter, which originated out of the Clean Air Act (U.S. EPA, 2012b). The NAAQS sets limits on exposure to particulate matter and other criteria air pollutants because they are known human health hazards (U.S. EPA, 2012b). Other criteria air pollutants include lead, carbon monoxide, sulfur oxides (SO_x), nitrogen oxides (NO_x), and ozone (O_3) (U.S. EPA, 2012b). $PM_{2.5}$ has an annual standard that cannot

exceed 15 µg/m^3, while the 24-hour standard cannot exceed 35 µg/m^3 (U.S. EPA, 2012b). This standard recently decreased from 65 µg/m^3 (U.S. EPA, 2012b). Some cities around the world, such as Cairo, witness 24-hour concentrations for PM2.5 that frequently exceed 150 µg/m^3 (Abu-Allaban et al. 2007). Other cities like Beijing frequently exceed 24-hour PM2.5 concentrations of 350 µg/m^3 (He et al. 2001).

The OSHA PEL for particulate matter is 15 mg/m^3. This standard relates to acute exposures to particulate matter, rather than chronic exposures. Chronic effects to particulate matter are not tracked by OSHA. Therefore, the potential for chronic effects exists, especially at lower levels, but the standards are not currently in place to regulate particulate matter and chronic health effects.

Particulate Matter Health Effects

Many health studies have found an association between exposure to particulate matter and premature death (USEPA, 2010). While particulate matter causes respiratory issues, such as respiratory symptoms, asthma attacks, and bronchitis, it also can lead to cardiovascular symptoms, cardiac arrhythmias, and heart attacks (USEPA, 2010). These particular health effects demonstrate a need for continuing research into reducing exposures.

One study that investigated short-term effects of exposure to $PM_{2.5}$ in children with asthma found that for a short-term increase in 10 µg/m^3 $PM_{2.5}$ lagged one day, the odds ratio/relative risk for an asthma attack was 1.16 (95% CI: 1.03, 1.30) (Slaughter et al. 2003).

Further, another study found that an association exists with every 10 µg/m^3 increase in fine particulate matter ($PM_{2.5}$) exposure and a 15-27% increase in the relative risk for lung cancer after adjusting for confounders such as cigarette smoking, radon exposure, and occupational exposures (Turner et al. 2011). The study looked at approximately 189,000 never smokers over a period of 26 years to determine if any association existed (Turner et al. 2011).

Finally, a third study found that with short-term exposures to $PM_{2.5}$ there is an association with increased mortality (Garrett et al. 2011). The study found that in older

age groups (>= 65) the risk of cardiovascular mortality associated with $PM_{2.5}$ exposure was 2.39% with each increase in 10 $\mu g/m^3$ of exposure (Garrett and Casimiro, 2011).

The results of these three studies, which represent a small sample of the far-reaching compilation of epidemiologic studies linking particulate matter and disease, indicate that short-term exposures to $PM_{2.5}$ can lead to serious health effects including severe asthma attacks, increased odds for developing lung cancer, and increased cardiovascular mortality in older individuals. For those who face occupational exposures to $PM_{2.5}$, not only do they face acute and sub-acute exposures, but they also face sub-chronic and chronic exposures, which are associated with a multitude of health effects, warranting the importance of further research to reduce or mitigate the exposure hazards.

As Naeher et al. 2007 discusses, chronic pulmonary dysfunction may result from repeated exposure to smoke, especially for individuals that do not use respirators. However, the potential for these chronic effects to diminish in off-seasons has not yet been determined. Therefore, the potential for long-term effects exists from repeated exposures. One group that consistently faces occupational exposure to $PM_{2.5}$ in wood smoke is wildland firefighters during controlled burns and wildfires.

Wood Smoke: Classification and Health Effects

Wood smoke consists of a mixture of particulate matter, sulfur and nitrogen oxides, volatile organic compounds, free radicals, chlorinated dioxins, and many other compounds (Naeher et al. 2006). Many of these compounds are known carcinogens and can lead to a multitude of adverse effects (Simpson and Naeher, 2010). Epidemiologic studies have shown an association between wood smoke exposure and increased mortality, hospitalization, and variability in heart rate (Hinwood et al. 2008).

One of the major sources of wood smoke comes from indoor fires, which might include cooking, heating, or smoking food (Clarke et al. 2007). Though most people in the U.S. do not rely on wood-fire stoves for cooking, close to three billion people worldwide depend on biomass in the form of wood as a source for energy and cooking (Clarke et al. 2007). This dependence leads to reductions in indoor air quality and greater negative health effects. However, due to the poor ventilation conditions, the problem only is exacerbated for many of the people in these parts of the world.

In addition to indoor smoke exposure, outdoor smoke exposure from fires also contribute to the decrease in health for many of these individuals. When major brush and forest fires occur, at least in the developed world, wild fire smoke can lead to communal increases in asthma, chronic obstructive pulmonary disorder (COPD), and bronchitis (Dills et al. 2006). In developing nations, the risks expand to COPD, asthma, lung cancer, the development of cataracts, and otitis media (the inflammation of the middle ear), since wood smoke concentrations tend to be higher in developing nations (Dills et al. 2006). Further, tuberculosis and child acute respiratory infections become exacerbated by wood smoke because it irritates the lungs (Clarke et al. 2007). Vegetation and forest fires in 1997-98 exposed an estimated 70 million Southeast Asians to wood-smoke (Dills et al. 2006). Because of these fires, 527 deaths occurred, along with 15,800 hospitalizations from smoke inhalation (Dills et al. 2006).

Wildland Firefighter Exposure to Wood Smoke

Occupational exposure to wood smoke in wildland firefighters is an important area of study because of the potential for the numerous health effects associated with particulate matter exposure (Reinhardt et al. 2000). 40 percent of firefighter medical problems during the 1988 Yellowstone fire were associated with respiratory illnesses due to particulate matter exposure (Naeher et al. 2005). An estimated 80,000 firefighters each year in the United States contribute to the management of controlled burns and wildland fire events, including forest and brush fires (Betchley et al. 1997). With so many wildland firefighters that mitigate both brush and forest fires, while also performing controlled burns, the potential for respiratory illnesses developing from occupational exposure to wood smoke is substantial.

As of 1997, up to 70,000 controlled burns were conducted during an active fire season in the U.S. (Ward et al. 1989, Betchley et al. 1997). Southern land managers in particular, participated in some of the greatest volumes of controlled burning in the U.S. (Naeher et al. 2006). Each year, land managers in the southern part of the U.S. conduct prescribed burning on 6-8 million acres of forests and agricultural lands per year as a means of managing the lands (Naeher et al. 2006). This actually accounts for more prescribed burning than anywhere else in the country (Naeher et al. 2006).

An active fire season can last four to six months or even longer, creating the potential for cross-seasonal occupational wood smoke exposures in wildland firefighters. Although wildland firefighters might not be exposed to the higher concentrations of smoke associated with urban fires, their exposure period is much longer, with typical shifts lasting 12-18 hours (Betchley et al. 1997). In the initial fire suppression period, these shifts can last up to 48 hours, placing the firefighters at greater risk for health effects. With particularly large fire events, these workers might fight for up to 14 days and longer with nearly constant exposure (Betchley et al. 1997).

One study reported cross-shift occupational exposures to wood smoke from prescribed burns of 630 $\mu g/m^3$ $PM_{3.5}$ with maximum concentrations of 6900 $\mu g/m^3$, over shifts that lasted 11.5 hours, while a second study reported cross-shift occupational exposures to wood smoke from wildfires of 500 $\mu g/m^3$ $PM_{3.5}$ with maximum concentrations of 2930 $\mu g/m^3$ over shifts that lasted 10.4 hours on average (Reinhardt et al. 2000, Reinhardt et al. 2004). Naeher et al. 2005 reported mean full work shifts for $PM_{2.5}$ exposures during prescribed burns of approximately 385 $\mu g/m^3$ with a range of 21.3 to 2462 $\mu g/m^3$ (Naeher et al. 2006).

Previous studies on urban firefighters indicate that during acute exposures to smoke, respiratory function measured by forced expiratory volume (FEV), forced vital capacity (FVC), and forced expiratory flow (FEF) decreases (Tepper et al. 1991, Betchley et al. 1997). Many epidemiologic studies investigate health effects by measuring FEV, FVC, and FEF, demonstrating that for firefighters, lowered function with respect to these measures can indicate serious respiratory effects.

Exposure Assessment

Trying to assess occupational wood smoke exposure accurately can be extremely difficult. Because of spatial and temporal variations in the wood smoke, accurate measurement of occupational wood smoke exposure can be challenging (Simpson and Naeher, 2010). One area might encounter different mixing in the air, leading to diverging concentrations. People constantly move between microenvironments as well, leading further to variations in exposure (Simpson and Naeher, 2010). When taking all of these

different conditions into account, it makes it difficult to capture the full variability of the exposure for the individual (Simpson and Naeher, 2010).

The traditional method of determining exposures involves taking personal samples of compounds, such as carbon monoxide and particulate matter. Considering that fire regimes produce higher quantities of $PM_{2.5}$ and CO than other sources of $PM_{2.5}$ and CO, such as a diesel engine, the potential for accurately determining source emissions is higher when these compounds are compared between multiple sources and each other. $PM_{2.5}$ and CO are both characteristic of combustion, but alone, $PM_{2.5}$ could represent dust, while CO represents incomplete combustion from a car engine. Therefore, comparing the two exposure variables and especially the concentrations is important for determining sources.

One of the greatest limitations of sampling pumps is the added bulkiness of the pumps. Over a shift of 8-14 hours, a firefighter will no longer want to wear a heavy, cumbersome pump. The potential for losing pumps or having subjects remove them before the end of a sampling period can be extremely high, especially on a stressful day for the subject. Therefore, with this in mind, trying to determine a method that is more feasible for the subjects, but also increases the accuracy of determining potential health outcomes from the occupational exposure is necessary. One potential means is to use the process of biological monitoring.

Biomarkers of Exposure

Biomonitoring involves utilizing biomarkers of exposure as a means for quantifying the dose-response exposure relationship with other more harmful compounds. By measuring biomarkers of wood smoke exposure, the ambient concentration of wood smoke can be determined (Simpson et al. 2010). This can lead to a greater understanding of the exposure period and the potential for developing negative health effects. Wood smoke biomarker compounds can include polycyclic aromatic hydrocarbons (PAHs), levoglucosan, and methoxyphenols. Each of these will be discussed to determine their benefits and drawbacks.

Polycyclic Aromatic Hydrocarbons

Among other potential pathways, PAHs form during combustion of biomass, such as wood. Some of these compounds include naphthalene and pyrene as well as their metabolites, 2-naphthol and 1-hydroxypyrene. The PAHs form during pyrolysis which led to these compounds being used as biomarkers of wood smoke exposure in a study on Brazilian charcoal workers performed by Kato et al. 2004.

However, as Simpson et al. 2010 observed, PAH compounds "are by no means specific to wood smoke, [but] are a component of incomplete combustion and are present in a variety of PM sources, including vehicle exhaust, gas and coal combustion, and cooking fumes," (Simpson et al. 2010). Taking this into account, several confounders exist that can distort a true association between the pyrene/naphthalene biomarkers and wood smoke as the primary source. Therefore, by understanding the limitations of PAHs as biomarkers of wood smoke exposure, due to the multitude of confounders, it seems that a more viable candidate is needed.

Levoglucosan as a Potential Exposure Biomarker

Another likely candidate as a biomarker is levoglucosan. Levoglucosan forms from the pyrolysis of cellulose making it a potential biomarker (Simpson et al. 2010). The compound is abundant in wood-smoke, which makes it promising for future study. Levoglucosan can be used as a tracer for $PM_{2.5}$ and represents about 2.8-3.8% of $PM_{2.5}$ mass from foliar fuels (Hays et al. 2002) and closer to 5.7% as measured in wildland forest fires in Georgia (Miglaccio et al. 2009, Lee et al. 2005).

Due to the inherent nature of levoglucosan forming from pyrolysis of cellulose, hypothetically the compound should enter the body via inhalation of wood smoke and thus serve as a potential tracer for wood smoke exposure. A few major studies have been performed to test this hypothesis and the results were mixed. One study by Miglaccio et al. 2009 found that mice exhibited higher levels of levoglucosan after being exposed to wood smoke. School children from Libby, Montana also exhibited urinary levoglucosan mean concentrations of 55 ng/mg creatinine (Miglaccio et al. 2009, Simpson et al. 2010)

demonstrating a presence of the compound in the body and suggesting some association as a predictor of wood smoke exposure.

A study performed by Hinwood et al. 2008 found no real difference between pre-exposure levoglucosan concentrations and post-exposure concentrations in twelve participants of a fire-training exercise, but the study did conclude that levoglucosan was present in the body and the mean concentration was 4700 ng/mg creatinine (Hinwood et al. 2008). This may or may not be explained by the presence of wood smoke.

Bergauff et al. 2010 investigated the potential of urinary levoglucosan as a biomarker of wood smoke exposure in two different controlled settings, including a campfire exposure study and a wood stove exposure study. The results of the campfire study showed individual $PM_{2.5}$ exposures of 0.84 to 2.99 mg/m^3 and individual levoglucosan exposures of 76 to 256 $\mu g/m^3$ (Bergauff et al. 2010). However, there was no consistent response between wood smoke exposure and urinary levoglucosan.

Similarly, the results of the wood stove exposure trial showed individual $PM_{2.5}$ exposures that ranged from 1.15-1.97 mg/m^3 (Bergauff et al. 2010). Again, the urinary levoglucosan measurements from the subjects showed no consistent response and no consistent association as a predictor of wood smoke (Bergauff et al. 2010). What the study demonstrated was that levoglucosan was present in the body, but it could not be determined if the presence was due to wood smoke or some other source. Levoglucosan is found in food like caramel, which can explain the potential for concentrations in the subjects' urine prior to the wood smoke exposure trials.

Methoxyphenol Compounds as Potential Biomarkers of Wood Smoke Exposure

Methoxyphenol compounds are formed from the pyrolysis of lignin, which is a polymer found in wood (Simpson et al. 2010). Unlike levoglucosan, which can be found in many foods as well as wood smoke, methoxyphenol compounds are unique to wood-smoke and only a few food sources (Simpson et al. 2010). This makes controlling for confounders much easier for determining consistency between wood smoke exposure and concentrations of urinary methoxyphenols, rather than levoglucosan. The results of several studies demonstrate promising aspects of methoxyphenol compounds as potential biomarkers of wood smoke exposure.

Dills et al. 2006 determined methoxyphenol concentrations in subjects after exposing them to wood smoke from a campfire. The results indicated increased concentrations of urinary methoxyphenols (Dills et al. 2006). 74 percent of the variance in urinary methoxyphenol concentrations was explained by the $PM_{2.5}$ exposure. These results were more consistent for an association with 12 hour post-exposures compared to 24-hour post-exposures (Dills et al. 2006). Considering that methoxyphenols have a half-life in the body of 2-6 hours (one half of the concentration is metabolized in the body every two to six hours) (Dills et al. 2006), this would account for why the 12-hour samples appeared to demonstrate more of an association than for 24-hour post-exposure samples. When the samples were corrected for the creatinine concentrations, it disregarded the need to know the urinary formation period (Dills et al. 2006). Additionally, the methoxyphenol compounds correlated strongly with airborne exposure to levoglucosan. The research group suggested in their conclusions that methoxyphenols could be used as biomarkers for exposures of >700 $\mu g/m^3$ wood smoke (Dills et al. 2006).

Some of the methoxyphenol compounds, like vanillin or eugenol, are found in foods, which can confound associations with wood smoke exposure (Dills et al. 2006). However, unlike PAH compounds, which have many potential sources, methoxyphenol compounds, with the exception of a few foods, are found mostly in the lignin-rich (woody) parts of plants (Dills et al. 2006).

Conclusions from the Literature Review

When compared to other potential biomarkers, urinary methoxyphenol compounds suggest the greatest potential as viable biomarkers of wood smoke exposure. Two previous studies showed promising results. One included the study of wood-smoke exposure to wildland firefighters (Neitzel et al. 2009), while the other investigated residents in rural Guatemala and their exposures to wood smoke from indoor cooking (Clarke et al. 2007). Although the Neitzel et al. 2009 study had a small sample size of 20 subjects, it demonstrated the viability of urinary methoxyphenols as dose-dependent predictors of wood smoke exposure. Based of the results of the Neitzel paper and the Clarke paper, this research project will explore the dose-dependent relationship of urinary methoxyphenol biomarkers as viable predictors of wood smoke exposure in a group of

wildland firefighters with a greater sample size than the pilot study performed by the Neitzel et al. study.

Specific Aims and Hypothesis:

Occupational and environmental exposure to particulate matter from the burning of biomass presents a serious issue for people in the United States. Biomass in the form of wood smoke can lead to adverse health effects including respiratory impairment, asthma, cardiovascular disease, and increased mortality, especially with people who are immune-compromised.

Wildland firefighters face greater occupational and environmental exposures to wood smoke than other groups. Due to shifts of 12-18 hours over long seasons, which can last for several months, wildland firefighters face a greater potential for adverse health effects due to inhalation of biomass smoke. During a fire regime, brush, forests, and other forms of vegetation can ignite, leading to greater inhalation of toxic compounds such as particulate matter, carbon monoxide, volatile organic compounds, and free radicals. Each of these present a serious risk of negative health effects for those exposed.

During an active fire season, many wildfires can occur, leading to greater numbers of firefighters needed to control the fires. Additionally, controlled burns are conducted periodically throughout a season to clear out brush and prevent larger fires from occurring. Because of the prevalence of both wildfires and controlled burns, the risk may increase over time for individuals exposed to the smoke produced by these fires.

At the present moment, it is difficult to assess accurately exposure from wood smoke, due to variable conditions, wood types, and individual variations in the human body, such as metabolism. Because of this, using active or passive monitors can only provide an estimate of exposure to wood-smoke during a fire or a controlled burn, due to a lack of specificity for wood smoke. Biomonitoring however can provide a more accurate assessment by accounting for individual variations such as metabolism and respiration.

Due to the limitations of other forms of exposure assessment, this project seeks to use biomonitoring to investigate the dose-response relationship for the excretion of urinary methoxyphenols in a cohort of wildland firefighters exposed to wood-smoke. Methoxyphenols are components of wood smoke that are produced by the pyrolysis of lignin and can serve as biomarkers of exposure. The group of wildland firefighters specifically is a cohort of United States Forest Service firefighters stationed at the

Savannah River Site (SRS) Forest Station in Aiken, SC and includes men and women between 21 and 54 yrs old.

Overall Scientific Aim:

To investigate the dose-response relationship of wood smoke exposure and concentrations of urinary methoxyphenol compounds in wildland firefighters to determine if methoxyphenol compounds can serve as suitable biomarkers of wood smoke exposure.

Specific Aim 1: To investigate if occupational exposure to wood smoke, as measured by particulate matter, carbon monoxide, and levoglucosan, influences creatinine-corrected urinary concentrations of methoxyphenol compounds.
Hypothesis: Occupational exposure to wood smoke is associated with increases in urinary concentrations of methoxyphenol compounds, which can serve as viable biomarkers of wood smoke exposure.

Specific Aim 2: In order to evaluate whether vegetation type differentially affects the dose-response relationship between exposure to wood smoke and methoxyphenol concentrations, we will develop an a multiple linear regression model, with an interaction term for fuel type, which will account for vegetation type.
Hypothesis: Vegetation type differentially affects the dose-response relationship between exposure to wood smoke and methoxyphenol concentrations.

Chapter Two: Methods

Location of Study and Subject Requirement

The data for this study was collected in the winter of 2008 and 2009 at the Savannah River Site Forest Station, a National Environmental Research Park in Aiken, South Carolina. The area was approximately 29 percent hardwood or mixed (pine, hardwood) and approximately 68 percent pine (Neitzel et al. 2009). The study investigated USFS forest fighters whose occupation involved burning 15,000 to 18,000 acres per year to restore longleaf fire savannah communities and wetlands native to the area.

The cohort of wildland firefighters included 18 subjects of which 16 were male and two were female. Personal work-shift exposures to carbon monoxide (CO), particulate matter less than or equal to 2.5 µm in diameter (PM2.5), and levoglucosan (LG) were measured in wood smoke.

17 of the 18 subjects responded to survey questions concerning demographic information. The results indicated that the firefighters ranged in age from 21-54 years old with an average age of 31.5 years old (SD: 8.3 years). Subjects volunteered to enlist into the study after the purpose of it was explained to them. Each volunteer signed a consent form to participate. Finally, the University of Georgia Institutional Review Board for inclusion of human subjects approved the study.

Personal Exposure Monitoring: $PM_{2.5}$ and CO

Full-shift personal exposure monitoring of wood smoke was performed on wildland firefighters during the dormant burn season of spring 2008 and spring 2009. There were a total of 155 person-day samples collected over 32 days during the study.

$PM_{2.5}$ exposure was monitored using SKC Air Check Model 2000 pumps (SKC Inc., Eighty Four, PA) attached to BGI Triplex Cyclones (BGI Inc., Waltham, MA), which were fastened to each firefighter. The pump flow rate was set at 1.5 L/min to achieve a 50 percent aerodynamic cut-off point at 2.5µm for the cyclone. Pre- and post-sampling flow rates were measured with a BIOS Dry Cal Defender 510 Model (BIOS International, Butler, NJ). $PM_{2.5}$ was collected on Gelman 37 mm filters (Pall Corp., Ann

Arbor, MI). Each filter contained a 100% poly-tetra-fluoro-ethylene (PTFE) membrane. The filters had a pore size of 2.0µm with polymethylpentene (PMP) support rings.

Real-time personal exposure monitoring of carbon monoxide (CO) was performed using Dräger PAC III single gas monitors (Dräger Safety Inc., Pittsburgh, PA) outfitted with CO sensors. Personal monitoring occurred in the breathing zone of each firefighter and the sampling instruments were placed in the pockets of the vests worn by each of them.

$PM_{2.5}$ Gravimetric Analysis

PM filters were stored in a climate-controlled laboratory for a minimum of 48 hours before they were weighed for the pre- and post-sample collection. Both weights (pre- and post-shift) were measured twice, with a Cahn C-35 microbalance that achieved a sensitivity of ±1µg, following the guidelines set forth by the U.S. EPA SOP (1998). The filters weights were determined by calculating the average pre-shift weight, which then was subtracted from the average post-shift weight of the filter. The TWA $PM_{2.5}$ concentrations then were calculated as the concentration of $PM_{2.5}$ per m^3 air.

Levoglucosan Analysis

Analysis of levoglucosan was undertaken at the University of Washington using previously described methods (Neitzel et al. 2009, Simpson et al., 2004, 2005). The protocol for levoglucosan from the filter $PM_{2.5}$ and its analysis involved many steps. Each PTFE filter membrane was cut from the support ring using a custom made Teflon/stainless steel cutter. The filters were spiked with a recovery standard, which was composed of deuterated levoglucosan (d7-levoglucosan) in silane treated headspace vials. Each of the filters was extracted for sonication for one hour in a freshly prepared solution of 3.6µM triethylamine in ethyl acetate. The extract volumes were reduced to approximately 0.5 mL in a Turbovap II concentrator under nitrogen. Extracts were filtered through a 0.45µm PTFE syringe filter into silanized autosampler vials and internal standards (triisopropylbenzene) and anhydroheptulose were added, which were used to monitor the efficiency of derivatization.

The derivatization reagent, methylsilyltrifluoroacetanide (MSTFA) together with trimethylchlorosilane and pyridine, was added to extracts and allowed to react in the dark for at least 6 hours. The derivatized extracts then were injected into a GC column by splitless injection. Quantification was performed using the *m/z* peaks of 204 (levoglucosan) and 206 (d7-levoglucosan) relative to a response at *m/z* 189 (triisopropylbenzene) and the calibration curve. The calibration was set to quantify the levoglucosan concentration within a range of 0.1-100 µg/mL in the extract.

Analysis for Methoxyphenols and Creatinine in Urine Samples

Urine samples were collected from the study participants, each day prior to the start of shift and immediately at the completion of the shift. The firefighters were given specimen collection cups to take home for the collection of their first morning void. At the end of the shift, the firefighters again were given sample collection cups to collect the post-shift voids. Each specimen container was barcode labeled, scanned into an electronic database, and logged into a logbook. The samples were checked for the proper seal on the containers, bagged, and placed in a cooler filled with dry ice. After the coolers were filled, they were shipped to the National Center for Environmental Health laboratory at the Center for Disease Control in Atlanta, Georgia for temporary storage.

The urine samples in the current study were shipped to the University of Washington, to be analyzed for methoxyphenols and creatinine. Urine samples were analyzed for creatinine and methoxyphenols. Urinary creatinine was measured by a clinical laboratory using a colorimetric assay, and the creatinine measurements were used to correct for diuresis.

The analytical procedure for methoxyphenols has been described previously (Neitzel et al. 2009, Dills et al. 2001, 2006). 13 methoxyphenol compounds were measured, which included guaiacol, methylguaiacol, 2,3-dimethoxyphenol, ethylguaiacol, propylguaiacol, cis-isoeugenol, trans-isoeugenol, syringol, methylsyringol, ethylsyringol, allylsyringol, propylsyringol, and acetosyringol. The choice for these compounds was determined *a priori* based on previous studies in humans performed by Dills et al. 2001, 2006, and Neitzel et al. 2008.The analytical procedure involved acid hydrolysis of 10 mL aliquots of urine to deconjugate the methoxyphenols. The samples then were applied to

ion exchange solid phase extraction columns. The methoxyphenols were eluted with ethylacetate and analyzed by gas-chromatography-mass spectrometry (GC/MS). The deuterated methoxyphenols (synthesized in house) were amended to urine samples prior to extraction to monitor the analyte recovery. Assay blanks (10 mL H_2O) and the urine samples were fortified with methoxyphenols included with each batch of firefighter urine samples to monitor the assay precision and accuracy. For analyte identification, retention times for the analyte peaks were referenced to their respective retention time standards (each analyte's recovery time). The window of acceptance was approximately (± 0.002 min), and was based on the variability of the relative retention times observed in the calibrants.

Statistical Methods:

Statistical analyses of the atmospheric contaminants and urinary methoxyphenols were conducted using Stata/IC 11.2 for Windows (Stata Corp., College Station, TX USA). Histograms of the $PM_{2.5}$, CO, and LG exposure data were not normally distributed. For that reason, these compounds were log-transformed. Spearman's correlation coefficients were determined for the exposure variables in the exploratory analysis to assess the interaction between $PM_{2.5}$, CO, and LG. However, considering that all of the exposure variables were log-normally distributed, Pearson's correlation tables were created to assess associations between the log-transformed exposure and outcome variables. The Pearson's correlation data was used to inform choices of exposure and outcome variables included in subsequent models.

Because the half-life of urinary methoxyphenols is on the order of two to six hours (Dills et al. 2006), it is expected that post-shift urine voids will result in increased concentrations of urinary methoxyphenols. As performed in prior studies (Netizel et al. 2009), baseline concentrations of pre-shift concentrations were compared to post-shift data values. This association was assessed by including pre-shift concentration as a covariate of post-shift concentration and exposure.

Additionally, cross-shift changes in the concentrations were calculated to assess differences in concentrations of methoxyphenols over one shift. Cross-shift changes in concentrations were calculated by subtracting pre-shift data values from post-shift data

values. Negative cross-shift values were censored from the analysis because the values could not be log-transformed.

The urinary methoxyphenol pre- and post-shift concentrations were not normally distributed. For this reason, the variables were log-normally distributed. The histograms representing these variables can be found in the appendix (Figs. A7, A8, A10, and A11). The cross-shift concentrations were normally distributed, but considering the exposure variables were log-normally distributed, the cross-shift concentrations were log-normally distributed for easier interpretation of the regression models. The histograms of the cross-shift variables also can be found in the appendix (Figures A9 and A12).

Simple and multiple linear regression models then were developed for post-shift and cross-shift methoxyphenols. The R^2 values for the base models were used in a predictive capacity for determining which biomarker to select for further multiple regression models. The results of these analyses are featured in the discussion section of this thesis.

Chapter Three: Discussion of Results

Summary of Data Structure: Work Shift Duration, Pump Time, and Consecutive Work Shifts

Eighteen subjects participated in the study, and exposure and biomarker data were collected on a total of 136 work shifts. The number of shifts worked is unique for each firefighter. Although a total of 165 work shifts are recorded, urine samples were not collected for 29 of the work shifts. The distribution of shifts ranged from one to twenty-three shifts per firefighter, with twelve of the firefighters working less than ten shifts, three working ten to twenty shifts, and three working more than twenty shifts (see Appendix Table A1 and A2).

Two approaches were examined in order to determine the duration of work shift, for the purpose of calculating time weighted average (TWA) exposures (Refer to Figure A1 - A3). In the first approach, shift duration was determined by the length of time the sampling pump was worn by the subject. Additionally, the firefighters documented the length of their work shifts in diaries that logged daily activities concerning the fire regimes. Ultimately, pump time was considered the most appropriate means of determining the exposure and calculating the eight-hour TWA.

Exposure Variables: $PM_{2.5}$, Carbon Monoxide, and Levoglucosan

Personal exposure data are summarized in Table 1 and Table A3. Eight-hour time weighted averages (TWA) for each exposure variable were calculated to standardize the exposures to an eight-hour work shift. The eight-hour TWA was calculated by multiplying the concentration of pollutant by the work shift duration (in hours), and then dividing the (concentration x time) by eight hours. This calculation was based on the formula provided by OSHA 1910.1000(d)(1)(i).

Table 1: Descriptive Statistics of 8-hr TWAs for Exposure Variables

Variable	Vegetation Type	N	Mean	SD	Min	25th %ile	Median	75th %ile	Max
CO 8hr TWA (ppm)	Mixed	39	1.5	1.2	0.037	0.65	1.2	1.9	4.7
	Pine	79	2.7	2.2	0.021	1.2	2.3	3.6	9.8
	Total	118	2.3	2.0	0.021	0.95	1.8	3.1	9.8
PM 8-hr TWA ($\mu g/m^3$)	Mixed	36	570	430	66	280	510	680	2200
	Pine	78	750	540	120	400	610	920	2700
	Total	114	700	510	66	360	540	880	2700
LG 8-hr TWA ($\mu g/m^3$)	Mixed	35	24	29	0.21	6.2	14	24	140
	Pine	73	58	72	0.048	12	32	72	320
	Total	108	47	64	0.048	9.2	22	54	320

The summary data for the exposure variables ($PM_{2.5}$, CO, and LG) indicate that each variable produced different concentrations based on vegetation type. The mixed vegetation produced lower concentrations of $PM_{2.5}$, CO, and LG compared to the pine vegetation.

The OSHA PEL for carbon monoxide 8-hr TWA exposures is 50 ppm and the NIOSH REL is 35 ppm, indicating that all of the firefighters were well below the occupational exposure limit. The 8-hour TWAs for carbon monoxide are much lower than the OSHA PEL. This indicates that although the firefighters were not wearing respirators, the potential for human health effects from exposure to carbon monoxide was low.

The OSHA PEL for particulate matter is 15,000 $\mu g/m^3$, indicating that the subjects in this study were well below the OSHA limit. There is no OSHA PEL for levoglucosan, but it does serve as a fraction of $PM_{2.5}$ that is specific to plant matter. A higher concentration of levoglucosan indicates that a greater volume of plant matter burned. If levoglucosan is compared to $PM_{2.5}$, a fraction greater than 1.5 percent (Neitzel et al. 2009) would indicate more of the $PM_{2.5}$ would be due to wood smoke compared to another source, such as dust.

Fine et al. 2001 investigated smoke exposures based on different wood types and found different woods and burn conditions affect smoke chemistry. Depending on the wood type, the group found that differing levels of $PM_{2.5}$ are produced (Fine et al. 2001). Additionally, they found that there was no observed correlation with wood moisture content and $PM_{2.5}$. Finally, they determined that hard wood combustion emissions contain greater concentrations of syringols compared to soft woods. This is another reason that fuel type was investigated in this study.

The intensity of the fire as well as the composition of the wood type can contribute to the amount of CO, $PM_{2.5}$, and LG that is produced. It is possible that in general soft woods like pine vegetation produce greater concentrations of CO, $PM_{2.5}$, and LG, when compared to hard woods. However, this needs to be determined in a future study with a greater sample area of hard woods.

Examination of Correlations Amongst Urinary Biomarker, and Between Biomarkers and Exposure Measures

As a first step towards examining potential associations between urinary methoxyphenols and measurements of wood smoke exposure, we calculated Pearson and Spearman correlation Tables. See appendix A21-29 for Spearman's and Pearson's tables.

Table 3: Spearman Correlation of Exposure Variables

All Data	**ρ**	**N**	**p-value**
$PM_{2.5}$ vs. CO	0.65	122	<0.0001
$PM_{2.5}$ vs. LG	0.52	122	<0.0001
CO vs. LG	0.52	117	<0.0001
Data w/out Levoglucosan/$PM_{2.5}$<1.5%	**ρ**	**N**	**p-value**
$PM_{2.5}$ vs. CO	0.73	105	<0.0001
$PM_{2.5}$ vs. LG	0.71	105	<0.0001
CO vs. LG	0.63	100	<0.0001
Mixed Wood Compartments	**ρ**	**N**	**p-value**
$PM_{2.5}$ vs. CO	0.73	40	<0.0001
$PM_{2.5}$ vs. LG	0.51	40	0.0009
CO vs. LG	0.53	39	0.0006
Soft Wood Compartments	**ρ**	**N**	**p-value**
$PM_{2.5}$ vs. CO	0.61	81	<0.0001
$PM_{2.5}$ vs. LG	0.49	81	<0.0001
CO vs. LG	0.42	77	0.0001

Table 3 describes Spearman correlation coefficients for the exposure variables $PM_{2.5}$, CO, and LG. When including all the data, we see that $PM_{2.5}$ correlates more strongly with CO than with LG. Additionally, if the fire smolders, CO and $PM_{2.5}$ will still be produced, while LG may not be produced to the same extent as during the more intense fire stage. LG has similar correlations with CO and with $PM_{2.5}$.

Neitzel & colleagues suggested that samples with LG/$PM_{2.5}$ ratios that are small might be attributed to other sources rather than wood smoke (eg. re-suspended ash & dust), and used a cutoff LG: $PM_{2.5}$ ratio of >1.5 % to identify such samples (Neitzel et al. 2009). Table 3 indicates that the associations for all three exposure variables are higher when samples with low LG:$PM_{2.5}$ ratio are excluded, compared to the full exposure data

set. In addition, correlations between the exposure variables were assessed after stratifying by fuel type. The correlation between exposure variables for mixed woods is somewhat higher than the soft woods.

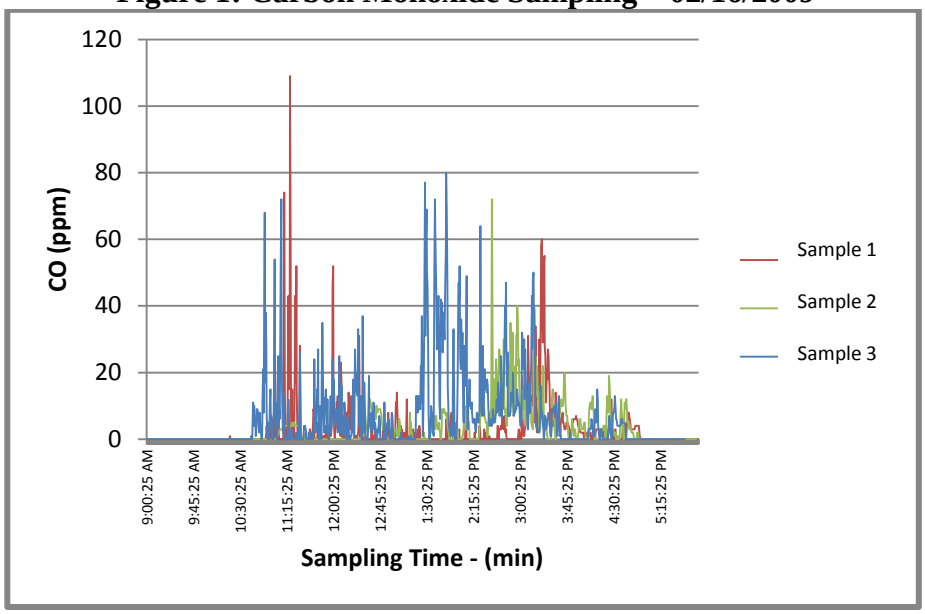
One primary question to consider is whether the exposure variables could act as surrogates of each other. The data in Table 4 indicates that there is a higher correlation between $PM_{2.5}$ and CO compared to CO and LG or $PM_{2.5}$ and LG. In contrast, a previous study in wildland firefighters (Neitzel et al. 2009) reported that LG and CO were more correlated than LG and PM.

The Pearson's correlation tables (refer to Figures A24-A29 in appendices) indicate correlation amongst clusters of compounds. Guaiacyl-type compounds area highly correlated with each other, specifically methylguaiacol, ethylguaiacol, and propylguaiacol. Syringyl-type compounds are also highly correlated with each other, specifically syringol, methylsyringol, ethylsyringol, and propylsyringol. Guaiacol compounds are more correlated as clusters in pine vegetation, while syringols are more correlated as clusters in mixed vegetation. The exposure variables also correlate with specific methoxyphenols. CO shows the highest correlation amongst guaiacols. Interestingly, some of the cross-shift syringyl-type compounds correlate with guaiacyl-type compounds in mixed vegetation, suggesting that there is a higher percentage of both hard and soft woods in the mixed vegetation.

CO Real-Time Data

The CO data presented in the preceding table represents concentrations averaged over approximately 6-8 hours. Figure 1 below plots continuous measures of CO exposure averaged every minute for workers on one representative sampling day. Several different peak exposures of carbon monoxide occurred, which can be interpreted as intense burn episodes that produced high quantities of wood smoke. Additionally, after the intense burns, periods of smoldering occurred, which also produced CO. However, there were long periods of no exposure at the beginning and end of the work shifts. This represents the one to two hour period of travel time to the burn site and the return back to the fire station after each work shift, where exposure to carbon monoxide was miniscule or did not occur.

Figure 1: Carbon Monoxide Sampling – 02/16/2009



In this particular example, the peaks in CO concentration for Sample 1, Sample 2, and Sample 3 range from 80 to 110 ppm. These however should not be directly compared to the OSHA PEL for carbon monoxide of 50 ppm, the NIOSH REL for carbon monoxide of 35 ppm, or the ACGIH TLV for carbon monoxide of 25 ppm, which are based on 8-hour time weighted averages (TWAs). Levels of carbon monoxide exposure that are greater than 1500 ppm are considered immediately dangerous to life or health (IDHL). These results indicate that the firefighters are well below the IDHL level for CO.

Outcome Data: Methoxyphenol Summary Statistics and Censored Explanations

This next section discusses the pre-, post-, and cross-shift urinary methoxyphenol concentrations that were measured for each of the firefighters.

Table 2: Post-shift, Pre Shift, Cross-Shift Methoxyphenol Descriptive statistics Prior to Replacing Censored Values

Pre-Shift Data (Creatinine Adjusted µg/mg)						Post-Shift Data (Creatinine Adjusted µg/mg)					Cross-Shift Data (Creatinine Adjusted µg/mg)				
Variable	**N**	**Mean**	**SD**	**Min**	**Max**	**N**	**Mean**	**SD**	**Min**	**Max**	**N**	**Mean**	**SD**	**Min**	**Max**
GU	82	0.69	0.59	0.068	2.89	80	0.81	0.58	0.16	2.7	67	0.12	0.47	-1.06	1.8
MEGU	85	0.067	0.070	0.0064	0.44	97	0.22	0.26	0.028	2.06	81	0.16	0.26	-0.23	1.63
23DMP	92	0.02	0.033	0.00029	0.21	92	0.024	0.026	0.00094	0.15	84	0.0012	0.03	-0.12	0.10
ETGU	99	0.083	0.16	0.000093	1.14	101	0.077	0.088	0.0027	0.52	99	-0.0047	0.17	-1.0	0.52
SY	87	0.02	0.026	0.00054	0.13	85	0.16	0.34	0.076	2.4	75	0.14	0.36	-0.11	2.37
PRGU	83	0.016	0.055	0.00012	0.32	97	0.011	0.024	0.00047	0.23	79	-0.0050	0.063	-0.31	0.23
CISOEU	74	0.022	0.024	0.00096	0.15	85	0.075	0.11	0.0025	0.76	67	0.064	0.13	-0.052	0.74
MESY	86	0.0074	0.016	0.00020	0.082	86	0.074	0.18	0.0016	1.17	72	0.059	0.14	-0.063	0.94
TISOEU	82	0.022	0.029	0.00053	0.17	88	0.082	0.10	0.0023	0.47	72	0.060	0.10	-0.084	0.43
ETSY	83	0.0066	0.082	0.00012	0.043	80	0.042	0.10	0.00088	0.68	66	0.041	0.11	-0.031	0.68
ALLSY	84	0.18	0.69	0.00055	4.69	74	0.21	0.48	0.0015	2.50	65	-0.053	0.89	-4.54	2.48
PRSY	62	0.0021	0.0057	0.00011	0.048	73	0.0069	0.017	0.000088	0.12	46	0.15	0.0099	-0.035	0.048
ACETSY	81	0.023	0.025	0.00093	0.13	79	0.033	0.024	0.0019	0.11	68	0.0083	0.029	-0.090	0.10

Summary statistics for the outcome variables are presented in Table 2 (histograms illustrating the data distribution for the outcome variables are presented in figures A7-A11 in the appendix). The data indicates that the most abundant compound present for post- and pre-shift data is guaiacol. The highest cross-shift concentration is observed with methylguaiacol. The least abundant compound measured is propylsyringol.

Tables A6-A8 present the censored data and indicate that the syringol compounds are heavily censored, especially for cross-shift samples. Based on these results, it can be interpreted that the urinary syringols are not as reliable a measure of wood smoke exposure as the guaiacol compounds. 67 percent of the work shifts, the firefighters worked in soft wood vegetation (pine wood). The other 33 percent of work shifts were in areas of mixed hard wood and soft wood vegetation. As pine is the dominant vegetation in the area, the potential for syringols being produced is much lower, since syringols are more associated with hard wood vegetation rather than soft wood vegetation (Dills et al. 2001). In contrast, soft woods including pine, have higher concentrations of guaiacols (Dills et al. 2001).

Potential Confounders of Associations Between Urinary Methoxyphenol Concentrations and Woodsmoke Exposure in the Firefighter Cohort

Questionnaires were given to the firefighters after each shift to document qualitative aspects of the study, such as if the firefighter smoked cigarettes or was exposed to secondhand cigarette smoke. Further, the questions investigated if smoked or grilled foods were consumed, as well as the intensity of the burn for the day. This section discusses some of the potential confounders of the study based on the questionnaire.

Arnarp et al. 1989 found that when cigarettes are burned, guaiacol compounds form during the pyrolysis of the tobacco material. Based on the results shown in Appendix Table A14 from the questionnaire data, no significant influence of smoking on post-shift biomarker concentrations was observed in this study. Ultimately, there are so few instances of smoking during the course of this study, that the statistical power is not adequate to demonstrate any association.

Refer to the appendix (Tables A15-A20) for the results of the t-tests performed on the methoxyphenols and grilled and smoked foods. The data indicates that grilled foods

do not produce any significant association with grilled foods and urinary methoxyphenol concentrations in the subjects. Smoked food consumption also exhibits no significant association on urinary methoxyphenols.

Respirators were not used by any of the subjects during the duration of the study.

Linear Regression Analyses

To reduce the number of potential outcome variables examined in the linear regression analyses, specific methoxyphenol compounds were selected based on analytical reasons and correlation between compounds. For the data censored due to non-detects or values below the minimum reporting limit, the concentrations were replaced with the limit of detection divided by the square root of 2. However, if too many of the concentrations were replaced by this new value, the actual concentration could become biased. Therefore, compounds with more than 20 percent of the post- or cross-shift values censored for analytical reasons were excluded from the regression models.

Table 4: Base Regression Models for Exposure and Methoxyphenol Variables for Post-Shift Samples

Post-Shift Creatinine Adjusted Methoxyphenols (µg/mg creatinine)										
Log Transformed Predictor Variable	**Log Transformed Outcome Variable**	**N**	**Coefficient**	**Std. Err.**	**Coefficient P-Value**	**95% CI**		**Model R^2**	**F-statistic**	**F-statistic P-value**
PM2.5 8hr-TWA	Guaiacol	85	0.55	0.29	0.07	-0.04	1.13	0.04	3.47	0.07
	Methylguaiacol	93	0.54	0.15	<0.0001	0.24	0.84	0.13	13.04	0
	Ethylguaiacol	94	0.58	0.15	<0.0001	0.29	0.88	0.14	15.37	0
	Propylguaiacol	**94**	**0.69**	**0.13**	**<0.0001**	**0.44**	**0.94**	**0.25**	**29.81**	**<0.00001**
	Cis-Isoeugenol	84	0.65	0.21	0	0.24	1.07	0.11	9.83	0
	Syringol	94	0.25	0.2	0.22	-0.15	0.66	0.02	1.51	0.22
	Methylsyringol	94	0.38	0.21	0.07	-0.03	0.79	0.04	3.36	0.07
	Ethylsyringol	85	0.33	0.2	0.11	-0.07	0.72	0.03	2.68	0.11
CO 8hr-TWA	Guaiacol	85	0.19	0.19	0.34	-0.2	0.57	0.01	0.92	0.34
	Methylguaiacol	92	0.37	0.1	<0.0001	0.18	0.56	0.14	14.72	0
	Ethylguaiacol	93	0.45	0.1	<0.0001	0.25	0.65	0.18	19.83	<0.00001
	Propylguaiacol	**93**	**0.57**	**0.09**	**<0.0001**	**0.39**	**0.75**	**0.31**	**40.89**	**<0.00001**
	Cis-Isoeugenol	83	0.4	0.15	0.01	0.11	0.7	0.08	7.42	0.01
	Syringol	93	0.03	0.14	0.85	-0.26	0.31	0	0.04	0.85
	Methylsyringol	93	0.12	0.15	0.43	-0.18	0.41	0.01	0.62	0.43
	Ethylsyringol	84	0.14	0.14	0.32	-0.14	0.42	0.01	1	0.32
Levoglucosan 8-hr TWA	Guaiacol	85	0.19	0.14	0.19	-0.1	0.47	0.02	1.73	0.19
	Methylguaiacol	93	0.28	0.07	<0.0001	0.13	0.42	0.14	14.22	0
	Ethylguaiacol	94	0.28	0.07	<0.0001	0.14	0.43	0.14	14.76	0
	Propylguaiacol	93	0.31	0.06	<0.0001	0.18	0.43	0.21	23.88	<0.00001
	Cis-Isoeugenol	84	0.36	0.1	<0.0001	0.17	0.55	0.14	13.65	0
	Syringol	94	0.09	0.1	0.38	-0.11	0.29	0.01	0.79	0.38
	Methylsyringol	94	0.17	0.1	0.1	-0.03	0.37	0.03	2.76	0.1
	Ethylsyringol	85	0.15	0.1	0.13	-0.04	0.35	0.03	2.36	0.13

Table 4 summarizes the base regresion models (post-shift biomarker levels) for each of the compounds that was selected for further analysis based on Pearson's Correlation tables as well as the chemical censoring explanations. The compounds that were chosen represent compounds from both syringyl-type compounds and guaiacyl-type compounds. This creates an understanding of all of the most highly correlated compounds and which compounds demonstrate the most association with exposure to $PM_{2.5}$, CO, and levoglucosan.

After investigating the Pearson's correlation tables (Appendix Tables A24-A29), it was determined that the eight-hour time wieghted average exposure to CO is more correlated with the outcome variables than $PM_{2.5}$ or LG. For this reason, CO was chosen for multiple regression models that included multiple urinary outcome variables.

Finally, after examining the correlation coefficient values in the simple linear regression model, it was determined that propylguaiacol exhibits the strongest association with the CO eight-hour time weighted average exposure. For this reason, we chose to use propylguaiacol to represent the other (correlated) methoxyphenols in the subsequent multiple linear regression models that we developed.

In building the multiple linear regression models, we considered several covariates that would plausibly influence the association between wood smoke exposure and biomarker response. Covariates of the model included fuel type, the interaction between fuel type and CO, and the log-transformed pre-shift propylguaiacol concentration. The model explains 33% of the variability in log-transformed urinary propylguaiacol concentrations.

Other variables that were initially explored as potential covariates included the CO:PM ratio and the duration of time between the last CO exposure and the post-shift urine void. The CO:PM ratio variable was considered because it acts as a potential means of identifying sources with higher levels of CO and $PM_{2.5}$ compared to sources with no CO, such as dust. However, this ratio did not exhibit a cut-off point for a percentage of CO:PM that was specific to wood smoke. Further, the variable was not statistically significant when it was included in the multiple linear regression models and did not greatly affect the R^2 value.

The variable explaining the duration of time from the last CO exposure and the post-shift urine void was also considered as a means of determining if post-shift urine samples were indicative of recent wood smoke exposure. Considering the short half-life of methoxyphenols, especially propylguaiacol (~2 hours), it seemed necessary to test for this duration and compare it

to the post-shift concentrations. However, it does not exhibit a significant statistical association when included in the model. Further, there are several missing data values for this variable, reducing the sample power greatly.

As discussed earlier in this chapter, it is possible the pre-shift levels of methoxyphenols (either from workshifts on previous days, or from non-occupational sources of woodsmoke exposure) could influence post-shift biomarker levels, and could affect the association beween workshift exposures and post-shift biomarker levels.

In Table 5 and 6, we included pre-shift urine concentrations in the model as one approach to adjust for the effect of pre-shift urine concentration on the association of interest. An alternative approach is to calculate the cross-shift change in biomarker levels, as described by Neitzel (2009). We conducted the regression analyses using cross shift-change in urinary concentration as the outcomes variables.

The base model of the association between post-shift propylguaiacol associations and work shift exposure to CO had a better R^2 value than the model based on cross-shift change in propylguaiacol (0.31 vs. 0.25). Similarly the fully developed model for propylguaiacol and CO based on post-shift biomarker measurements has an R^2 of 0.33, vs. 0.27 for the fully developed model based on cross shift measurements. The coefficient for pre-shift propylguaiacol in the post-shift model is not statistically significant, indicating that the two variables are independent of each other. Because they are independent of each other, the potential for pre-shift concentrations to influence post-shift concentrations is not a significant association according to the results of the regression model. Therefore, it signifies that cross-shift concentrations of propylguaiacol increase with exposure to wood smoke, independent of prior exposures.

Table 5: Multiple Linear Regression Model of Carbon Monoxide on Post-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine)

Post-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine)												
Log Transformed Predictor Variable	**Log Transformed Outcome Variable**	**Covariate**	**N**	**Coefficient**	**Std. Err.**	**Coefficient P-Value**	**95% CI**		**Model R^2**	**Adj. R^2**	**F-statistic**	**F-statistic P-value**
Log[CO]	Log[PRGU]		87	0.55	0.12	<0.0001	0.31	0.79	0.33	0.30	10.13	<0.0001
		Fuel Type		-0.38	0.21	0.080	-0.81	0.046				
		Interaction 2		-0.10	0.21	0.62	-0.52	0.31				
		PRGU Pre		-0.0076	0.066	0.91	-0.14	0.12				

Note: Fuel Type refers to Pine=0, Mixed=1; Interaction 2 = CO 8-hour TWA x Fuel Type; PRGU Pre is the variable for log-transformed pre-shift propylguaiacol concentration (µg/mg creatinine

Table 6: Post-shift creatinine adjusted propylguaiacol without Substituted LOD/sqrt(2)

Post-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine) without Substituted LOD/sqrt(2)												
Log Transformed Predictor Variable	**Log Transformed Outcome Variable**	**Covariate**	**N**	**Coefficient**	**Std. Err.**	**Coefficient P-Value**	**95% CI**		**Model R^2**	**Adj. R^2**	**F-statistic**	**F-statistic P-value**
Log[CO]	Log[PRGU]		74	0.58	0.14	<0.0001	0.31	0.86	0.33	0.30	8.64	<0.0001
		Fuel Type		-0.41	0.25	0.10	-0.92	0.085				
		Interaction 2		-0.14	0.23	0.56	-0.59	0.32				
		PRGU Pre		-0.015	0.072	0.84	-0.16	0.13				

Table 7: Base Regression Models for Association between Exposure and Cross-Shift change in Urinary Methoxyphenols

Cross-Shift Creatinine Adjusted Methoxyphenols (µg/mg creatinine)										
Log Transformed Predictor Variable	**Log Transformed Outcome Variable**	**N**	**Coefficient**	**Std. Err.**	**Coefficient P-Value**	**95% CI**		**Model R2**	**F-statistic**	**F-statistic P-value**
PM2.5 8hr-TWA	Methylguaiacol	77	0.75	0.18	<0.00010	0.39	1.11	0.19	17.	<0.00010
	Ethylguaiacol	62	0.51	0.21	0.02	0.10	0.93	0.09	6.0	0.020
	Propylguaiacol	**70**	**0.91**	**0.20**	**<0.00010**	**0.52**	**1.3**	**0.24**	**22.**	**<0.00010**
	Ethylsyringol	59	0.30	0.35	-0.39	0.99	0.38	0.01	0.77	0.38
CO 8hr-TWA	Methylguaiacol	78	0.74	0.11	<0.0010	0.53	0.96	0.38	47.	<0.00010
	Ethylguaiacol	60	0.54	0.16	<0.00010	0.23	0.86	0.17	12.	<0.00010
	Propylguaiacol	**69**	**0.69**	**0.15**	**<0.00010**	**0.40**	**0.99**	**0.25**	**22**	**<0.00010**
	Ethylsyringol	58	0.20	0.22	-0.25	0.65	0.37	0.01	0.81	0.37
Levoglucosan 8-hr TWA	Methylguaiacol	77	0.37	0.09	<0.00010	0.19	0.56	0.18	16.	<0.00010
	Ethylguaiacol	62	0.20	0.11	0.070	-0.020	0.43	0.05	3.3	0.070
	Propylguaiacol	**70**	**0.41**	**0.11**	**<0.00010**	**0.20**	**0.62**	**0.18**	**15.**	**0.00030**
	Ethylsyringol	59	0.16	0.15	-0.15	0.46	0.3	0.02	1.1	0.30

Table 8: Multiple Linear Regression Models showing association between Mean 8-hour Carbon Monoxide Time Weighted Average Concentration and Cross-Shift change in Propylguaiacol (µg/mg creatinine)

Cross-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine)												
Predictor Variable	**Outcome Variable**	**Covariate**	**N**	**Coefficient**	**Std. Err.**	**Coefficient P-Value**	**95% CI**		**Model R^2**	**Adj. R^2**	**F-statistic**	**F-statistic P-value**
CO 8-hr TWA	**Propylguaiacol**		**69**	0.69	0.15	<0.0001	0.40	0.99	0.25	0.24	22	<0.0001
CO 8-hr TWA	**Propylguaiacol**		**68**	0.80	0.18	<0.0001	0.43	1.17	0.27	0.24	8.07	0.0001
		Fuel Type		-0.20	0.34	0.55	-0.88	0.47				
		Interaction 2		-0.42	0.30	0.14	-1.1	0.15				

Note: Fuel Type: Pine=0, Mixed=1; Interaction 2 = CO 8-hour TWA x Fuel Type

In addition to the post-shift regression models, cross-shift models were also developed to determine if there was an assocation between exposure variables and urinary methoxyphenol concentrations. The cross-shift base regression models were determined for similar compounds as the models based on the post-shift concentrations. However, these compounds were further limited by some of the higher number of censored values due to concentrations below minimum-reporting limits, as well as non-detection (especially in the pre-shift samples). Because many of the cross-shift samples were negative, they were censored from the regression analysis because they could not be log-transformed. Further, the correlation coefficients determined by the Pearson's correlation tables limited the compounds to include in the cross-shift base regression models to methylguaiacol, ethylguaiacol, propylguaiacol, and ethylsyringol. As with the post-shift samples, the carbon monoxide exposure variable exhibited the highest correlation coefficient values for the exposure variables. The base regression models based on cross-shift change are presented in Table 7.

Table 8 illustrates the difference is cross-shift concentrations and exposure, which can be compared to the models based on the post-shift concentrations and exposure. The models initially included other covariates to explain the interaction of wood smoke exposure and methoxyphenol concentrations, but each covariate did not elicit an association. The fuel type variable and interaction variable still present important features of the model, which is why they are included. Because the fuel types have been shown in previous studies to produce different combinations of urinary methoxyphenols, it was necessary to maintain these variables in the model to take into account the different fuel types. Nevertheless, in this particular model, fuel type and the interaction term [fuel type, CO] were not statistically significant.

Chapter Four: Conclusions

The first hypothesis stated that occupational exposure to wood smoke is associated with cross-shift changes in concentrations of methoxyphenol compounds, which can serve as viable biomarkers of wood smoke exposure. The results identify that occupational exposure to wood smoke is associated with increases in concentrations of individual biomarkers. Using cross-shift changes in urinary propylguaiacol concentration as the outcome of interest, we find that work shift exposure to CO is significantly associated with cross-shift change in urinary propylguaiacol concentration, specifically log transformed work shift exposure to CO explains 25% of the variability in the log transformed cross-shift change in urinary propylguaiacol concentration. Similarly, when using post-shift changes in urinary propylguaiacol concentration as the outcome of interest, we find that work shift exposures to CO is significantly associated with post-shift change in urinary propylguaiacol concentration, specifically log transformed work shift exposure to CO explains 33% of the variability in the log transformed post-shift change in urinary propylguaiacol concentration.

The post-shift urine void occurred immediately after the end of the shift, which in relation to the end of exposure, ranged from one to two hours. With a relatively short half-life of the methoxyphenol compounds in the body (~2-6 hours), it is possible that the half-life influenced the post-shift concentrations and ultimately the associations between wood smoke exposure and urinary propylguaiacol concentrations.

This result differs from the Dills et al. 2006 study, in that this study focused on CO as the exposure variable rather than $PM_{2.5}$ as was performed in the earlier study. The earlier study focused on the association of clusters of methoxyphenols with exposure. In this study, we explored associations with individual methoxyphenols.

Neitzel et al. 2009 reported associations between CO exposure and specific guaiacol compounds in urine. Both studies found that LG exhibited lower associations with individual compounds compared to CO. The results of the Neitzel study also indicated that CO as an exposure variable resulted in the highest R^2 values with methoxyphenols compared to LG or CO, which is something this study found as well. However, the R^2 value in the Neitzel study for CO exposure and propylguaiacol concentration was 0.59 compared to this study, which was 0.31.

In the current study, CO was used as a surrogate exposure variable for $PM_{2.5}$ and LG for many reasons. During a fire event, volatile organic compounds are created from combustion. However, during incomplete combustion from any source, CO is produced. Depending on the burn conditions, the fuel, and the intensity of the fire, as well as the stage of the fire, such as during a blaze compared to smoldering, the levels of CO will fluctuate. However, in the current study, CO is more specific to wood smoke than other sources because the levels from wood smoke are much higher than others source that could potentially confound any wood smoke exposure, such as exhaust from a vehicle. Therefore, CO serves as a viable surrogate of the other exposure variables because it is specific to combustion, which occurs during a fire event. Additionally, the Pearson's tables indicate that the highest correlations were observed between CO and other exposure variables and CO and the outcome variables (Refer to Pearson's tables, Appendix A24-A29).

$PM_{2.5}$ was not chosen primarily because the Pearson correlation coefficients were not as strong as CO. Additionally, $PM_{2.5}$ is not entirely specific to wood smoke. Even if total particulate matter were used as an exposure variable, it would contain some PM derived from dust and dirt, rather than wood smoke per se.

Levoglucosan was not chosen as the exposure variable because the correlations between other exposure variables and methoxyphenols were lower than the $PM_{2.5}$ or CO in many cases. Further, LG can be impacted by several factors such as the wood type, combustion features, and cellulose content in the wood (Bergauff et al. 2010).

The second hypothesis stated that vegetation type differentially affects the dose-response relationship between exposure to wood smoke and methoxyphenol concentrations. For this reason, the linear regression models included fuel type and an interaction term of fuel type multiplied by the exposure variable as covariates. The Pearson's correlation tables (refer to Appendix Tables A24-A29) suggest that urinary guaiacol compounds are more associated with pine vegetation while urinary syringol compounds are more associated with mixed vegetation. This assessment agrees with prior literature that investigated fuel type and methoxyphenol concentrations in subject groups (Dills et al. 2001, Dills et al. 2006, Neitzel et al. 2009). This is also consistent with the Fine et al. 2001 study, which demonstrated that syringol compounds are more abundant in hard wood smoke compared to soft wood smoke.

However, despite the initial results of the Pearson's correlation tables that would suggest an interaction between vegetation type and urinary methoxyphenol concentrations, ultimately this interaction was not witnessed, as indicated by the p-values for the fuel type and interaction term in the regression models, which were not statistically significant. The other potential covariate logCO/PM that was evaluated was found not to be significant in the multiple regression models, therefore was not included in the final models.

The data might not support the interaction, but we are not in a position to completely rule out fuel type and its interaction with the exposure variables, specifically CO. When the confidence interval for the fuel type and interaction term are considered, it indicates that the additional effect between CO and urinary propylguaiacol when coded for mixed versus pine vegetation could be greater. Additionally, preliminary studies suggest that fuel type influences concentrations of guaiacyl-type compounds and syringyl-type compounds. Much like Neitzel's studies, investigating the effects of clusters of methoxyphenols could demonstrate a greater association with CO exposure.

None of the models we developed effectively predicted biomarker concentrations based on the exposure variables as evidenced by the low R^2 values for the models. This is somewhat in contrast to the Neitzel study, which included multiple exposure variables and a composite outcome variable with an R^2 value of 0.75. Therefore, in the future, including LG and CO as predictor variables could increase the predictability of the model. Further, investigating clusters of methoxyphenols, such as guaiacol clusters and syringol clusters could potentially present a more viable option.

As a suggestion for future work, it would be interesting to conduct the same study in a different location that contained a higher proportion of mixed vegetation, or better still, hardwood vegetation compared to pine vegetation. The results of this study are in larger part specific to pine vegetation because pine dominated the burn area, producing predominantly guaiacol compounds. Considering that wildfires occur in many Western states, in addition to the southeastern part of the U.S. where the data was collected for this study, it would be appropriate to conduct a similar study on brushfire exposures, especially in places such as Nevada, Arizona, Utah, Texas, or other Western states. In this way, the results of the pine vegetation in this study could be compared to other vegetation types to verify our findings, which suggest that a difference exists in the concentrations of biomarkers in firefighters, associated with differing fuel

types. Further, subsequent studies in areas with different fuels could determine if a different biomarker of exposure should be sought for a specific type of vegetation. This would contribute greatly to the field of occupational exposures to wood smoke in wildland firefighters, because it could reveal if certain areas are prone to higher concentrations of one biomarker compared to others.

Another aspect for future study would involve investigating subject specific differences in urinary methoxyphenol concentrations in wildland firefighters. Considering that biomonitoring takes into account metabolism and higher rates of breathing, the concentration of biomarker in the firefighter could be greatly influenced by the amount of wood smoke that is inhaled into the lungs, given the particular activity. Understanding if greater physical activity and exertion could influence biomarker concentrations, could lead to a greater understanding of smokes exposures and differences in reaching permissible exposure limits.

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Appendices:

This section describes the number of work shifts that each firefighter worked and the number of consecutive work shifts that occurred.

Summary of Data Structure: Work Shift Duration, Pump Time, and Consecutive Work Shifts

Table A1: Number of Total and Consecutive Shifts Worked for Each Subject

Subject	Number of Shifts Worked	Did they Work Consecutive Shifts?	Number of Consecutive Shifts Worked
BIO 001	23	Y	10
BIO 002	21	Y	6
BIO 003	7	Y	1
BIO 004	3	N	0
BIO 005	5	N	0
BIO 006	14	Y	2
BIO 007	18	Y	5
BIO 008	6	Y	1
BIO 009	5	Y	1
BIO 010	3	N	0
BIO 011	5	Y	2
BIO 012	9	Y	2
BIO 013	1	N	0
BIO 014	12	Y	5
BIO 015	2	N	0
BIO 016	1	N	0
BIO 018	1	Y	1
Total	136	Y=11, N=6	36

Table A1 represents the number of shifts worked for each subject in the study as well as how many consecutive shifts the subjects worked. “Number of consecutive shifts” is a summation of the number of times one shift occurred the day following a previous shift. For instance, if a firefighter worked one shift on 2/8/2008, a consecutive shift would be regarded as a shift worked on 2/9/2008. It does not indicate how many days any one series of consecutive shifts lasted. This table indicates that the range of work shifts monitored for each subject is between one and twenty-three, while the number of consecutive work shifts ranges from zero to ten, depending on the subject.

Figure A1: Shift Length Duration by Pump Time and Work Shift Diary Entry

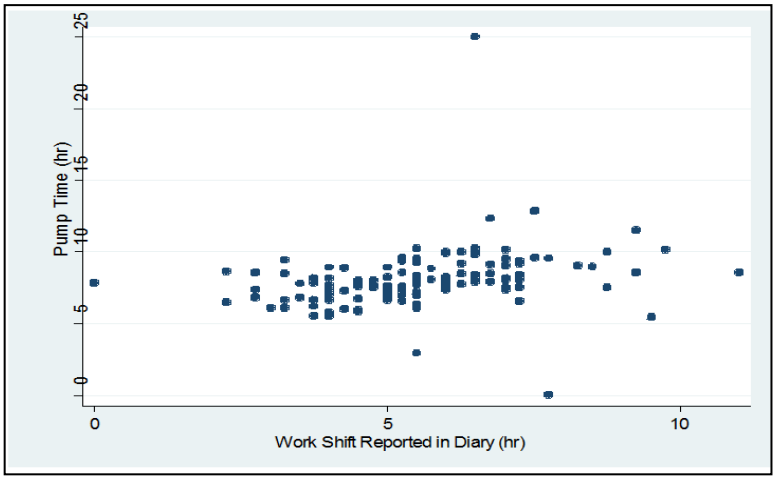


Figure A1 indicates that the shift duration based on the diary entry differed from the pump sampling time. Several of the work diary entries reported shifts of less than five hours, with one reported for zero hours. The work shift diaries convey an approximation of the shift duration, but they have limitations due to firefighters potentially reporting values that differ from the actual work shift durations.

Consecutive shifts were determined to understand if the exposure from one shift could influence the following shift's exposure. Although initially the consecutive shifts were considered for inclusion in the multiple linear regression models, ultimately including the pre-shift urinary concentration variable, or a cross-shift change in concentration in the model controlled for the consecutive shifts.

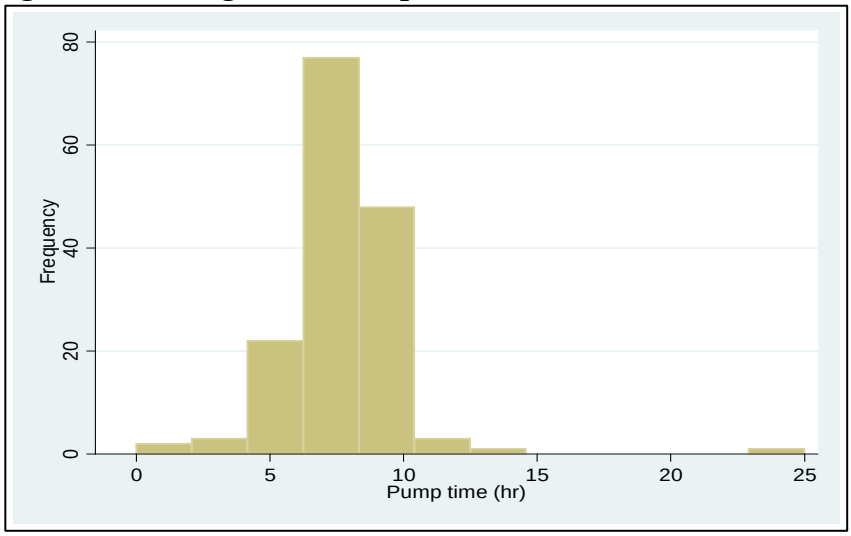
Table A2 summarizes the distribution of monitored work shifts for each subject across the two years of the study.

Table A2: Work Shift Matrix Stratified by Subject ID Including Consecutive Work Shifts

	Unique ID																
Date	BIO 001	BIO 002	BIO 003	BIO 004	BIO 005	BIO 006	BIO 007	BIO 008	BIO 009	BIO 010	BIO 011	BIO 012	BIO 013	BIO 014	BIO 015	BIO 016	BIO 018
1/16/08	1	1	1		1												
1/17/08	1	1				1	1										
1/30/08		1			1		1										
2/6/08		1	1	1		1	1	1									
2/16/08	1				1		1		1	1	1						
2/17/08	1	1					1				1	1					
2/18/08	1	1					1	1	1			1					
2/21/08	1	1	1			1	1		1								
2/22/08	1	1				1			1								
2/29/08	1	1		1		1		1				1					
3/12/08	1	1	1									1	1				1
3/13/08	1	1	1	1		1	1										
3/14/08	1						1					1					
3/15/08			1			1	1		1			1					
3/20/08		1	1		1	1	1					1					
3/27/08		1				1						1					
3/28/08		1						1				1					
1/11/09	1					1								1	1		
2/1/09	1	1					1							1			
2/10/09	1	1												1			
2/11/09	1						1							1			
2/17/09	1	1									1			1			
2/18/09	1	1									1			1			
2/26/09	1				1		1			1				1			
2/27/09	1						1							1			
2/28/09		1				1	1										
3/7/09						1				1							
3/10/09	1										1			1		1	
3/11/09		1					1							1			
3/12/09	1					1		1						1	1		
3/13/09	1					1		1									
3/20/09	1	1					1							1			

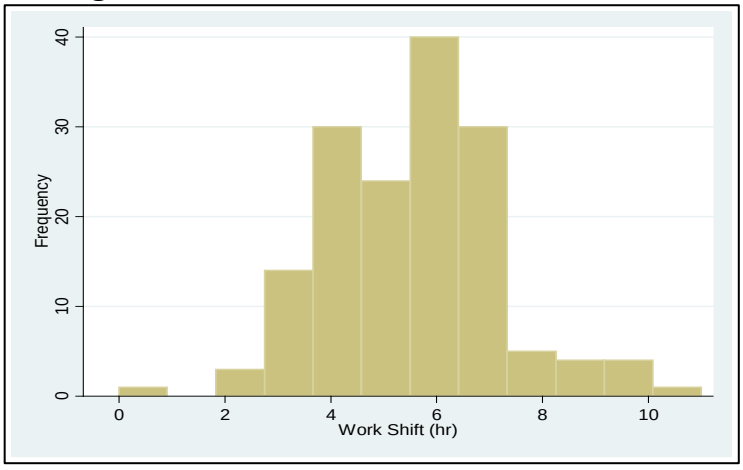
Work Shift by Pump Time

Figure A2: Histogram of Pump Time to Indicate Shift Duration



Figures A2 and A3 illustrate the distribution of work shift durations based on pump time (Figure A2) and workers diaries (Figure A3).

Figure A3: Histogram of Work Shift Duration Based on Work Diaries (hr)



The sampling pump time durations range from zero to twenty-five hours, with an average duration between six and ten hours. The pumps that reported work shift duration values of less than five hours can be attributed to a number of factors. One reason could be due potentially to days with small or no fires, leading to a truncated shift. Another factor could be that the pump failed, due to a flow fault. The field data logs might shed light on this issue, but will need to be investigated further.

The pump that measured twenty-five hours is not a realistic value for work shift duration. Besides a pump malfunction, the other potential reason for a value of twenty-five hours would be a pump that was left on after the end of the shift until the following shift, and was not accounted for in the work shift diary or sampling pump notes.

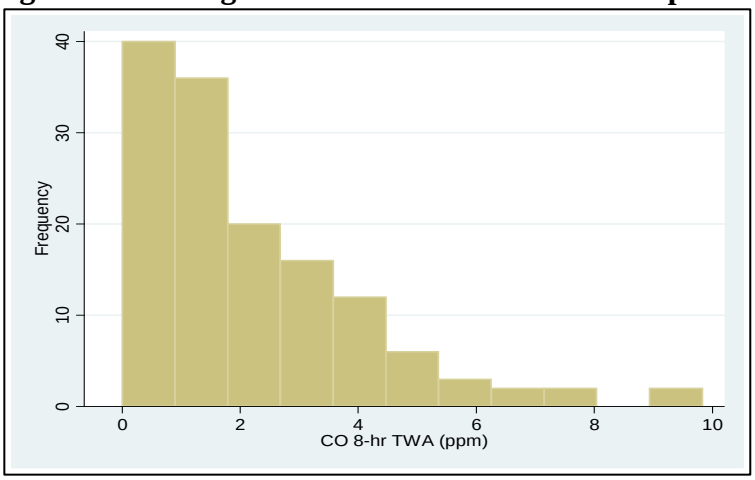
Overall, there is also much greater variation in the work diaries compared to the pumps. The pump time is approximately one to two hours greater on average than the work shift duration reported in the diaries. The reason that these two measures of work shift duration differ may be due to travel time between the fire site and the stage area where firefighters' personal sampling pumps were started and stopped. Some firefighters may have interpreted shift duration to mean only the time they spent at the fire site.

The sampling pumps provide a more reliable record of the actual work shift compared to the diaries, since each pump was manually started at the beginning of each work shift, and manually stopped at the end of each work shift. This allows for a defined interval with a known beginning and known stopping point based on the sampling pump's internal clock. Additionally, these values were provided in the field data logs. Considering that the smoke concentrations can be standardized to an eight-hour work shift, it seems more appropriate to use the pump times rather than the times reported in the diaries as the indicator of work shift. Unfortunately, if the pump was left on well after the end of the shift - as may have been the case with the one sample that recorded a shift duration of 25hr - it generates discrepancies in the data.

Further, if the pump was not turned on or if the pump failed, the shift duration information was no longer available. However, considering that the work shift diaries were not standardized for the whole group, it is difficult to determine the accuracy of the information as well as the interpretation of what the wildland firefighters constituted as a work shift, i.e. Work shift starts upon departure from the base or arrival at the field site. For these reasons, pump times were used for determining the shift times for wood smoke exposures in the data set.

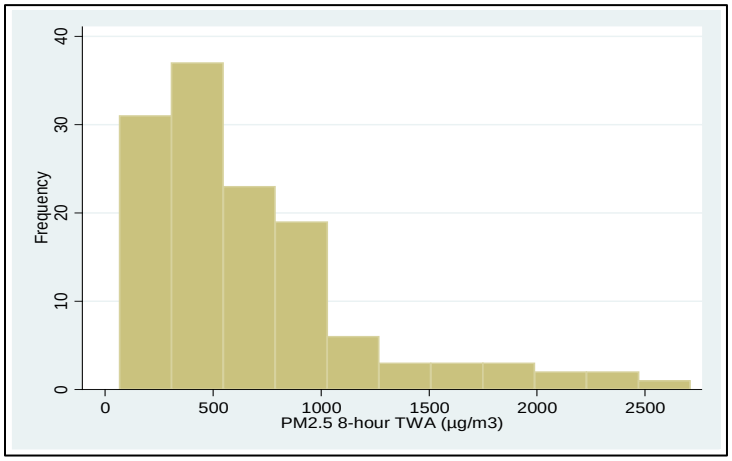
See the following figures for histograms of the 8-hr TWA exposures for CO, $PM_{2.5}$ and Levoglucosan.

Figure A4: Histogram of 8 Hour TWA for CO Exposure



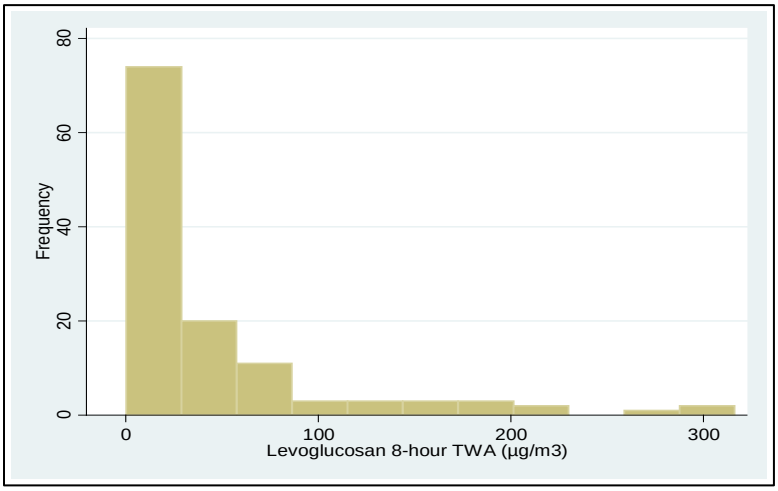
The 8-hour TWA for carbon monoxide is skewed to the right. The data indicates that the exposure was relatively low for most of the firefighters with concentrations ranging between 0.02 to 5 ppm, and approximately 80 of the samples resulting in concentrations less than 2 ppm. The OSHA PEL for carbon monoxide 8-hr TWA exposures is 50 ppm and the NIOSH REL is 35 ppm, indicating that all of the firefighters were well below the occupational exposure limit.

Figure A5: PM 2.5 8-hour TWA



The $PM_{2.5}$ 8-hr TWA is heavily skewed to the right, with mean exposures that range between 100 and 1000 µg/m^3. Approximately 65 subjects were exposed to concentrations that were 500 µg/ m^3 or less, while approximately 43 subjects were exposed to concentrations between 500 and 1000 µg/m^3. The OSHA PEL for particles is 15,000 µg/m^3, indicating that the subjects in this study were well below the OSHA limit.

Figure A6: Levoglucosan 8-hr TWA



The levoglucosan 8-hr TWA is heavily skewed to the right, with most of the subjects facing exposures between 0.05 and 50 µg/m^3. Approximately 75 of the subjects were exposed to 8-hour time weighted exposures of levoglucosan that measured 0-35 µg/m^3. Considering the LG represents a fraction of the $PM_{2.5}$ exposure, this indicates 2-5% of the $PM_{2.5.}$

Table A3: Comparison of Geometric Mean Exposure Variables, Dichotomized by Vegetation Type. Calculated using Two-Sample t-test with Unequal Variance

Exposure Var.	Vegetation Type	N	GM	GSE	GSD	[95% Conf. Interval]	
Log (CO) 8hr-TWA (ppm)	Mixed	39	0.041	0.16	1.01	-0.29	0.37
	Pine	79	0.61	0.12	1.06	0.37	0.85
	Combined	118	0.42	0.099	1.08	0.23	0.62
	Difference = Mean(Mixed)-Mean(Pine)		-0.57	0.20		-0.97	-0.17
	P-value = 0.0059		t = -2.8				
Log(PM2.5) 8hr-TWA (µg/m3)	Mixed	36	6.1	0.12	0.70	5.9	6.4
	Pine	78	6.4	0.076	0.67	6.3	6.6
	Combined	114	6.3	0.065	0.69	6.2	6.4
	Difference = Mean(Mixed)-Mean(Pine)		-0.29	0.14		-0.57	-0.0098
	P-value = 0.043		t = -2.1				
Log(LG) 8hr-TWA (µg/m3)	Mixed	35	2.50	0.22	1.27	2.1	3.0
	Pine	73	3.3	0.18	1.51	2.9	3.6
	Combined	108	3.0	0.14	1.48	2.8	3.3
	Difference = Mean(Mixed)-Mean(Pine)		-0.73	0.28		-1.3	-0.17
	P-value = 0.0109		t = -2.6				
Log(LG/PM) Ratio	Mixed	40	0.41	0.029	0.18	0.35	0.47
	Pine	81	0.50	0.024	0.22	0.46	0.55
	Combined	121	0.47	0.019	0.21	0.44	0.51
	Difference = Mean(Mixed)-Mean(Pine)		-0.093	0.038		-0.17	-0.018
	P-value = 0.016		t = -2.5				

Note: Log(LG/PM) was determined by taking the log(8-hr LG TWA) and dividing it by log(8-hr PM2.5 TWA) rather than calculating the log of the original LG/PM ratio.

As shown in Table A3, vegetation type has a significant influence on CO exposures. Similarly, vegetation type has a significant influence on $PM_{2.5}$ and LG exposures.

Methoxyphenol Distribution of Concentrations

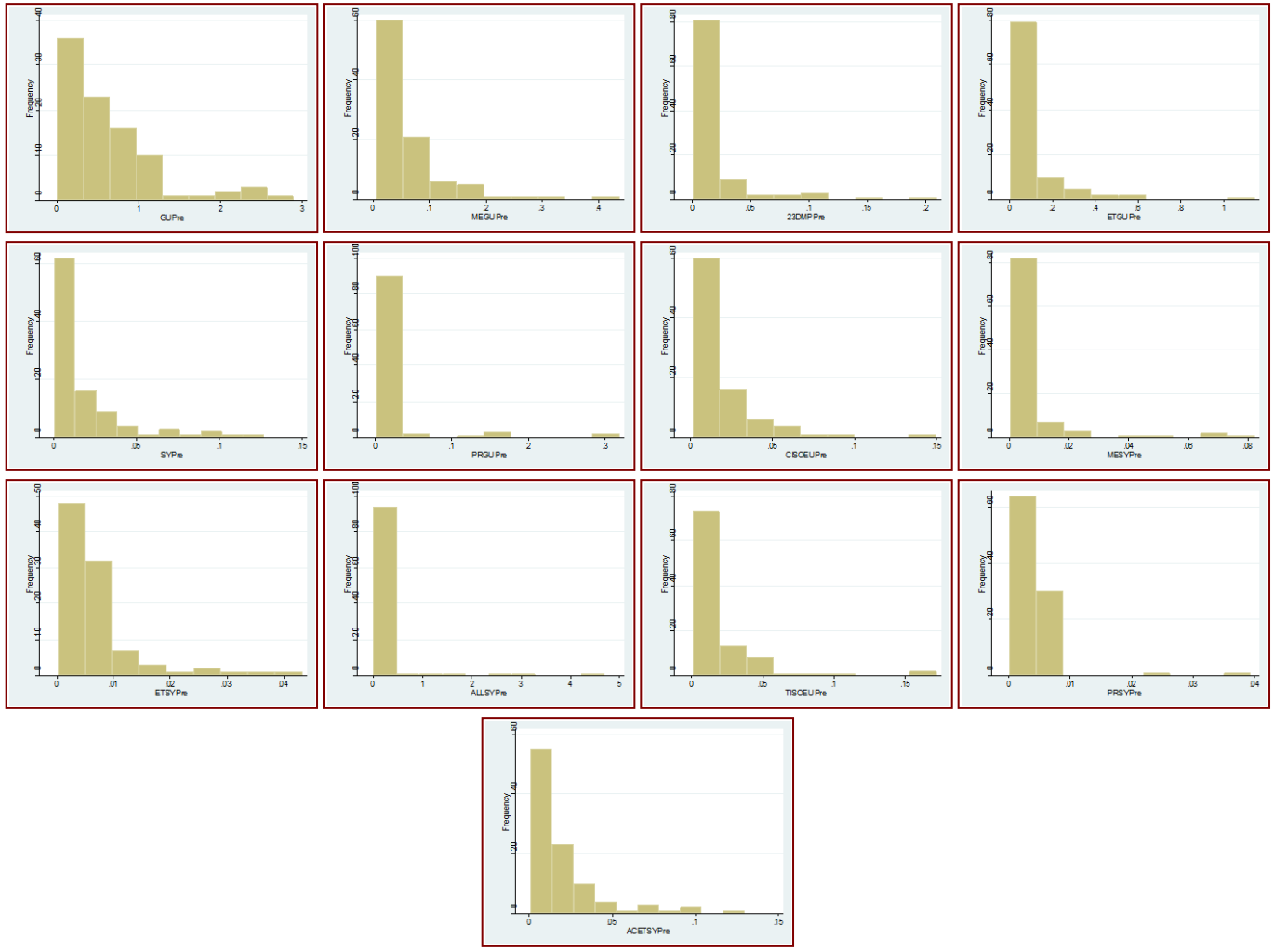
This next section investigates the different histograms of the methoxyphenol concentration distributions.

As tables A7-A10, all of the post-, pre- and cross-shift methoxyphenol compounds are skewed to the right and do not present a symmetric distribution. This indicates that they should be normalized before being added to the regression model.

Unlike the pre- and post-shift methoxyphenol compounds, many of the cross-shift methoxyphenols are normally distributed. The normally distributed methoxyphenol compounds include guaiacol, methylguaiacol, 2,3-dimethylguaiacol, ethylguaiacol, propylguaiacol, and acetylsyringone. The other compounds include cis-isoeugenol, trans-isoeugenol, syringol, methylsyringol, ethylsyringol, propylsyringol, and allylsyringol and are skewed. This indicates that they should be log-transformed for the regression analysis. The cross-shift compounds that initially were normally distributed also are log-transformed to maintain uniformity in the interpretation of the cross-shift compound.

Figures A7(a-m): Post-Shift Methoxyphenol Concentration Histograms

Figures A8(a-m): Pre-Shift Methoxyphenol Concentration Histograms



Figures A9(a-m): Cross-Shift Methoxyphenol Concentration Histograms

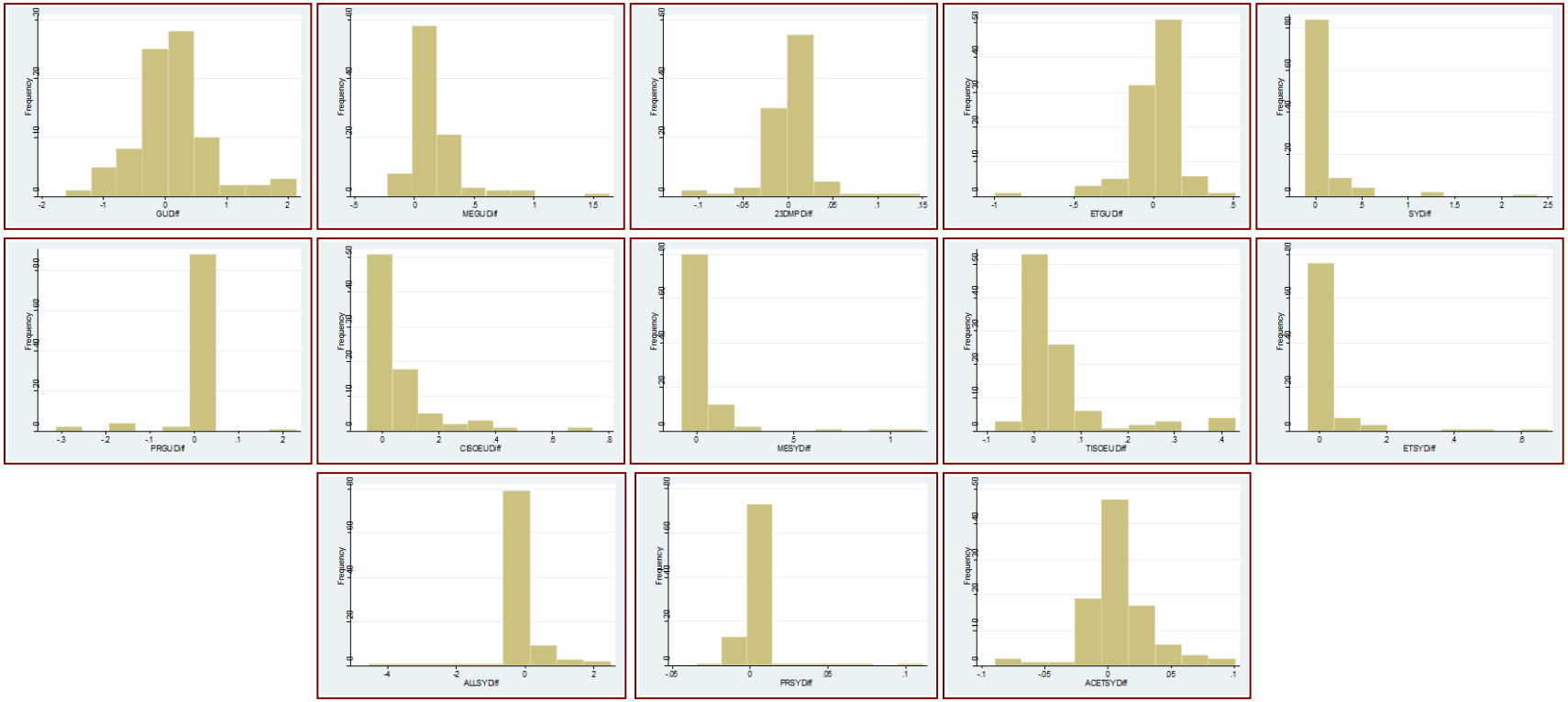


Figure A10(a-m): Log-Transformed Post-Shift Methoxyphenol Histograms

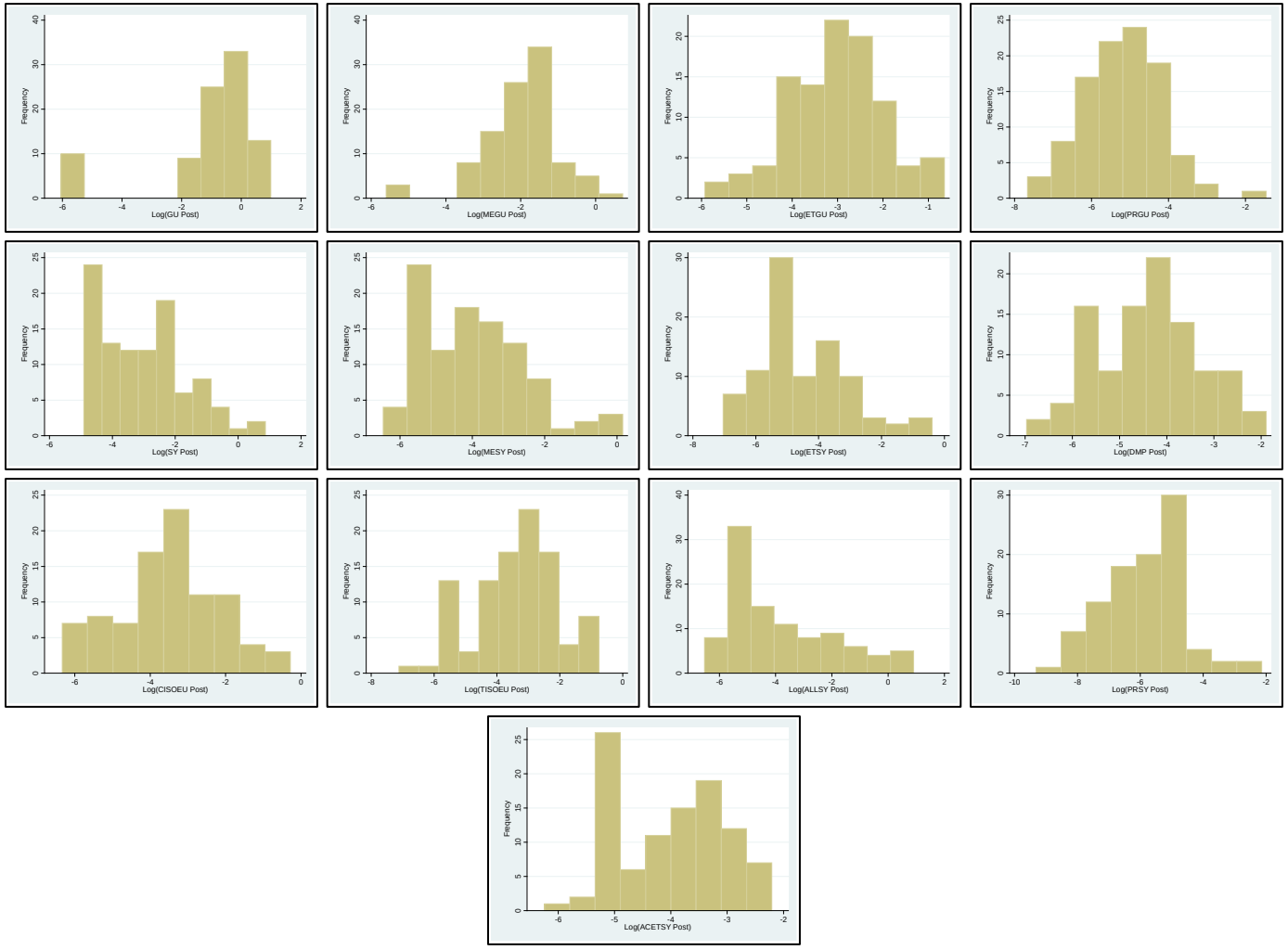


Figure A11(a-m): Log-Transformed Pre-Shift Methoxyphenol Histograms

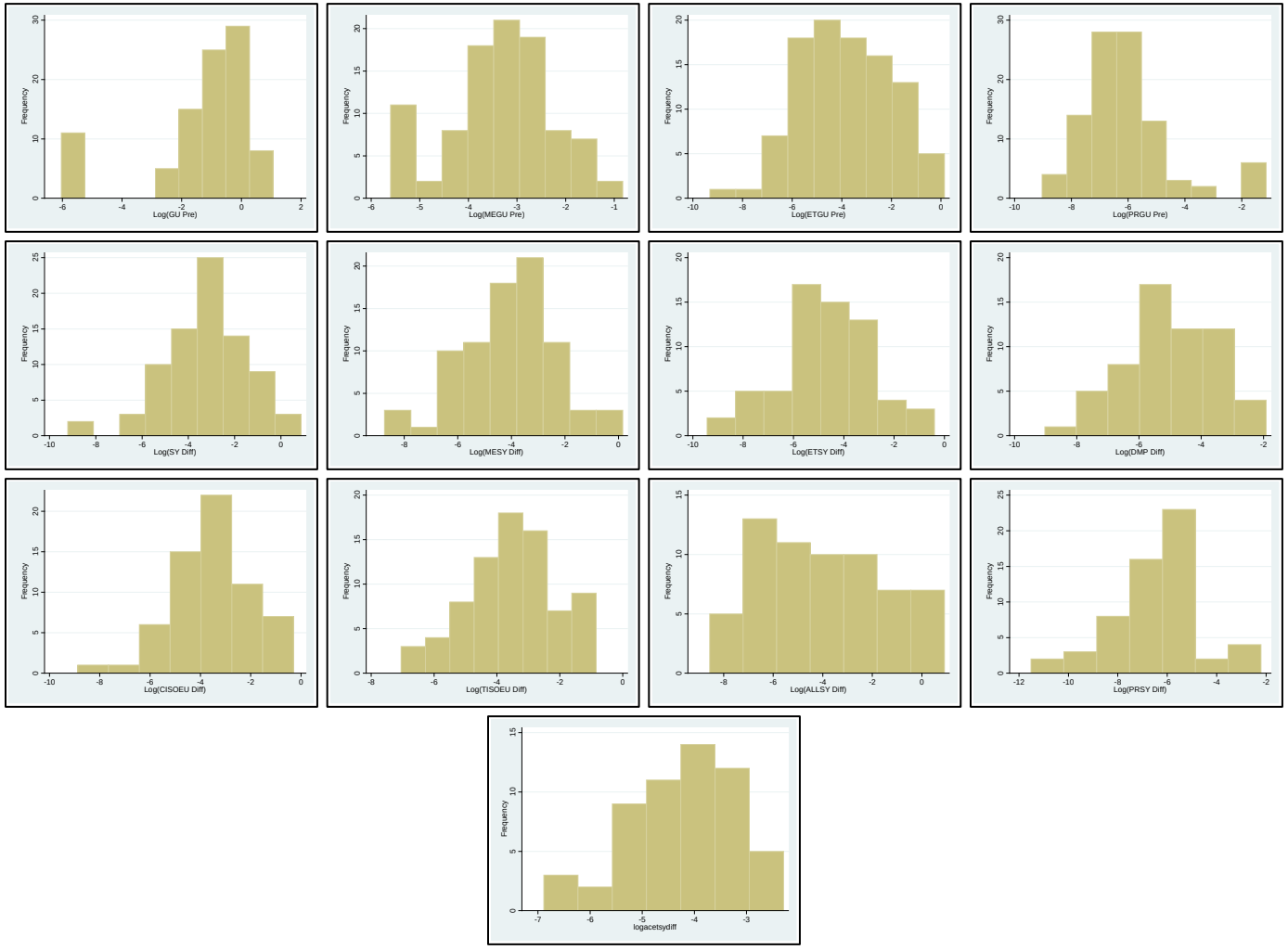
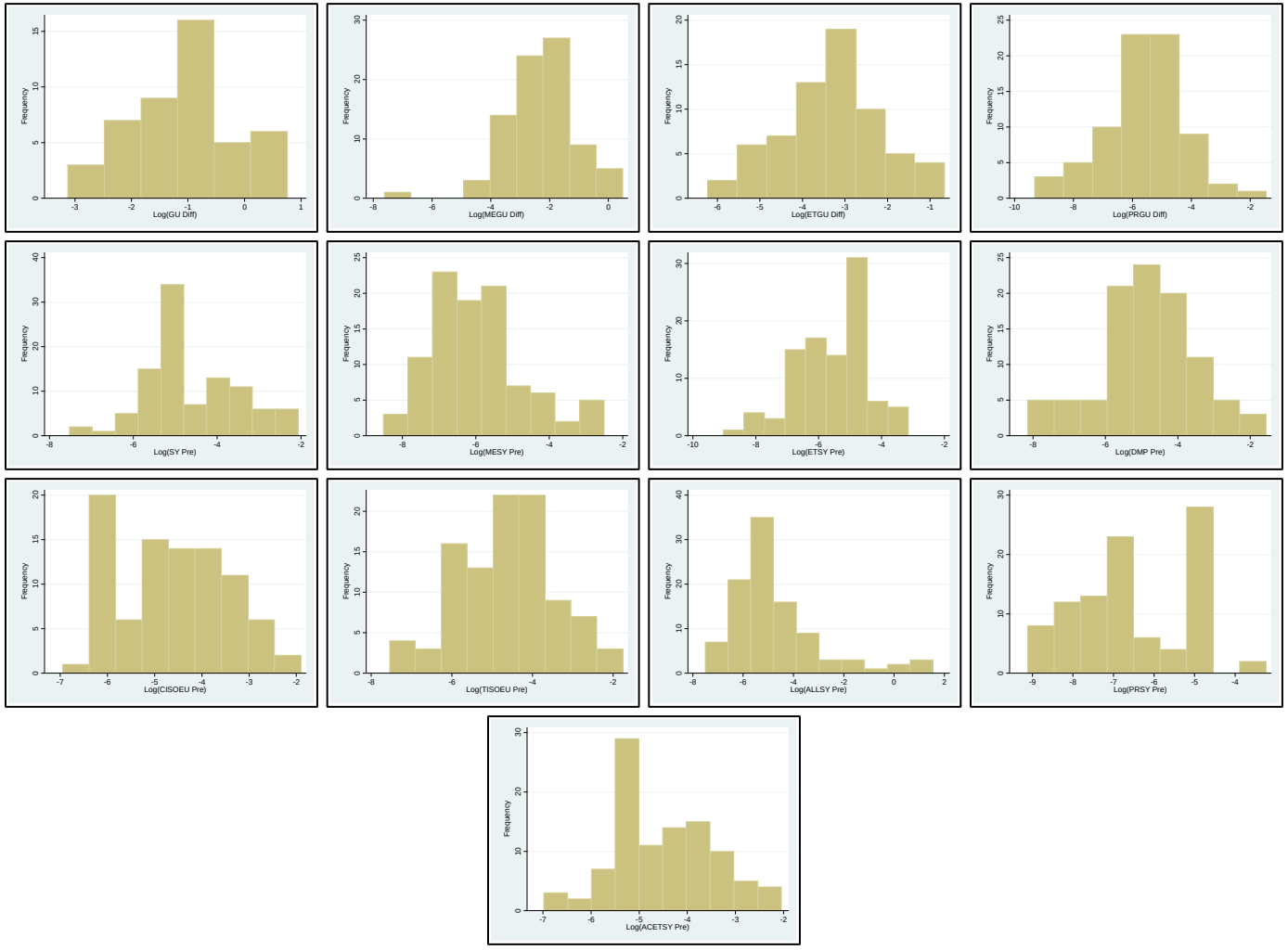


Figure A12(a-m): Log-Transformed Cross-Shift Methoxyphenol Histograms



Methoxyphenol Chemical Censoring Explanations

This section discusses the reasons for why compounds were censored during chemical analysis as well as the frequency of analytes that were censored.

Table A4: Explanation of Methoxyphenol Censored and Non-Censored Data Concerning Creatinine Concentrations and Air Data

Creatinine Too Low	10
No Air Data	14
No Urine Data	19
Neither Urine Nor Air Data	13
Valid Data	101
Total Data	157

This table explains how the data was censored prior to creatinine corrections. Of the 157 total samples, which accounted for both exposure and post-exposure samples, 56 were censored. The reasons for censoring included no air or urine data or both, or the creatinine factor was too low to allow for an accurate analysis of the compounds. 101 of 157 were analyzed after creatinine corrections were performed.

Table A5: Percentage of Methoxyphenol Compounds Censored for Analytical Reasons Based on Information in Tables 24 and 26

Compound	Percentage Censored for Post Shift	Percentage Censored for Cross Shift
Guaiacol	9	25
Methylguaiacol	3	17
2,3-Dimethoxyphenol	9	18
Ethyl Guaiacol	0	0
Syringol	16	33
Propylguaiacol	3	20
Cis-Isoeugenol	6	27
Methylsyringol	15	35
Trans-Isoeugenol	12	36
Ethylsyringol	12	22
Allylsyringol	25	52
Propylsyringol	23	47
Acetosyringone	20	46

*Note: Highlighted Values Refer to Analytes with Low Number of Censored Compounds

Analytical reasons that could lead to a compound being censored are listed in Table A4. Censorship was performed for recovery outside quality assurance limits, invalid reference retention times, and especially non-detection or concentrations below the minimum reporting limit. The frequency of non-detection or compounds that reported concentrations below the minimum reporting limit was much higher than other censoring reasons. Frequency of censored data is shown in Table A5.

Table A6 details the censorship statistics for the post-shift samples after creatinine correction. The majority of the compounds were censored due to non-detection or concentrations that were below the minimum-reporting limit. The highest censored compounds were allylsyringol, propylsyringol, and acetylsyringone, while the lowest censored compounds were ethylguaiacol, methylguaiacol, and propylguaiacol.

Table A7 details the censorship for the pre-shift samples after creatinine correction. The majority of the compounds were censored due to non-detection or concentrations that were below the minimum-reporting limit. The highest censored compounds were propylsyringol, and cis-isoeugenol, while the lowest censored compounds were ethylguaiacol, and 2,3-dimethylguaiacol.

Table A8 details the censorship for the cross-shift samples after creatinine correction. The majority of the compounds were censored due to non-detection or concentrations that were below the minimum-reporting limit. The highest censored compounds were propylsyringol, allylsyringol, guaiacol, ethylsyringol, cis-isoeugenol, and acetylsyringone, while the lowest censored compounds was ethylguaiacol.

Table A6: Determination of Censored Variables for the Post-Shift Creatinine Corrected Data

Censored Post-Shift Data Explanation								
Compound	Total Samples	AI	ND or MR	RY	RT	AX	Total Censored	Total Uncensored
GU	101	5	10	2	3	1	21	80
MEGU	101	1	3	0	0	0	4	97
23DMP	101	0	9	0	0	0	9	92
ETGU	101	0	0	0	0	0	0	101
SY	101	0	16	0	0	0	16	85
PRGU	101	1	3	0	0	0	4	97
CISOEU	101	1	6	0	9	0	16	85
MESY	101	0	15	0	0	0	15	86
TISOEU	101	1	12	0	0	0	13	88
ETSY	101	8	12	0	1	0	21	80
ALLSY	101	0	25	0	2	0	27	74
PRSY	101	1	23	1	3	0	28	73
ACETSY	101	0	20	1	0	1	22	79

Note: AI: Refers to the area not being valid due to an interference during GC-MS analysis; **ND or MR:** Refers to chemical non-detection or below the minimum reporting limit; **RY:** Recovery outside quality assurance limits or was indeterminate during analysis; **RT:** Reference Retention Time is invalid (0, error, or blank); **AX:** Response likely exceeds detector saturation limit or area not valid due to other causes

Table A7: Determination of Censored Variables for the Pre-Shift Creatinine Corrected Data

Censored Pre-Shift Data Explanation								
Compound	Total Samples	AI	ND or MR	RY	RT	AX	Total Censored	Total Uncensored
GU	101	6	11	2	0	0	19	82
MEGU	101	4	11	1	0	0	16	85
23DMP	101	0	7	1	1	0	9	92
ETGU	101	0	0	0	2	0	2	99
SY	101	0	13	1	0	0	14	87
PRGU	101	1	13	1	3	0	18	83
CISOEU	101	0	15	1	11	0	27	74
MESY	101	3	11	1	0	0	15	86
TISOEU	101	0	17	1	1	0	19	82
ETSY	101	2	13	1	2	0	18	83
ALLSY	101	0	16	1	0	0	17	84
PRSY	101	2	34	0	3	0	39	62
ACETSY	101	1	19	0	0	0	20	81

Table A8: Determination of Censored Variables for the Difference of Cross-Shift Creatinine Corrected Data

Censored (Cross Shift) Data Explanation								
Compound	Total Samples	AI	ND or MR	RY	RT	AX	Total Censored	Total Uncensored
GU	101	9	17	4	3	1	34	67
MEGU	101	5	14	1	0	0	20	81
23DMP	101	0	15	1	1	0	17	84
ETGU	101	0	0	0	2	0	2	99
SY	101	0	25	1	0	0	26	75
PRGU	111	2	16	1	3	0	22	79
CISOEU	101	1	18	1	14	0	34	67
MESY	101	3	25	1	0	0	29	72
TISOEU	101	1	26	1	1	0	29	72
ETSY	101	9	22	1	3	0	35	98
ALLSY	101	0	34	1	1	0	36	65
PRSY	101	2	47	1	5	0	55	99
ACETSY	101	1	31	1	0	0	33	68

This section details some of the statistical analyses that were performed to determine if any of the variables would confound the association between the exposure variables and the methoxyphenols.

Exposure to Cigarette Smoke

The results of the research indicates that guaiacol compounds form in cigarette smoke, which then are inhaled into the body. During wood smoke exposure, guaiacols are formed from the pyrolysis of lignin. This makes cigarette smoke a potential confounder of wood smoke exposure Therefore, a Student's t-test was performed to determine if smoking effects guaiacol concentrations in the cohort of firefighters for this study.

Table A9: Number of Smokers in Firefighter Cohort

Variable: Did you smoke?	Freq.	Percent
No	129	98.47
Yes	2	1.53
Total	131	100

The results represented in Table A9 demonstrate that only two subjects smoked during the study.

Table A10: Number of Cigarettes Smoked

Unique ID	Did_you_smoke?	If_yes,_how_many cigarettes?	
BIO 008	Yes	1	-
BIO 013	Yes	-	6

As shown in Table A10 subject# BIO 008 smoked one time during the study, while subject# BIO 013 smoked six times.

Table A11: Frequency of Secondhand Smoke

Around_smokers?	Freq.	Percent	Cum.
No	102	77.86	77.86
Yes	29	22.14	100
Total	131	100	

There is a total of 29 firefighter*shifts of secondhand cigarette smoke. Firefighter*shifts refers to the number of instances the firefighters in the study were exposed to secondhand smoke for a work shift. Although there are only 18 subjects, some of these subjects

received secondhand smoke multiple times. This can be interpreted as stating if one firefighter was exposed to secondhand smoke on one day and secondhand smoke on another day, there would be a total of two firefighter*shifts of secondhand smoke.

Table A12: Number of times Around Smokers Based on Time Intervals

Unique ID	Around_smokers?	Number of Times around Smokers (min) based on time intervals			
		>5 min	10 min	20 min	120 min
BIO 001	Yes		2		
BIO 002	Yes	4	1		
BIO 003	Yes		4		
BIO 006	Yes	1	2	1	
BIO 007	Yes	7			
BIO 011	Yes				1
BIO 012	Yes	3			
BIO 013	Yes				

Table A12 explains which subjects were around other subjects who smoked during the study as well as the interval of time they were with subjects who smoked. There were 15 instances in which non-smokers were around a subject who smoked for less than 5 minutes, while nine subjects were near to other subjects who smoked for approximately 10 minutes. One subject was exposed to secondhand smoke for 20 minutes and another for 120 minutes. BIO 013 reported an exposure for 600 minutes, but this was considered unlikely.

Table A13: Frequency of Smoking and Exposure to Second-Hand Smoke

Around_smokers?	No		Yes	
Did_you_smoke?	No	Yes	No	Yes
Unique ID				
BIO 001	21	0	2	0
BIO 002	14	0	6	0
BIO 003	4	0	4	0
BIO 004	3	0	0	0
BIO 005	5	0	0	0
BIO 006	9	0	4	0
BIO 007	10	0	7	0
BIO 008	5	1	0	0
BIO 009	5	0	0	0
BIO 010	2	0	1	0
BIO 011	3	0	1	0
BIO 012	4	0	3	0
BIO 013	0	0	1	1
BIO 014	12	0	0	0
BIO 015	2	0	0	0
BIO 016	1	0	0	0
BIO 018	1	0	0	0
Total Number	101	1	27	1

Table A13 summarizes by subject people that smoked during the sampling period as well as the subjects who were near to the subjects who smoked.

When secondhand smoke is considered, similar results are obtained. No association exists between secondhand smoke and guaiacol or methylguaiacol concentrations in wildland firefighters. The duration of exposure to secondhand smoke is less than or equal to ten minutes for twenty-four of the twenty-six instances that this occurred, compared to wood smoke exposures that ranged from 5-8 hours. Based on the ratio of wood smoke exposure to cigarette exposure, the likelihood of cigarette smoke inhalation contributing significantly to the concentration of guaiacols and guaiacol derivatives in the urine seems unlikely.

Table A14: Comparison of Post-shift Geometric Mean Methoxyphenol Concentrations in Smokers and Non-Smokers.*

Variable	Smoked?	N	Mean	[95% Conf. Interval]		P-value
Log(GU Post)	No	87	-1.16	-1.5	-0.69	0.398
	Yes	2	0.06	-7.51	7.63	
Log(MEGU Post)	No	97	-2.00	-2.21	-1.79	0.987
	Yes	2	-1.99	-8.95	4.98	
Log (PRGU Post)	No	99	-5.19	-5.39	-4.98	0.9272
	Yes	2	-5.25	-10.1	-0.4	
Log(MEGU Cross)	No	80	-2.25	-2.52	-1.99	0.7254
	Yes	2	-2.56	-16.55	11.44	
Log (PRGU Cross)	No	73	-5.57	-5.89	-5.24	0.1933
	Yes	2	-6.87	-17.5	3.77	

* **Used student's t-test with equal variance**

Given the relatively short duration of exposure two cigarette smoke reported by the two subjects who smoked, the inhaled dose of cigarette smoke compared to wood smoke on the two occasions would likely have been much smaller in comparison to the exposure to wood smoke.

For this particular study, it appears that cigarette smoking did not contribute to the exposure-biomarker relationship in any significant way. However, smoking still presents a major potential confounder and in future studies should be controlled if similar associations between wood smoke and urinary methoxyphenols are to be determined (Arnarp et al. 1989, Dills et al. 2001).

Grilled Food Confounding

Grilled foods can contain a variety of guaiacol and syringol compounds, either due to condensation of smoke onto the food during cooking, or deliberate addition of smoke flavoring agents (Dills et al. 2001, 2006, Neitzel et al. 2009). Thus it was necessary to examine whether consumption of smoked or grilled foods was a potential confounder of the association between occupational exposure to wood smoke and urinary methoxyphenols in this study.

Table A15: Grilled Food Consumption

Grilled Food Consumed	Freq.	Percent	Cum.
No	110	89.43	89.43
Yes	13	10.57	100
Total	123	100	

Table A15 indicates the frequency of subject*days for grilled food consumption. Consumption of grilled food was reported in association with thirteen of the 123 work shifts monitoring during the study.

Table A16: Grilled Food Frequency by Subject

Grilled Food	Freq.
BIO 001	1
BIO 002	5
BIO 006	2
BIO 009	2
BIO 012	1
BIO 014	1
BIO 015	1

Table A16 further demonstrates the frequency of grilled food consumption with stratification by subject. The grilled food consumption ranged from one to five times for each subject, with BIO 002 having the highest frequency of consumption of grilled foods.

Table A17 represents the results of the Student's t-tests that were performed to determine if consumption of grilled food confounds concentrations of urinary methoxyphenols in firefighters from wood smoke exposure. This may be due to a low number of subjects ingesting grilled foods, similar to subjects that smoked, where there were only two subjects who smoked during the sampling period. The low number of instances compared to the high number of non-instances makes it difficult to determine if grilled foods actually could confound this study. A previous pilot study by Dills et al. 2001 determined that grilled foods could potentially confound a dose-dependent relationship of urinary methoxyphenols and wood smoke exposure (Dills et al. 2001). Therefore, in the current study measures were taken to prevent ingestion of grilled foods during the duration of the study in the event that the wood smoke exposure could be confounded by grilled or smoked food exposure.

Table A17: Comparison of Geometric Mean Methoxyphenol Concentration in Urine Samples for Subjects that Consumed Grilled Foods and Subjects that Did Not Consume Grilled Foods*

Variable	Consumed Grilled Foods	N	Mean	[95% Conf. Interval]		P-Value
Log(GU Post)	No	82	-1.11	-1.5	-0.68	0.54
	Yes	5	-0.56	-1.5	0.40	
Log(MEGU Post)	No	88	-1.97	-2.2	-1.7	0.72
	Yes	7	-2.12	-2.7	-1.5	
Log(SY Post)	No	89	-3.14	-3.4	-2.8	0.55
	Yes	7	-2.81	-4.0	-1.7	
Log(CISOEU Post)	No	80	-3.5	-3.8	-3.2	0.92
	Yes	6	-3.56	-4.2	-2.9	
Log (PRGU Post)	No	89	-5.16	-5.4	-4.9	0.67
	Yes	7	-5.33	-5.9	-4.7	
Log(MEGU Cross)	No	74	-2.16	-2.4	-1.9	0.43
	Yes	4	-2.59	-3.5	-1.7	

*** Student's t-test with Equal Variance used for comparison**

Table A18: Smoked Food Consumption

Smoked Food	Freq.	Percent	Cum.
No	103	81.75	81.75
Yes	23	18.25	100

Table A18 indicates the frequency of subject*days of smoked food consumption. Smoked foods can confound methoxyphenol concentrations since both guaiacol and syringol compounds are present in smoked food. Therefore, Student's t-tests were performed to determine if there was a significant difference in estimated geometric mean methoxyphenol concentrations for subjects that ate smoked foods and subjects that did not.

Table A19: Smoked Food Consumption Frequency by Subject

Smoked Food	Freq.
BIO 001	4
BIO 002	11
BIO 003	1
BIO 004	1
BIO 006	1
BIO 007	2
BIO 009	2
BIO 012	1

Table A19 reflects the frequency of smoked food consumption. BIO 002 consumed smoked food 11 times, which was higher than any of the other subjects, which ranged from 1-4 instances.

Table A20 suggests that although there are twenty-three instances of smoked food consumption, which constitutes 18.25% of the sampling group, the association between concentrations of methoxyphenol compounds and smoked food exposure is not significant. This could be due to the low number of subject instances of smoked food exposure compared to the high number of wood smoke exposures.

Table A20: Comparison of Geometric Mean Methoxyphenol Concentration in Subjects that Consumed Smoked Foods and Subjects that Did Not Consume Smoked Foods*

Variable	Ate Smoked Foods?	Obs	Mean	[95% Conf. Interval]		P-value
Log(GU Post)	No	80	-1.1	-1.5	-0.63	0.83
	Yes	8	-1.2	-2.9	0.50	
Log(MEGU Post)	No	84	-2.0	-2.2	-1.8	0.58
	Yes	14	-1.8	-2.2	-1.5	
Log (PRGU Post)	No	84	-5.2	-5.4	-5.0	0.77
	Yes	15	-5.1	-5.5	-4.7	
Log(SY Post)	No	85	-3.1	-3.4	-2.8	0.94
	Yes	14	-3.1	-3.7	-2.6	
Log(MESY Post)	No	85	-4.0	-4.3	-3.7	0.85
	Yes	14	-4.1	-4.7	-3.5	
Log(MEGU cross-shift)	No	70	-2.1	-2.4	-1.9	0.27
	Yes	11	-2.5	-3.1	-2.0	

* Student's t-test with Equal Variance used

Spearman's and Pearson's Correlation Tables

This next section looks at the correlations between exposure variables and methoxyphenols using Spearman's and Pearson's correlation tables

Table A21-23 lists the Spearman Rho coefficients for each of the exposure variables and pre-shift (Table A21), post-shift (Table A22) and cross-shift (Table A23) methoxyphenol compounds and states which compounds correlate strongly compared to compounds that do not correlate as strongly. Generally, methylguaiacol, ethylguaiacol, cis-isoeugenol, and propylguaiacol correlate relatively strongly with each other. Post-shift and cross-shift concentrations of several of these compounds also had relatively good correlations with CO.

Table A21: Spearman Correlation of Pre-Shift Variables

	PM 2.5 (µg/m3)	CO (ppm)	LG (µg/m3)	GU PRE	MEGU PRE	DMP PRE	ETGU PRE	SY PRE	PRGU PRE	CISOEU PRE	MESY PRE	TISOEU PRE	ETSY PRE	ALLSY PRE	PRSY PRE
CO (ppm)	0.75														
LG (µg/m3)	0.58	0.54													
GU PRE	0.17	-0.025	0.031												
MEGU PRE	0.16	0.019	0.15	0.29											
DMP PRE	0.17	0.012	0.27	0.58	0.38										
ETGU PRE	0.14	-0.093	0.14	0.54	0.36	0.60									
SY PRE	0.018	-0.23	-0.18	0.48	0.26	0.44	0.56								
PRGU PRE	0.057	0.12	-0.014	0.086	0.091	-0.11	0.19	0.11							
CISOEU PRE	0.032	-0.072	-0.028	0.16	0.30	0.15	0.034	0.14	0.10						
MESY PRE	0.071	-0.16	-0.19	0.23	0.36	0.20	0.19	0.59	0.18	0.56					
TISOEU PRE	0.16	0.0089	0.15	0.490	0.47	0.45	0.44	0.35	0.13	0.52	0.37				
ETSY PRE	0.077	-0.095	0.0046	0.25	0.14	0.27	0.56	0.64	0.2	0.21	0.49	0.28			
ALLSY PRE	0.060	0.014	-0.15	0.057	0.12	-0.033	-0.0024	0.029	0.34	0.22	0.20	0.37	-0.11		
PRSY PRE	0.022	-0.046	-0.24	-0.030	-0.0097	-0.048	0.064	0.31	0.36	-0.10	0.098	-0.14	0.34	0.17	
ACETSY PRE	0.13	-0.079	0.28	0.093	0.29	0.48	0.51	0.12	0.15	0.30	0.15	0.32	0.21	0.060	-0.092

Table A22: Spearman Correlation of Post-Shift Variables

	PM 2.5 (µg/m3)	CO (ppm)	LG (µg/m3)	GU POST	MEGU POST	DMP POST	ETGU POST	SY POST	PRGU POST	CISOEU POST	MESY POST	TISOEU POST	ETSY POST	ALLSY POST	PRSY POST
CO (ppm)	0.68														
LG (µg/m3)	0.43	0.55													
GU POST	0.27	0.19	0.33												
MEGU POST	0.46	0.55	0.44	0.58											
DMP POST	0.11	0.10	0.22	0.68	0.51										
ETGU POST	0.38	0.43	0.35	0.63	0.85	0.57									
SY POST	0.14	0.14	0.19	0.48	0.45	0.50	0.50								
PRGU POST	0.51	0.62	0.44	0.45	0.91	0.43	0.84	0.47							
CISOEU POST	0.41	0.45	0.44	0.55	0.71	0.34	0.70	0.55	0.75						
MESY POST	0.20	0.14	0.26	0.38	0.48	0.41	0.47	0.77	0.47	0.43					
TISOEU POST	0.40	0.47	0.44	0.57	0.80	0.46	0.74	0.52	0.81	0.83	0.54				
ETSY POST	0.27	0.18	0.25	0.36	0.51	0.45	0.54	0.78	0.57	0.55	0.82	0.55			
ALLSY POST	0.23	0.19	0.11	0.30	0.33	0.37	0.31	0.22	0.26	0.22	0.20	0.38	0.20		
PRSY POST	0.16	0.22	0.11	0.26	0.49	0.36	0.57	0.54	0.55	0.42	0.51	0.39	0.57	0.17	
ACETSY POST	0.020	0.11	0.28	0.06	0.09	0.34	0.12	0.30	0.16	0.040	0.43	0.30	0.41	0.37	0.040

Table A23: Spearman Correlation of Cross-Shift Variables

	PM 2.5 (µg/m3)	CO (ppm)	LG (µg/m3)	GU CROSS	MEGU CROSS	DMP CROSS	ETGU CROSS	SY CROSS	PRGU CROSS	CISOEU CROSS	MESY CROSS	TISOEU CROSS	ETSY CROSS	ALLSY CROSS	PRSY CROSS
CO (ppm)	0.79														
LG (µg/m3)	0.55	0.56													
GU Cross	0.18	0.26	0.34												
MEGU Cross	0.47	0.59	0.54	0.58											
DMP Cross	0.020	0.050	0.20	0.32	0.19										
ETGU Cross	0.22	0.38	0.22	0.55	0.62	0.22									
SY Cross	0.24	0.30	0.43	0.46	0.52	0.40	0.54								
PRGU Cross	0.48	0.47	0.47	0.48	0.78	0.18	0.57	0.62							
CISOEU Cross	0.46	0.43	0.62	0.47	0.63	0.14	0.34	0.49	0.67						
MESY Cross	0.29	0.31	0.47	0.38	0.54	0.42	0.49	0.76	0.59	0.46					
TISOEU Cross	0.45	0.53	0.64	0.54	0.74	0.30	0.56	0.64	0.72	0.82	0.72				
ETSY Cross	0.28	0.21	0.45	0.28	0.36	0.45	0.37	0.73	0.52	0.39	0.78	0.53			
ALLSY Cross	0.22	0.35	0.34	0.11	0.32	0.00	0.10	0.40	0.29	0.19	0.28	0.34	0.18		
PRSY Cross	0.10	0.11	0.26	0.090	0.21	0.33	0.22	0.58	0.45	0.31	0.43	0.38	0.65	0.21	
ACETSY Cross	-0.14	0.030	0.050	0.12	0.070	0.32	0.10	0.27	-0.030	-0.12	0.39	0.18	0.26	0.52	0.11

Table A24: Pearson's Correlation Table for Exposure and Post-Shift Variables Stratified by Mixed Vegetation

	logpmtwa	logcotwa	loglgtwa	loggupost	logmegpost	logdmpost	logetgupost	logsypost	logprgupost	logcisoeupost	logmesypost	logtisoeupost	logetsypost	logallsypost	logprsypost
logcotwa	0.82*														
	0														
	40														
loglgtwa	0.41*	0.49*													
	0.0092	0.0017													
	40	39													
loggupost	0.16	0.12	0.0061												
	0.42	0.54	0.98												
	29	30	29												
logmegupost	0.28	0.12	-0.024	0.63*											
	0.13	0.50	0.90	0.00010											
	31	32	31	32											
logdmppost	0.14	0.084	0.089	0.42*	0.61*										
	0.45	0.65	0.63	0.016	0.00010										
	31	32	31	32	34										
logetgupost	0.46*	0.34	0.24	0.30	0.62*	0.62*									
	0.0096	0.060	0.19	0.092	0.00010	0.00010									
	31	32	31	32	34	34									
logsypost	0.45*	0.32	0.043	0.25	0.35*	0.35*	0.42*								
	0.011	0.078	0.82	0.16	0.045	0.041	0.013								
	31	32	31	32	34	34	34								
logprgupost	0.501*	0.47*	0.17	0.26	0.84*	0.51*	0.79*	0.46*							
	0.0048	0.0071	0.39	0.15	<0.00001	0.0022	<0.00001	0.0077							
	30	31	30	31	33	33	33	33							
logcisoeupost	0.42*	0.24	-0.11	0.14	0.25	0.14	0.41*	0.69*	0.55*						
	0.036	0.23	0.59	0.49	0.19	0.49	0.030	0.0	0.0027						
	25	26	25	27	28	28	28	28	27						
logmesypost	0.39*	0.30	0.052	0.40*	0.55*	0.51*	0.46*	0.75*	0.51*	0.41*					
	0.028	0.096	0.78	0.024	0.00070	0.0022	0.0065	<0.00001	0.0024	0.028					
	31	32	31	32	34	34	34	34	33	28					
logtisoeupost	0.40*	0.20	-0.12	0.34	0.73*	0.49*	0.56*	0.60*	0.64*	0.61*	0.71*				
	0.024	0.28	0.53	0.055	<0.00001	0.0035	0.00060	0.00020	0.0001	0.00060	0				
	31	32	31	32	34	34	34	34	33	28	34				
logetsypost	0.35	0.30	0.1	0.077	0.15	0.28	0.62*	0.75*	0.66*	0.61*	0.56*	0.34			
	0.064	0.11	0.56	0.69	0.41	0.12	0.00020	0.0	0.0001	0.00080	0.0009	0.059			
	29	30	29	30	32	32	32	32	31	27	32	32			
logallsypost	-0.16	-0.27	-0.28	0.0091	0.27	0.36*	0.22	0.17	0.1	0.31	0.16	0.46*	0.11		
	0.40	0.13	0.13	0.96	0.13	0.039	0.21	0.34	0.28	0.11	0.36	0.0065	0.55		
	31	32	31	32	34	34	34	34	33	28	34	34	32		
logprsypost	0.32	0.33	0.071	0.089	0.32	0.24	0.55*	0.58*	0.3*	0.52*	0.58*	0.44*	0.71	* -0.0200	
	0.087	0.067	0.71	0.64	0.071	0.17	0.00090	0.00040	0.0001	0.0053	0.00050	0.011	<0.00001	0.91	
	30	31	30	31	33	33	33	33	32	27	33	33	31	33	
logacetsypost	-0.21	-0.19	-0.088	0.047	0.12	0.31	0.052	0.28	0.13	0.046	0.28	0.30	0.31	0.50*	-0.053
	0.26	0.29	0.64	0.80	0.49	0.075	0.77	0.11	0.49	0.82	0.10	0.088	0.082	0.0025	0.77
	31	32	31	32	34	34	34	34	33	28	34	34	32	34	33

Table A25: Pearson's Correlation Table for Exposure and Post-Shift Variables Stratified by Pine Vegetation

	logpmtwa	logcotwa	loglgtwa	loggupost	logmegpost	logdmpost	logetgupost	logsypost	logprgupost	logcisoeupost	logmesypost	logtisoeupost	logetsypost	logallsypost	logprsypost
logcotwa	0.63*														
	<0.00001														
	81														
loglgtwa	0.39*	0.35*													
	0.00030	0.0020													
	81	77													
loggupost	0.23	0.088	0.20												
	0.098	0.53	0.14												
	55	54	55												
logmegupost	0.35*	0.44*	0.46*	0.35*											
	0.0064	0.00050	0.00020	0.0084											
	61	59	61	56											
logdmppost	0.067	0.071	0.12	0.62*	0.32*										
	0.61	0.59	0.34	<0.00001	0.0093										
	62	60	62	57	65										
logetgupost	0.30*	0.45*	0.42*	0.37*	0.56*	0.44*									
	0.019	0.00030	0.00080	0.0052	<0.00001	0.00030									
	62	60	62	57	65	66									
logsypost	-0.052	-0.16	0.070	0.41*	0.35*	0.38*	0.34*								
	0.69	0.23	0.59	0.0014	0.0046	0.0018	0.0050								
	62	60	62	57	65	66	66								
logprgupost	0.44*7	0.54*	0.52*	0.25	0.66*	0.42*	0.75*	0.23							
	0.00040	<0.00001	<0.00001	0.062	<0.00001	0.00050	<0.00001	0.066							
	63	61	62	57	65	66	66	66							
logcisoeupost	0.24	0.25	0.49*	0.29*	0.59*	0.33*	0.61*	0.32*	0.59*						
	0.076	0.059	0.0001	0.034	<0.00001	0.0093	<0.00001	0.011	0						
	58	56	58	54	61	62	62	62	62						
logmesypost	0.041	-0.083	0.15	0.37*	0.42*	0.36*	0.37*	0.85*	0.30*	0.3353*					
	0.75	0.53	0.25	0.0046	0.00050	0.0026	0.0021	<0.00001	0.014	0.0077					
	62	60	62	57	65	66	66	66	66	62					
logtisoeupost	0.25	0.31*	0.45*	0.34*	0.51*	0.39*	0.59*	0.38*	0.61*	0.8360*	0.4710*				
	0.055	0.016	0.00020	0.011	<0.00001	0.0014	<0.00001	0.0017	0	0	0.0001				
	61	59	61	56	64	65	65	65	65	61	65				
Logetsypost	0.047	-0.044	0.13	0.40*	0.40*	0.36*	0.47*	0.8207*	0.28*	0.3288*	0.8534*	0.3976*			
	0.73	0.76	0.34	0.0043	0.0016	0.0053	0.00020	<0.00001	0.033	0.0125	0	0.002			
	55	53	55	50	59	59	59	59	59	57	59	58			
Logallsypost	0.061	0.19	0.22	0.33*	0.25	0.33*	0.13	0.24	0.22	0.2026	0.3270*	0.3418*	0.10		
	0.64	0.16	0.10	0.015	0.052	0.0070	0.30	0.052	0.083	0.1205	0.0084	0.0061	0.45		
	60	58	60	55	63	64	64	64	64	60	64	63	57		
Logprsypost	-0.11	0.13	0.049	-0.014	0.12	0.33*	0.34*	0.32*	0.16	0.0959	0.3006*	0.1436	0.48*	0.058	
	0.40	0.34	0.71	0.92	0.35	0.0085	0.0068	0.011	0.21	0.4741	0.0176	0.2695	0.00020	0.66	
	58	57	58	54	61	62	62	62	62	58	62	61	56	60	
logacetsypost	-0.14	-0.013	0.11	0.31*	0.061	0.31*	0.094	0.45*	0.11	0.1165	0.5002*	0.3361*	0.40*	0.55*	0.16
	0.29	0.92	0.42	0.020	0.63	0.014	0.46	0.00020	0.37	0.3713	<0.00001	0.0071	0.0020	<0.00001	0.24
	60	58	60	55	63	64	64	64	64	61	64	63	58	63	60

Table A26: Pearson's Correlation Table for Exposure and Cross-Shift Variables Stratified by Mixed Vegetation

	logpmtwa	logcotwa	loglgtwa	loggudiff	logmegudiff	logdmpdiff	logetgudiff	logsydiff	logprgudiff	logcisoeudiff	logmesydiff	logtisodiff	logetsydiff	logallsydiff	logprsydiff
logcotwa	0.82*														
	<0.00001														
	40														
loglgtwa	0.41*	0.49*													
	0.0092	0.0017													
	40	39													
loggudiff	0.081	0.16	-0.031												
	0.79	0.59	0.92												
	13	14	13												
logmegudiff	0.46*	0.26	0.028	0.46											
	0.020	0.17	0.90	0.095											
	25	26	25	14											
logdmpdiff	0.34	0.56*	0.50	0.42	0.67*										
	0.21	0.024	0.059	0.20	0.0061										
	15	16	15	11	15										
logetgudiff	0.57*	0.46*	0.24	0.86*	0.74*	0.79*									
	0.013	0.048	0.33	0.0013	0.0011	0.0041									
	18	19	18	10	16	11									
logsydiff	0.1	0.087	-0.046	-0.077	0.19	0.28	0.15								
	0.14	0.69	0.83	0.79	0.39	0.29	0.55								
	24	24	24	14	22	16	18	0.25							
logprgudiff	0.43	0.30	0.20	0.45	0.63*	0.50	0.68*	0.32							
	0.055	0.19	0.37	0.17	0.0039	0.12	0.0076	18							
	21	21	21	11	19	11	14	0.63*	0.60						
logcisoeudiff	0.47	0.54*	-0.0013	0.80*	0.66*	0.72*	0.84*	0.0083	0.051						
	0.059	0.020	1.0	0.010	0.0051	0.0088	0.0024	16	11						
	17	18	17	9.0	16	12	10	0.61*	0.61*	0.57*					
logmesydiff	0.47*	0.43*	0.16	0.32	0.69*	0.81*	0.63*	0.0021	0.0052	0.021					
	0.015	0.028	0.45	0.27	0.00040	0.00020	0.0088	23	19	16					
	26	26	26	14	22	16	16	0.50*	0.52*	0.82*	0.75*				
logtisodiff	0.56*	0.32	-0.053	0.52*	0.65*	0.64*	0.74*	0.013	0.019	<0.00001	<0.00001				
	0.0037	0.11	0.80	0.048	0.00030	0.0061	0.00070	24	20	18	24				
	25	26	25	15	26	17	17	0.82*	0.47	0.63*	0.69*	0.72*			
logetsydiff	0.22	0.20	0.094	0.34	0.22	0.75*	0.70*	<0.00001	0.076	0.022	0.0024	0.0005			
	0.37	0.41	0.70	0.31	0.38	0.0085	0.0052	19	15	13	17	19			
	19	19	19	11	18	11	14	0.27	0.23	-0.075	-0.0072	0.069	-0.040		
logallsydiff	-0.30	-0.38	-0.32	0.41	0.042	0.58	0.40	0.26	0.40	0.79	0.98	0.77	0.88		
	0.18	0.084	0.14	0.24	0.86	0.077	0.14	20	17	15	20	21	17		
	22	22	22	10	19	10	15	0.65*	0.62*	0.16	0.38	0.58*	0.55	0.32	
logprsydiff	0.34	0.17	-0.13	0.23	0.33	0.17	0.49	0.0091	0.024	0.64	0.21	0.024	0.051	0.31	
	0.21	0.54	0.63	0.55	0.24	0.62	0.11	15	13	11	13	15	13	12	
	15	16	15	9.0	14	11	12								
logacetsydiff	-0.43	-0.491*	-0.32	0.54	0.056	0.35	0.21	0.39	0.042	0.48	0.17	0.25	0.23	0.82*	0.10
	0.092	0.045	0.23	0.088	0.84	0.40	0.54	0.17	0.89	0.16	0.53	0.35	0.46	0.0001	0.78
	16	17	16	11	16	8.0	11	14	13	10	15	16	13	16	10

Table A27: Pearson's Correlation Table for Exposure and Cross-Shift Variables Stratified by Pine Vegetation

	logpmtwa	logcotwa	loglgtwa	loggudiff	logmegudiff	logdmpdiff	logetgudiff	logsydiff	logprgudiff	logcisoeudiff	logmesydiff	logtisodiff	logetsydiff	logallsydiff	logprsydiff
logcotwa	0.63*														
	<0.00001														
	81														
loglgtwa	0.39*	0.35*													
	0.0003	0.0020													
	81	77													
loggudiff	0.27	0.49*	0.48*												
	0.17	0.0078	0.011												
	27	28	27												
logmegudiff	0.39*	0.73*	0.51*	0.65*											
	0.0043	<0.00001	0.00010	0.00030											
	51	51	51	26											
logdmpdiff	-0.0035	0.15	0.11	0.67*	0.45*										
	0.98	0.37	0.50	0.00070	0.0077										
	39	37	39	22	34										
logetgudiff	0.12	0.31	0.17	0.72*	0.68*	0.51*									
	0.44	0.051	0.28	0.00020	<0.00001	0.0031									
	43	40	43	22	39	32									
logsydiff	-0.11	-0.12	0.0085	0.31	0.28	0.45*	0.39*								
	0.46	0.43	0.95	0.096	0.064	0.0063	0.015								
	50	49	50	29	45	35	38								
logprgudiff	0.49*	0.55*	0.46*	0.69*	0.67*	0.43*	0.69*	0.28							
	0.00040	0.00010	0.0010	0.00020	<0.00001	0.010	<0.00001	0.071							
	48	47	48	24	45	34	37	41							
logcisoeudiff	0.39*	0.47*	0.52*	0.57*	0.71*	0.28	0.52*	0.42*	0.76*						
	0.012	0.0035	0.00060	0.0061	<0.00001	0.17	0.0043	0.0093	<0.00001						
	40	37	40	22	35	26	28	37	36						
logmesydiff	0.069	-0.11	0.17	0.32	0.29	0.36*	0.28	0.75*	0.36*	0.25					
	0.64	0.45	0.24	0.10	0.051	0.036	0.099	<0.00001	0.020	0.14					
	49	49	49	27	45	35	36	49	42	35					
logtisodiff	0.30*	0.41*	0.55*	0.75*	0.71*	0.29	0.66*	0.32*	0.60*	0.74	* 0.42*				
	0.045	0.0054	0.00010	<0.00001	<0.00001	0.11	<0.00001	0.035	<0.00001	<0.00001	0.0056				
	45	44	45	25	40	32	33	44	41	37	43				
logetsydiff	0.048	0.067	0.12	0.42*	0.36*	0.60*	0.47*	0.67*	0.36*	0.27	0.82*	0.43*			
	0.77	0.69	0.47	0.044	0.033	0.00040	0.0073	<0.00001	0.034	0.15	<0.00001	0.0096			
	39	38	39	23	36	30	31	40	35	31	39	35			
logallsydiff	0.14	0.26	0.36*	0.54*	0.32	0.67*	0.31	0.42*	0.27	0.018	0.51*	0.40*	0.53*		
	0.42	0.13	0.034	0.020	0.075	0.00010	0.12	0.018	0.12	0.93	0.0034	0.020	0.0047		
	35	34	35	18	32	27	26	32	34	26	31	33	27		
logprsydiff	-0.23	-0.018	-0.022	0.52*	0.37*	0.53*	0.44*	0.63*	0.12	0.25	0.51*	0.33	0.49*	0.29	
	0.17	0.92	0.90	0.019	0.036	0.0035	0.019	0.0001	0.50	0.20	0.0026	0.070	0.0062	0.17	
	38	37	38	20	32	28	28	33	33	27	33	31	30	24	
logacetsydiff	-0.35*	-0.16	-0.049	-0.14	-0.067	0.48*	0.13	0.48*	0.13	-0.17	0.41*	-0.051	0.40*	0.47*	0.14
	0.043	0.39	0.79	0.59	0.72	0.0096	0.51	0.0057	0.50	0.43	0.021	0.78	0.039	0.0096	0.48
	33	32	33	18	32	28	28	32	31	24	32	31	27	29	26

Table A28: Pearson's Correlation Table for Exposure and Pre-Shift Variables Stratified by Mixed Vegetation

	logpmtwa	logcotwa	loglgtwa	loggupre	logmegupre	logdmppre	logetgupre	logsypre	logprgupre	logcisoeupre	logmesypre	logtisoeupre	logetsypre	logallsypre	logprsypre
logcotwa	0.82*														
	0														
	40														
loglgtwa	0.41*	0.49*													
	0.0092	0.0017													
	40	39													
loggupre	0.060	-0.095	-0.019												
	0.77	0.63	0.93												
	27	28	27												
logmegupre	0.084	-0.024	0.12	0.37*											
	0.66	0.90	0.54	0.046											
	30	31	30	29											
logdmppre	-0.13	-0.18	-0.0087	0.32	0.60*										
	0.48	0.33	0.96	0.087	0.00030										
	30	31	30	30	32										
logetgupre	0.060	-0.20	-0.018	0.33	0.45*	0.54*									
	0.75	0.27	0.92	0.072	0.011	0.0011									
	31	32	31	30	32	33									
logsypre	0.10	-0.045	0.040	0.28	0.49*	0.65*	0.58*								
	0.59	0.81	0.83	0.14	0.0054	0.00010	0.00040								
	31	31	31	29	31	32	33								
logprgupre	-0.026	-0.0069	-0.26	0.061	0.24	0.27	0.38*	0.30							
	0.90	0.97	0.17	0.76	0.20	0.14	0.030	0.099							
	30	30	30	28	30	31	32	32							
logcisoeupre	-0.052	-0.17	-0.24	0.071	0.30	0.19	-0.012	0.29	0.41*						
	0.80	0.41	0.25	0.73	0.12	0.33	0.95	0.13	0.032						
	26	27	26	25	27	28	29	28	27						
logmesypre	-0.11	-0.19	-0.074	0.052	0.43*	0.57*	0.22	0.72*	0.31	0.44*					
	0.55	0.32	0.70	0.80	0.018	0.00090	0.24	<0.00001	0.092	0.021					
	30	30	30	28	30	31	32	32	31	27					
logtisoeupre	0.19	0.033	-0.12	0.27	0.51*	0.32	0.41*	0.47*	0.47*	0.57*	0.38*				
	0.30	0.86	0.50	0.14	0.0030	0.071	0.015	0.0061	0.0063	0.0014	0.034				
	31	32	31	30	32	33	34	33	32	29	32				
logetsypre	-0.23	-0.35	-0.066	0.12	0.36*	0.47*	0.53*	0.57*	0.56*	0.32	0.57*	0.46*			
	0.22	0.057	0.72	0.55	0.045	0.0068	0.0017	0.00050	0.0010	0.092	0.00060	0.0077			
	31	31	31	29	31	32	33	33	32	28	32	33			
logallsypre	0.12	-0.0044	-0.42*	0.21	0.22	0.18	0.29	0.26	0.35*	0.33	0.17	0.59*	0.12		
	0.53	0.98	0.020	0.28	0.24	0.31	0.11	0.14	0.048	0.087	0.36	0.00030	0.52		
	31	31	31	29	31	32	33	33	32	28	32	33	33		
logprsypre	-0.21	-0.17	-0.071	-0.35	0.16	0.20	0.18	0.16	0.51*	0.093	0.13	0.11	0.27	0.31	
	0.29	0.39	0.72	0.075	0.41	0.29	0.32	0.40	0.0044	0.65	0.50	0.57	0.14	0.094	
	28	29	28	27	29	30	31	30	29	26	29	31	30	30	
logacetsypre	0.074	0.00060	-0.055	0.063	0.42*	0.28	0.45*	0.27	0.34	0.078	0.17	0.36*	0.29	0.40*	0.086
	0.70	1.0	0.77	0.74	0.018	0.12	0.0093	0.13	0.064	0.69	0.35	0.039	0.12	0.024	0.65
	30	31	30	29	31	32	33	32	31	28	31	33	32	32	30

Table A29: Pearson's Correlation Table for Exposure and Pre-Shift Variables Stratified by Pine Vegetation

	logpmtwa	logcotwa	loglgtwa	loggupre	logmegupre	logdmppre	logetgupre	logsypre	logprgupre	logcisoeupre	logmesypre	logtisoeupre	logetsypre	logallsypre	logprsypre
logcotwa	0.63*														
	<0.00001														
	81														
loglgtwa	0.39*	0.35*													
	0.00030	0.002													
	81	77													
loggupre	0.15	-0.12	0.063												
	0.27	0.37	0.64												
	58	56	58												
logmegupre	0.12	-0.057	0.11	0.39*											
	0.37	0.67	0.41	0.0025											
	59	58	59	59											
logdmppre	0.12	-0.010	0.18	0.42*	0.17										
	0.35	0.94	0.16	0.00070	0.18										
	61	59	61	61	62										
logetgupre	-0.028	-0.12	0.15	0.32*	0.22	0.46*									
	0.83	0.38	0.25	0.012	0.093	0.00010									
	60	58	60	60	61	64									
logsypre	0.034	0.018	0.082	0.30*	0.13	0.52*	0.52*								
	0.79	0.89	0.53	0.019	0.30	<0.00001	<0.00001								
	62	60	62	62	63	65	64								
logprgupre	0.072	0.26	0.070	-0.11	0.020	-0.10	0.023	0.070							
	0.58	0.050	0.60	0.41	0.88	0.43	0.86	0.59							
	60	58	59	59	60	62	61	63							
logcisoeupre	0.10	0.15	0.20	0.16	0.23	0.24	0.12	0.35*	-0.024						
	0.45	0.28	0.14	0.26	0.087	0.071	0.36	0.0066	0.86						
	56	53	56	55	56	58	57	59	57						
logmesypre	0.091	0.084	0.06	0.022	0.13	0.14	0.18	0.65*	0.20	0.56*					
	0.49	0.53	0.64	0.87	0.31	0.27	0.15	<0.00001	0.12	<0.00001					
	60	58	60	60	61	63	62	64	61	57					
logtisoeupre	0.074	0.24	0.25	0.33*	0.21	0.27*	0.35*	0.41*	0.046	0.61*	0.43*				
	0.58	0.071	0.058	0.012	0.11	0.034	0.0056	0.00070	0.72	<0.00001	0.00040				
	60	58	60	60	61	63	62	64	62	58	63				
logetsypre	0.10	0.12	0.099	-0.039	0.081	0.14	0.55*	0.57*	0.15	0.25	0.59*	0.50*			
	0.46	0.37	0.46	0.77	0.54	0.28	<0.00001	<0.00001	0.25	0.068	<0.00001	<0.00001			
	58	56	58	58	59	61	60	62	59	55	62	61			
logallsypre	0.26*	0.090	0.082	0.029	0.087	0.13	0.026	0.18	-0.042	0.26*	0.33*	0.28*	0.15		
	0.042	0.50	0.53	0.82	0.50	0.29	0.84	0.15	0.75	0.048	0.0079	0.025	0.25		
	62	60	62	62	63	65	64	66	63	59	64	64	62		
logprsypre	0.0099	0.17	-0.014	-0.41*	-0.14	0.052	0.028	0.21	0.36*	-0.099	0.20	-0.0071	0.42*	0.11	
	0.94	0.19	0.92	0.0012	0.27	0.69	0.83	0.090	0.0050	0.46	0.11	0.96	0.0008	0.40	
	60	58	60	60	61	63	62	64	61	57	63	62	61	64	
logacetsypre	0.035	-0.021	0.21	0.13	0.17	0.54*	0.53*	0.41*	0.045	0.35*	0.27*	0.26*	0.26*	0.39*	0.021
	0.79	0.87	0.11	0.33	0.18	0	0	0.0005	0.73	0.0059	0.030	0.036	0.04	0.0014	0.87
	62	60	62	62	63	65	64	66	63	59	64	64	62	66	64

Tables A24-A29 illustrate Pearson's correlation coefficients for the exposure and outcome variables when stratified for different fuel types (pine and mixed vegetation). Because past research indicates that methoxyphenol compounds are heavily influenced by vegetation type (Dills et al. 2001, 2006, Neitzel et al. 2009), we decided that it was appropriate to stratify the data by fuel type prior to examining the correlations. Additionally, since the compounds are log-normally distributed, the data were log-transformed prior to calculation of correlation coefficients.

Based on the results, many of the guaiacol-type compounds in the Pearson's tables based on pine vegetation correlate strongly with each other, especially in post-shift urine samples and cross-shift urine samples. Syringol-type compounds also correlate with each other in post-shift and cross-shift urine samples - more so for mixed vegetation.

Further, the compounds correlated with the exposure variables. Although guaiacols elicited a greater association with the exposure variables for post-shift concentrations, the syringols also exhibited association, but to a lesser degree. This indicates that for this particular dataset, guaiacols elicit a greater degree of association with exposure variables than syringol compounds because the vegetation is dominated by pine.

Although guaiacol-type compounds dominate soft woods, while syringol-type compounds dominate hard woods, it is interesting to note that there is still a presence of both guaiacol-type and syringol-type compounds in pine and mixed vegetation. As noted with the Spearman correlation tables, several of the methoxyphenols were moderately correlated with the exposure variables when considering post-shift or cross-shift data. For several of the guaiacols measured in post shift samples (or expressed as a cross-shift change), the correlations between outcome variables and CO exposure were stronger than the correlation between CO and LG.

Simple and Multiple Regression Models

The following tables include several regression models in addition to those shown in the main body of the thesis.

Table A30: Simple and Multiple Linear Regression Models of Geometric Mean 8-hour Carbon Monoxide Time Weighted Average Concentration with Post-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine)

Post-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine)												
Predictor Variable	Outcome Variable	Covariate	N	Coefficient	Std. Err.	Coeff. P-Value	95% CI		Model R^2	Adj. R^2	F-statistic	F-statistic P-value
Log[CO]	Log[PRGU]		93	0.57	0.090	<0.0001	0.39	0.75	0.31	0.30	40.89	<0.00001
Log[CO]	Log[PRGU]		88	0.56	0.096	<0.0001	0.37	0.75	0.30	0.29	18.49	<0.0001
		Log(PRGU Pre)		0.015	0.065	0.82	-0.11	0.14				
Log[CO]	Log[PRGU]		92	0.55	0.11	<0.0001	0.33	0.78	0.34	0.31	14.8	<0.0001
		Fuel Type		-0.35	0.20	0.085	-0.75	0.05				
		Interaction CO		-0.078	0.19	0.69	-0.31	0.46				
Log[CO]	Log[PRGU]		87	0.55	0.12	<0.0001	0.31	0.79	0.33	0.30	10.13	<0.0001
		Fuel Type		-0.38	0.21	0.080	-0.81	0.046				
		Interaction CO		-0.10	0.21	0.62	-0.52	0.31				
		PRGU Pre		-0.0076	0.066	0.91	-0.14	0.12				

Note: Interaction 2 = Fuel Type x CO 8-hr TWA; Fuel Type: 0=Pine, 1=Mixed

Table A31: Multiple Linear Regression Model of Mean 8-hour Carbon Monoxide Time Weighted Average Concentration on Cross-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine)

Cross-Shift Creatinine Adjusted Propylguaiacol (µg/mg creatinine) without Substituted LOD/sqrt(2)												
Log Transformed Predictor Variable	**Log Transformed Outcome Variable**	**Covariate**	**N**	**Coefficient**	**Std. Err.**	**Coefficient P-Value**	**95% CI**		**Model R^2**	**Adj. R^2**	**F-statistic**	**F-statistic P-value**
CO 8-hr TWA	**Propylguaiacol**		**58**	0.84	0.18	<0.0001	0.48	1.20	0.33	0.29	8.94	0.0001
		Fuel Type		-0.19	0.34	0.57	-0.87	0.49				
		Interaction 2		-0.52	0.31	0.098	-1.1	0.098				

Note: Interaction 2 = Fuel Type x CO 8-hr TWA; Fuel Type: 0=Pine, 1=Mixed