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Indoor and outdoor particulate matter and endotoxin concentrations in an intensely agricultural county

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Abstract

The objectives of this study were to characterize rural populations' indoor and outdoor exposure to PM_{10}, $PM_{2.5}$, and endotoxin and identify factors that influence these concentrations. Samples were collected at 197 rural households over five continuous days between 2007 and 2011. Geometric mean indoor PM_{10} (21.2 μg m^{-3}) and $PM_{2.5}$ (12.2 μg m^{-3}) concentrations tended to be larger than outdoor PM_{10} (19.6 μg m^{-3}) and $PM_{2.5}$ (8.2 μg m^{-3}) concentrations (PM_{10} p= 0.086; $PM_{2.5}$ p <0.001). Conversely, GM outdoor endotoxin concentrations (1.93 EU m^{-3}) were significantly larger than indoor (0.32 EU m^{-3}) (p<0.001). Compared to measurements from previous urban studies, indoor and outdoor concentrations of PM_{10} and $PM_{2.5}$ in the study area tended to be smaller while, ambient endotoxin concentrations measured outside rural households were 3-10 times larger. Contrary to our initial hypothesis, seasonality did not have a significant effect on mean ambient PM_{10} concentrations; however, endotoxin concentrations in the autumn were almost seven-times larger than winter. Excluding home cleanliness, the majority of agricultural and housing characteristics evaluated were found to be poorly associated with indoor and outdoor particulate and endotoxin concentrations.

Keywords

rural air quality; particulate matter; endotoxin

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Introduction

Approximately 194 million people live in rural areas throughout the United States, Canada, and European Union, however there is paucity of exposure assessment data on these individuals.[1] Occupational studies have shown that agricultural workers are regularly exposed to large concentrations of particulate matter (PM) and endotoxin while performing common tasks, such as crop harvesting, grain processing, and livestock production.[2-8] However, the effect of agricultural activities on rural air quality has not been well characterized and population-based exposure information is needed.

PM suspended in the ambient air is a heterogeneous mixture of inorganic and organic substances, the composition of which can vary depending on the source, season, and meteorological conditions.[9] Health effects from PM are determined by both the pathogenic effect of the substance and the area in which it deposits in the lung.[10] Epidemiological studies have demonstrated a clear association between exposure to PM and a number of adverse health effects including respiratory, cardiac, and all-cause mortality.[11-18] As part of the Clean Air Act, the United States Environmental Protection Agency (EPA) has promulgated air quality standards for two size fractions of particulate, PM_{10} (aerodynamic diameter ≤ 10 μm) and $PM_{2.5}$ (aerodynamic diameter ≤ 2.5 μm).[16] Fine particulate ($PM_{2.5}$), produced through combustion processes, is more efficiently inhaled than larger coarse particles (aerodynamic diameter >2.5 μm and ≤ 10 μm) and can potentially deposit deeper in the lungs.[17] Therefore, ambient exposure to $PM_{2.5}$ may have a larger impact on human health than PM_{10}.[15,17]

The vast majority of air quality studies have focused on urban areas, which compared to rural, may vary considerably in terms of composition of PM.[9] Agricultural air has a larger fraction of organic dust which is a mixture of plant and animal matter, microorganisms, and bio-aerosols.[19] Exposure to organic dust can cause a variety of acute or chronic conditions that are separate and distinct from health effects associated with urban PM exposure. Occupational workers exposed to large concentrations of organic dust can develop organic toxic dust syndrome, which is characterized by fever, chills, malaise, and dyspnea.[20,21] Long-term exposures can cause decreased lung function as well as chronic bronchitis, asthma-like syndrome, and wheezing.[7,22,23] Adverse health effects have also been linked to populations environmentally exposed. During 1985-1986, a series of asthma epidemics was found to be caused by environmental exposure to soybean dust in Barcelona, Spain.[24] Schwartz (1999) concluded that environmental exposure to organic dust among rural populations is one of the most important exposures in the progression of childhood asthma.[25]

The concentration of endotoxins in the inhaled organic dust fraction appears to be an important factor in the progression and development of respiratory diseases.[26,27] Endotoxins are made up of lipids, proteins, and lipopolysaccharides and are capable of remaining airborne for long periods of time due to their small size.[28] However, endotoxins are often attached onto PM, and consequently the majority of endotoxin is found in the coarse fraction as opposed to the fine fraction of particulate samples. [29-31] Sources of endotoxins in rural

environments include animal confinements, grain storage facilities, and row crop harvesting.[2,7,26,27,32,33]

Indoor and outdoor PM_{10}, $PM_{2.5}$, and endotoxin samples were collected from 197 rural households over five continuous days from 2007-2011 in order to characterize exposure to participants in a prospective population-based health study. The goals of this study were to quantify airborne concentrations of PM_{10}, $PM_{2.5}$, and endotoxin in an intensely agricultural area and compare findings to reported concentrations from urban areas; identify factors contributing to rural PM and endotoxin concentrations in both ambient and indoor air in homes; and evaluate the effect of seasonal variation on PM and endotoxin levels.

Methods

Study Area and Recruitment

Keokuk County, located in east-central Iowa, is considered entirely rural with no towns having a population greater than 2,500 residents. According to the 2010 US census, the population of the county was 10,511.[34] The majority of the land area in the county was devoted to agricultural production (86%), with approximately 318,160 acres considered cropland, pastures, and trees. The primary crops grown in the county were corn and soybeans, accounting for 157 and 57 tonnes harvested in 2009, respectively.[35]

Households recruited from the third round (2006-2011) of the Keokuk County Rural Health Study (KCRHS). The KCRHS is a prospective population-based cohort study designed to primarily investigate the incidence of respiratory disease and injuries in an intensely agricultural county. Recruitment methodology for the KCRHS has been previously published.[36] Although the KCRHS enrolled participants using a stratified random sample of eligible households within the county, the environmental assessment of the homes was a non-random sample. Residential properties in Keokuk County are designated by the Tax Assessor's Office as either residential, if the home is located within a town, or agricultural if it is located outside a town. Households in this study were selected from the enrolled KCRHS participants based on their willingness to allow investigators access to their home, spatial location within the county, and household designation (town or agricultural). The recruitment goals were to sample from an even spatial distribution of homes throughout the county and to sample from at least 25% of homes located within a town.

Sample Collection

Indoor environmental samples were collected to monitor for PM_{10}, $PM_{2.5}$, temperature, relative humidity, CO, and CO_2; outdoor PM_{10} and $PM_{2.5}$ were collected over the same time period at least 3 m from the home and away from any large obstructions. All environmental samples were collected over a five-day period unless scheduling conflicts necessitated a four- or six-day sample. A Q-TRAK™ (TSI Inc., St. Paul, MN) monitored indoor temperature, relative humidity, CO, and CO_2. The Q-TRAK™ data-logged every 30 minutes, and measurements were averaged over the entire sampling period. To ensure accurate measurements, the Q-TRAK™ was calibrated on a monthly basis.

Indoor PM_{10} and $PM_{2.5}$ samples were obtained using Personal Environmental Monitors (PEM) (SKC Inc., Eighty Four, PA) attached to BGI (BGI Inc., Waltham, MA) personal sampling pumps operated at 4 L min^{-1}. The samplers, pumps, and Q-TRAK™ were located in an area where the family reported spending most of their time and at least 1 m above the ground. To reduce particle bounce, a thin layer of mineral oil was applied to the impaction plate prior to each sampling period.

Ambient PM samples were collected with a dichotomous sampler with a 10 μm inlet (Model 2000i, Thermo Fisher Scientific Inc., Franklin, MA). The sampler uses a virtual impactor to separate the particles into two fractions, coarse and fine. In order to achieve proper cut points, the flow rates were set to 1.67 L min^{-1} for the coarse flow and 15.00 L min^{-1} for the fine flow.

All PM samples were collected on 37 mm polytetrafluoroethylene (PTFE) filters with a 0.8 μm pore size (Pall Corporation, Ann Arbor, MI).The pumps were calibrated at the start of the sampling session and post-calibrated during retrieval with a TetraCal™ (BGI Inc., Waltham, MA) volumetric flow calibrator. The initial and final flow rates were averaged, and this average flow rate was used to determine the volume of air sampled. A sample was considered acceptable and included in the analysis if the average flow rate was within ±10% of the initial flow rate.

Filters were pre- and post-weighed with an electrical microbalance (Mettler MT5, Columbus, OH) with a sensitivity of 2.0 μg. Prior to weighing, all filters were stored in a temperature and humidity controlled room for at least 48 hours to allow for acclimatization to stable room conditions. Additionally, all filters were passed over a ^{210}Po alpha emitter to neutralize static charge. During each weighing session the accuracy of the micro-balance was assessed using calibrated laboratory weights (200, 100, and 20 mg). In addition, field blanks were evaluated for each sampling period. Since all field blanks did not deviate by more than ±0.05%, no blank correction was performed.

Once filters were post-weighed they were returned to their filter cassette and stored in a −20 °C freezer until endotoxin analysis could be performed. During the beginning of the study, filters were not immediately stored in the freezer and remained unfrozen for approximately two years. A study by Spaan et al. (2007) found a 10% higher estimated endotoxin concentration on filters stored in the freezer compared to those stored in a refrigerator. Researchers hypothesized this is due to the freeze-thaw cycle lysing bacteria and therefore allowing for greater detection.[37] Since all filters were eventually stored in the freezer, storage method should not have biased results.

Endotoxin Analysis

A subset of homes (n=117) were selected for endotoxin analysis. In order for the sample to be considered for endotoxin analysis, all indoor and outdoor measurements had to meet the flow rate and sampling time restrictions (n=159). All homes that met this criteria and had a confined animal feeding operation located on their property (<400 m) were selected for analysis (n=16). The remaining 101 homes were selected at random from the remaining samples. Only the coarse fraction (10-2.5 μm) of the outdoor PM sample was analyzed for

endotoxin; whereas, the entire indoor PM_{10} fraction (<10 μm) was assayed. Since previous studies have shown the coarse fraction of particulate samples contain the bulk of endotoxins, underestimation of ambient concentration was assumed to be minimal.[29-31]

The endotoxin extracted from the filters was evaluated using the kinetic chromogenic Limulus Amebocyte Lysate (LAL) assay that has been previously described by Thorne (2000). [38] Filters were extracted in 10.0 mL of pyrogen-free water and shaken for 1 hour at room temperature. One mL was pipetted into a cryovial and spun for 5 min at 600 ×g (Marathon 16KM) to decrease inhibition from filter particulate. The filter extracts were assayed using five-fold serial dilutions. Two-fold dilutions of the Control Standard Endotoxin were assayed to create a 12-point standard curve from 50.0 EU mL^{-1} to 0.0244 EU mL^{-1}. The samples and field blanks were assayed in 96 well microplates (Corning Inc, Corning, NY) and the rate change of absorbance was measured at 405 nm every 30 seconds for 90 min using a microplate reader (Molecular Devices SpectraMax 384 Plus, Sunnyvale, CA with Softmax PRO 4.0 analysis software).

Re-Sampled Households

Households with complete indoor and outdoor PM measurements (n=159) were eligible for re-sampling. Fifteen homes were selected at random and re-sampled for indoor and outdoor PM_{10} and $PM_{2.5}$. Since seasonality was hypothesized to effect PM concentrations, homes were re-sampled in a different season.

Seasonal Calculation and Meteorological Data

Mean daily precipitation (cm), relative humidity (%), and wind speed (ms^{-1}) data were obtained from a weather station located approximately 30 km southwest of the center of the county and considered representative of weather conditions throughout the county.[39] Daily meteorological conditions were subsequently averaged over the course of the multi-day sampling period. Sampling seasons were assigned based on the end sample date: Winter was defined as December, January, and February; spring was March, April, May; summer was June, July, and August; and autumn was September, October, and November. In Iowa, the majority of corn and soybean harvest occurs during the autumn months.

Questionnaires

A trained interviewer administered an environmental questionnaire to the home owner at the beginning of the assessment. The participant was also asked to identify all agricultural operations on their property within 0.4 km of the residence, which included whether the family raised livestock, had a confined animal feeding operation, and/or had grain storage bins. A cadastral map was used to determine the type of road surface on which the home was located (gravel vs. paved).

Qualitative Assessment of a Home's Cleanliness

During each environmental survey, a single interviewer rated the overall maintenance and condition of the home on a scale of 1 to 5, with 5 considered the cleanest, most well-maintained household. While accompanied by the home owner, the interviewer was able to walk through the living space of the home. However, in general, the interviewer did not

have access to all of the bedrooms in the home. In order to minimize bias, the home inspection was performed discretely during the walkthrough with the homeowner. The rating scale was based on visual inspection for dirt and mold on the ceiling, walls, and floor; clutter on the floor, countertops, cabinets, and tables; condition of exterior and interior of the home; peeling interior paint; visible pet hair on the floor and furniture upholstery; and whether the home had an insect or rodent problem assessed through questionnaire information. The five levels were subsequently collapsed into three home cleanliness categories, with low being designated as (1-2), medium (3) and high (4-5). This was done to increase the sample size in each category and achieve the requisite power to detect differences in the groups.

Statistical Analysis

SAS version 9.2 (SAS Institute Inc., Cary, NC) was used for all statistical analysis. PM and endotoxin data were checked for normality and determined to be log-normally distributed. If continuous predictor variables were missing, they were substituted with the median of all reported values for the variable; while missing categorical variables were substituted with the mode for the variable. Paired t-tests were used to investigate whether indoor air had significantly ($p<0.05$) different concentrations of PM and endotoxin compared to outdoor. Bivariate analysis was conducted on log-transformed outdoor PM and endotoxin concentrations to determine if concentrations differed by season. Tukey-Kramer multiple comparison tests were used to determine significant differences in mean concentrations ($p<0.05$) across seasons. Wilcoxon signed-rank tests were used to determine if re-sampled PM measurement differed significantly.

Multivariate analysis was conducted to determine associations between agricultural and environmental variables and indoor and outdoor PM and endotoxin concentrations. Backwards elimination was used to eliminate variables sequentially until only variables with a $p<0.05$ remained in the model. Due to meteorological conditions not being independent, outdoor PM and endotoxin samples were analyzed using a mixed model (PROC MIXED). Each sampling period was given a unique ID number which was entered into the “subject” statement. Since indoor samples could be treated as independent measurements, associations were determined using a general linear model (PROC GLM).

Results

General characteristics of the 197 homes surveyed in the study are shown in Table 1. The majority of were single-family homes (89%), located outside of designated towns (71%), built prior to 1950 (52%), and on gravel roads (55%). Participants typically heated their homes with natural gas or propane (76%), used an electric stove (68%) for cooking, and did not allow smoking inside the home (91%).

Summary results for indoor and outdoor PM and endotoxin data are shown in Table 2. The range of indoor concentrations of PM spanned two orders of magnitude (PM_{10}: 4.1 to 173.3 μg m^{-3}; $PM_{2.5}$: 1.4 to 187.7 μg m^{-3}), while outdoor PM levels were less varied and spanned only a single order of magnitude (PM_{10}: 6.2 to 56.2 μg m^{-3}; $PM_{2.5}$: 1.5 to 24.1 μg m^{-3}). Geometric mean (GM) indoor PM_{10} (21.2 μg m^{-3}) and $PM_{2.5}$ (12.2 μg m^{-3}) concentrations

tended to be larger than outdoor PM_{10} (19.6 μg m^{-3}) and $PM_{2.5}$ (8.2 μg m^{-3}) concentrations (PM_{10} p= 0.086; $PM_{2.5}$ p <0.001). Conversely, GM outdoor endotoxin concentrations (1.93 EU m^{-3}) were significantly larger than indoor (0.32 EU m^{-3}) (p<0.001).

A subset of homes (n=15) were re-sampled for indoor and outdoor PM_{10} and $PM_{2.5}$ (Table 3). Due to flow rate and sampling time restrictions, only indoor PM_{10} measurements contained all fifteen matched samples. Since the number of re-sampled homes was small, Wilcoxon signed-rank tests were used to evaluate pairwise differences in re-sampled homes. Results showed no significant difference in PM concentrations in re-sampled homes between sample periods; however, a lack of power may be responsible for the null finding.

Bivariate analysis was conducted to determine significant differences in ambient concentrations of PM and endotoxin by season (Table 4). No seasonal trend was observed in ambient PM_{10} concentrations. A seasonal trend was found in the outdoor endotoxin measurements, with autumn (2.63 EU m^{-3}) having approximately seven-times larger endotoxin concentrations compared to winter (0.39 EU m^{-3}). A seasonal trend was also detected in ambient $PM_{2.5}$ levels. Compared to other seasons, winter (10.6 μg m^{-3}) had significantly larger concentrations of $PM_{2.5}$, while autumn had the smallest (6.8 μg m^{-3}).

In mixed regression analysis (Table 5) the majority of agricultural and property variables were not found to be significantly associated with outdoor PM and endotoxin levels. One variable that was found to be associated with outdoor PM_{10} levels was home location (town vs. agricultural). After adjusting for significant covariates, residents living in agricultural areas had significantly larger PM_{10} concentrations (20.8 μg m^{-3}) than residents living in designated towns (17.6 μg m^{-3}). Interestingly, when controlling for home location, no significant increase in PM_{10} concentrations was found between homes situated on a paved roads compared to a gravel roads (p= 0.297). Additionally, no significant association was observed between ambient endotoxin concentrations and presence of livestock, swine confinements, and/or grain bins on the property. However, unmeasured variables such as distance and direction were not taken into consideration in the model and the magnitude of the association may have been attenuated.

Multiple linear regression analysis of the indoor sample results is presented in Table 6. Smoking, outdoor PM concentrations, and indoor relative humidity were all significantly (p<0.05) associated with indoor PM concentrations. When controlling for seasonality, indoor fine particulate concentrations were significantly larger in homes using a gas furnace and without central air conditioning. However, these factors did not affect indoor PM_{10} or endotoxins levels. One of the major predictors of indoor PM_{10} and endotoxin levels inside the home was cleanliness. Compared to a residence that scored high on the scale, a home that rated low had a mean increase of 7.8 μg m^{-3} of PM_{10} and 0.12 EU m^{-3} of endotoxin. A positive association (p=0.006) was also observed between indoor endotoxin levels and having a grain storage bin on the property. Adjusting for significant co-variates, homes with a grain bin on the property had a mean increase of 0.08 EU m^{-3}. Smoking was negatively associated with indoor endotoxin concentrations; however only 7% of homes sampled for endotoxin reported smoking in the home.

Discussion

Few published studies have characterized rural populations' air pollution exposure. Therefore, we were interested in comparing indoor and outdoor PM and endotoxin concentrations from an intensely agricultural area to measurements taken in urban centers. Mean concentrations of ambient PM_{10} and $PM_{2.5}$ observed in Keokuk County were approximately 35% smaller than levels recorded across 15 metropolitan sites in the US from 2005-2007.[16] Indoor PM_{10} and $PM_{2.5}$ levels also tended to be smaller than levels found in previous North American urban studies.[40-43] Smoking prevalence among agricultural populations is generally smaller than urban and this may partially account for the decreased levels of indoor PM observed in this study.[44]

In contrast to PM measurements, ambient endotoxin levels measured in the study area were larger than studies conducted in non-rural settings using a similar size selective sampler. Geometric mean endotoxin concentrations in Keokuk County (1.19 EU m^{-3}) were approximately three-times larger than ambient levels found in Southern California (0.44 EU m^{-3}), while endotoxin levels were an order of magnitude larger than measurements recorded in the urban areas of Germany and Sweden (0.05 EU m^{-3}).[29,31,45] Although outdoor endotoxin concentrations were larger in the study area compared to urban areas, indoor levels (0.21 EU m^{-3}) were on the same order of magnitude as concentrations found in Baltimore (0.13 EU m^{-3}; PM_{10} sample)[46], Paris (0.512 EU m^{-3} and 0.553 EU m^{-3}; total dust sample)[47], and Boston (0.77 EU m^{-3}; total dust sample)[48]. Geometric mean endotoxin concentrations in Keokuk County were similar to levels found in rural-Canada, which sampled 146 homes over five days during the winter (indoor=0.14 EU m^{-3} vs. outdoor=0.12 EU m^{-3}) and summer (indoor=0.47 EU m^{-3} vs. outdoor=1.57 EU m^{-3}) of 2007 using a coarse PM sampler.[49] However, maximum five-day concentrations observed in this study were larger than levels found in Canada, with outdoor levels in Keokuk County reaching 13.00 EU m^{-3}compared to 6.41 EU m^{-3}.[49]

We expected airborne PM_{10} levels to be significantly larger during the autumn, when row-crop harvesting generates large amounts of airborne dust. Results show that PM_{10} concentrations outside the home were not significantly increased and five-day mean concentrations were comparable to other seasons. Although mean levels were not significantly affected, autumn had the largest range (6.2-56.2 μg m^{-3}) and GSD (1.84) of any season. This large variation was also reflected when PM_{10} measurements were stratified by quartiles. Only 26% of the PM_{10} measurements were recorded in the autumn, yet 38% of measurements were in the upper quartile, while 40% of measurements were in the lower quartile. This finding indicates that during certain times in autumn, ambient levels of PM_{10} can be elevated, but quickly return to background levels, usually within a week. Future rural air quality studies may benefit from a shorter sampling period and identification of local agricultural activities in order to achieve better temporal resolution to determine peak exposures during harvest season.

Unlike PM concentrations, ambient endotoxin concentrations were significantly larger during autumn, a finding that is unique to this study. Two previous urban air studies found no significant increase in endotoxin concentrations during this season.[45,50] A study

conducted outside Munich, Germany observed a strong positive correlation between ambient temperature and increased endotoxin levels, with peak concentrations occurring during June and July, while mean concentrations in the autumn were comparable to levels found in the winter time.[50] Additionally, a 2004 study conducted in Southern California found no seasonal pattern in endotoxin concentrations. [45] Although more data are needed to assign causality, harvesting appears to be responsible for this seasonal trend, since urban studies did not find elevated concentrations of endotoxin during the autumn.

A major goal of this study was to determine if agricultural variables were predictive of indoor and outdoor PM_{10}, $PM_{2.5}$, and endotoxin concentrations. One of the most significant factors influencing airborne PM_{10} and endotoxin levels inside homes was the qualitative assessment of home cleanliness. This is consistent with previous studies which have found home cleanliness, assessed either through questionnaire data or interviewer rated, was associated with decreased levels of endotoxin in settled dust and airborne samples.[46,47,51,52] Adjusting for significant covariates, homes that scored in the lowest of the three categories for home cleanliness had an average increase of 7.8 μg m^{-3} of PM_{10} and 0.12 EU m^{-3} of endotoxin compared to homes rated cleanest. This finding has potential implication for children's health. An epidemiological study of asthmatic children in inner-city Baltimore, found a significant increase in the incidence of cough, wheezing, and chest tightness for every 10 μg m^{-3} of $PM_{2.5-10}$.[53] While a study using total dust samplers, conducted in Prince Edward, Canada, found an increase of 0.49 EU m^{-3} was significantly associated with larger incidences of respiratory illnesses in children below the age of two years.[54] Compared to total dust samplers, PM_{10} samplers may underestimate endotoxin concentrations and consequently health effects may be detected at lower concentrations. Although home cleanliness was found to be a significant predictor of PM_{10} and endotoxin it only explained 4% and 10% of the variability in indoor measurements, respectively. Consequently, visual inspection alone would not serve as a surrogate for quantitative exposure measurements.

Another factor which was shown to significantly increase indoor endotoxin levels was the presence of grain storage bins on the property. Since outdoor levels were unaffected, grain bins may be a source of take-home exposure. Multiple agricultural studies have shown increased levels of pesticides inside rural households from take-home sources.[55-57] Recently, a study from the United Kingdom found larger levels of flour dust, an allergic sensitizer associated with occupational asthma, inside bakers' homes compared to non-bakers.[58] In the present study it is not clear whether larger endotoxin levels are associated with grain bins themselves or whether the bins are a proxy for unmeasured agricultural variables. Although more work is needed to determine the source, greater education among farmers about improved hygiene practices may decrease indoor endotoxin levels.

Gravel roads are often a source of nuisance dust in rural areas and can negatively impact EPA PM_{10} attainment status.[59] In multivariate modeling no significant increase in ambient PM_{10} was observed in samples collected outside homes located on unpaved roads. The lack of a significant increase was likely due to low vehicle traffic in the county (less than 100 vehicles per day)[60] and five-day averaging time. Findings from this study suggest that paving rural roads in low-vehicle traffic areas would do little to reduce ambient PM_{10}

exposure near homes and would not be beneficial given the increased costs of maintenance and construction.

This study had several limitations including non-specific survey questions to categorize exposure variables, potential underestimation of outdoor endotoxin levels, possible lack of generalizability due to the recruitment strategy of households, and small sample size for certain household characteristics. First, the lack of specificity in the environmental questionnaire may have caused possible misclassification of residential and agricultural variables. For example, regarding smoking status inside the home, participants were asked if household members or guests ever smoke in the residence. However, it was not known whether individuals smoked during the time of the sample collection. Consequently, estimation of the effect of predictors on concentrations of particulate and endotoxin may have been attenuated due to misclassification. Second, only the coarse fraction of the outdoor particulate sample was analyzed for endotoxin. As a result, this may have underestimated rural populations' exposure to airborne endotoxin. Third, households were recruited into the study through non-random sampling. This may limit the generalizabilty of this study if fundamental differences exist between homes selected for assessment and the underlying eligible population. Also, we could not account for changes in ambient PM and endotoxin concentrations by different years, since homes were not sampled in all season every year. Finally, smoking and the use of biomass for residential heating has been associated with increased indoor endotoxin levels in previous studies.[61-63] However, due to the small number of participants who smoked or burned biomass, we were unable to achieve enough power or large enough sample size to generalize results found in this study to the larger rural-population.

Conclusions

Results from this study show ambient endotoxin concentrations in an agricultural county in the Midwest US were elevated compared to those previously reported in urban areas; however, indoor and outdoor PM_{10} and $PM_{2.5}$ concentrations were smaller. Contrary to our initial hypothesis, there was no significant increase in five-day averaged outdoor PM_{10} during the harvest season. Conversely, concentrations of ambient endotoxin were significantly increased, a finding that seems unique to rural areas. In general, agricultural and housing variables were found to be poorly associated with indoor and outdoor PM_{10}, $PM_{2.5}$, and endotoxin concentrations. One variable that was found to be highly associated with indoor PM_{10} and endotoxin was our qualitative assessment of home cleanliness. Compared to a residence that scored high on the scale, a home that rated low had a mean increase of 7.8 μg m^{-3} of PM_{10} and 0.12 EU m^{-3} of endotoxin. This study demonstrated that a complete evaluation of exposures to $PM_{2.5}$ and endotoxin among residents of agricultural communities of the Midwest United States should incorporate both indoor and outdoor measurements.

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Table 1

Characteristics of the homes surveyed

Variable	n (%)
Homes surveyed	197
Homes re-sampled	15 (8)
Home designation	
Rural	140 (71)
Town	57 (29)
Type of housing	
Single-family home	175 (89)
Trailer	22 (11)
Road surface	
Paved	88 (45)
Gravel	109 (55)
Year of home construction	
Before 1900	27 (13)
1900-1949	76 (39)
1950-1969	31 (16)
1970-later	63 (32)
Smoking in the home	
Yes	18 (9)
No	179 (91)
Stove type	
Gas	63 (32)
Electric	134 (68)
Heating source	
Gas	148 (75)
Electric	16 (8)
Biomass	18 (9)
Fuel oil	9 (5)
Geo-thermal	4 (2)
Solar	1 (1)
Indoor dog/cat	
Yes	58 (29)
No	139 (71)

Table 2

Summary of indoor and outdoor PM_{10}, $PM_{2.5}$, and endotoxin concentrations

Pollutant	Location	N	Range*	Mean*	GM*	GSD	Median indoor/outdoor (I/O) ratio	Paired t-Tests *P*
PM_{10}	Indoor	203	4.1-173.3	26.5	21.2	1.91	1.08	0.086
	Outdoor	186	6.2-56.2	21.1	19.6	1.52		
$PM_{2.5}$	Indoor	199	1.4-187.7	16.2	12.2	2.05	1.45	<0.001
	Outdoor	182	1.5-24.1	9.1	8.2	1.58		
Endotoxin	Indoor	117	0.01-4.52	0.32	0.21	2.51	0.16	<0.001
	Outdoor	117	0.02-13.00	1.93	1.19	2.93		

*PM concentrations in μg m^{-3}; endotoxin concentrations in EU m^{-3}

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Table 3

Pairwise comparison of re-sampled homes by location and particulate matter size

Location	Pollutant	n	*p*
Outdoor	PM_{10}	13	0.436
	$PM_{2.5}$	12	0.190
Indoor	PM_{10}	15	0.146
	$PM_{2.5}$	12	0.380

Table 4

Bivariate analysis of outdoor PM_{10}, $PM_{2.5}$, and endotoxin by season

Season	PM_{10}		$PM_{2.5}$		Endotoxin	
	n	GM*	n	GM*	n	GM*
Winter	36	19.2[a]	48	10.6[a]	22	0.39[a]
Spring	48	18.8[a]	53	8.7[ab]	22	0.81[b]
Summer	53	20.9[a]	46	7.9[b]	37	1.33[b]
Autumn	49	19.2[a]	35	6.8[b]	36	2.63[c]

Tukey-Kramer multiple comparison tests using log transformed data. Same letters indicate no significant difference ($p > 0.05$) in the GM.

*PM concentrations in μg m^{-3} and endotoxin concentrations in EU m^{-3}.

Table 5

Multivariate analysis of outdoor log-transformed PM_{10}, $PM_{2.5}$, and endotoxin concentrations by major predictors

Variable	PM_{10} µg m^{-3}		$PM_{2.5}$ µg m^{-3}		Endotoxin EU m^{-3}	
	β	*p*	β	*p*	β	*p*
Intercept	1.987	<0.001	0.821	<0.001	1.049	0.002
Wind speed (m s^{-1})	−0.041	<0.001	−0.038	0.004	0.070	0.032
Precipitation (1 cm)	NS		NS		NS	
Relative humidity (%)	−0.008	<0.001	0.003	0.035	−0.017	<0.001
Season		0.030		0.004		<0.001
Winter	0.119	0.004	0.219	<0.001	Reference	
Spring	0.040	0.305	0.179	<0.001	0.139	0.185
Summer	0.008	0.802	0.030	0.435	0.528	<0.001
Autumn	Reference		Reference		0.780	<0.001
Agricultural household	0.073	0.019	NS		NS	
Home located on un-paved road	NS		NS		NS	
Grain storage bins on property	NS		NS		NS	
Cattle or swine raised on property	NS		NS		NS	
Swine confinement on property	NS		NS		NS	

NS variable's overall $p > 0.05$

Table 6

Multivariate analysis of indoor log-transformed PM_{10}, $PM_{2.5}$, and endotoxin concentrations by major predictors

Variable	PM_{10} μg m^{-3}		$PM_{2.5}$ μg m^{-3}		Endotoxin EU m^{-3}	
	β	*P*	β	*P*	β	*P*
Intercept	0.853	<0.001	0.364	0.051	−1.010	<0.001
Indoor relative humidity (%)	0.004	0.003	0.009	<0.001	NS	
Indoor CO_2 concentration (ppm)	NS		NS		NS	
Log outdoor PM_{10}	0.296	0.007	—		—	
Log outdoor $PM_{2.5}$	—		0.667	0.003	—	
Log outdoor endotoxin	—		—		0.282	<0.001
Home cleanliness		0.016	NS			0.001
Low	0.134	0.006			0.311	<0.001
Medium	0.090	0.034			0.198	0.016
High	Reference				Reference	
Indoor dog and/or cat	NS		NS		NS	
Smoking inside home	0.203	0.002	0.273	<0.001	−0.300	0.035
Gas stove	NS		NS		NS	
Gas furnace	NS		0.105	0.021	NS	
No central air conditioning	NS		0.145	0.003	NS	
Season	NS			0.008	NS	
Winter			0.104	0.174		
Spring			0.208	<0.001		
Summer			0.060	0.267		
Autumn			Reference			
Agricultural household	NS		NS		NS	
Home located on un-paved road	NS		NS		NS	
Grain storage bins on property	NS		NS		0.201	0.006
Non-confined cattle or swine raised on property	NS		NS		NS	
Swine confinement on property	NS		NS		NS	

NS variable's overall $p > 0.05$; — variable not included in analysis