Predictors of Airborne Endotoxin in the Home

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We identified home characteristics associated with the level of airborne endotoxin in 111 Boston-area homes enrolled in a cohort study of home exposures and childhood asthma, and we developed a predictive model to estimate airborne endotoxin. We measured endotoxin in family-room air and in dust from the baby's bed, family room, bedroom, and kitchen floor. Level of airborne endotoxin was weakly correlated ($r < 0.3$) with level of endotoxin in each of the four types of dust samples and was significantly correlated with endotoxin in family-room dust ($p < 0.05$). Endotoxin in family-room dust accounted for < 6% of the variability of airborne endotoxin. In a multivariate model, certain home characteristics were positively ($p < 0.05$) associated with airborne endotoxin. These included current presence of dog (difference in level, dog vs. no dog = 72%, partial $R^2 = 12.8\%$), past presence of dog (partial $R^2 = 5.5\%$), and endotoxin level in family-room dust (partial $R^2 = 5.3\%$). Use of a dehumidifier (partial $R^2 = 6.4\%$) was negatively associated ($p = 0.02$; difference = –31%) with airborne endotoxin. Other home characteristics were identified as important determinants of increased airborne endotoxin in this model, but individual coefficients were not statistically significant ($\alpha = 0.05$): total amount of fine dust collected in the home (partial $R^2 = 3.8\%$), concrete floor in family room (3.7%), water damage (3.6%), and use of cool-mist humidifier in past year (2.7%). This multivariate model explained 42% of the variability of airborne endotoxin levels, a substantial improvement over that with dust endotoxin alone. Airborne endotoxin in Boston-area homes appears to be determined by the presence of dogs, moisture sources, and increased amounts of settled dust. *Key words*: airborne endotoxin, dust endotoxin, predictive model, seasonal variability. *Environ Health Perspect* 109:859–864 (2001). [Online 14 August 2001]
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The prevalence of asthma has been increasing in the United States since the 1970s (*1,2*); however, the causes of the increase are not clearly understood. Some researchers have suggested that the increase might be caused by changes in the indoor environment and have reported the association of home characteristics with respiratory symptoms or physician-diagnosed asthma (*3–6*). Among home characteristics investigated, those that have been reported as significant predictors of respiratory symptoms and disease include home dampness, humidifier use, mold/mildew, water damage, and parental smoking (*3–6*). These predictors, however, may not be direct measures of exposure to target agents causing or aggravating disease, but instead are likely to be surrogate measures of exposure to something in air, such as allergens, endotoxin, fungal toxins, or particles from tobacco smoke.

Endotoxin is the biologic activity of lipopolysaccharide (LPS) which is a component of the outer membrane of gram-negative bacteria. Endotoxin has been long recognized as a health hazard for various occupations. Exposure to endotoxin by inhalation is a cause of humidifier fever (*7*) and of acute and chronic airflow limitation in cotton mill workers (*8,9*) and among agricultural workers (*10–13*). Endotoxin has also been detected in house dust and associated with severity of childhood and adult asthma (*14–17*). Recently, Von Ehrenstein et al. (*18*) have hypothesized that endotoxin may protect children in agricultural environments from onset of asthma. Endotoxin has numerous proinflammatory effects and can induce airway inflammation (*19,20*). The home environment may affect survival, growth, and proliferation of gram-negative bacteria. Therefore, endotoxin in the home may be associated with home characteristics. However, the number of reports about potential sources for endotoxin in the home or home characteristics associated with endotoxin is limited (*21–23*).

Measurement of endotoxin in dust has been used as a convenient way to measure exposure to endotoxin in the home (*14,15,17*) because house dust is easy to collect and assay. Another possible advantage of this measurement is that dust endotoxin could represent cumulative exposure to endotoxin because it may accumulate in settled dust. However, this is only a surrogate measure of airborne endotoxin. If airborne endotoxin at home represents true exposure, respiratory disease may be better correlated with a more direct measure of the inhaled dose, such as level of airborne endotoxin.

In some situations, however, measurement of airborne endotoxin in every home may not be practical, and house dust endotoxin may be the best surrogate for exposure. In these cases, combining measurement of dust endotoxin with other information about home characteristics may provide a better estimate of exposure to airborne endotoxin.

In this study, we hypothesized that certain home characteristics, especially those related to dampness, animals, and smoke exposure, would be associated with airborne endotoxin measured in a subset of homes from the Epidemiology of Home Allergens and Asthma Study (*24*). We then developed a predictive model to estimate airborne endotoxin on the basis of endotoxin in settled house dust and those home characteristics. The resulting multivariate regression model will be applied to an ongoing epidemiologic study, investigating the association of exposure to endotoxin in the home with health effects, to predict the level of airborne endotoxin in homes from measurements of dust endotoxin only and home characteristics.

Methods

Study cohort. The Epidemiology of Home Allergens and Asthma Study is an ongoing, longitudinal, closed birth-cohort study of children born to parents with histories of allergies and/or asthma. The aim of the study is to examine the role of exposure to indoor home allergens in the development of asthma/wheeze and allergic sensitization in early childhood. The Home Endotoxin and

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Childhood Asthma Study has supplemented this cohort study by examining the role of endotoxin exposure in these outcomes. A detailed description of the cohort (505 children from 499 families) was published previously (*24*). In brief, between September 1994 and June 1996, we obtained a daily list of all women who had just delivered babies at the Brigham and Women's Hospital and who lived in the Greater Boston area within Route 128. In screening, we included only mothers 18 years or older who did not plan to move in the next 12 months. We excluded newborn babies who were premature (< 36 weeks), had major congenital abnormalities, or were hospitalized in the neonatal intensive care unit.

Home visits and endotoxin sample collection. We visited the homes of all 499 infants within 2–3 months (first home visit) after the birth of the index child. If the first home visit was between 1 December 1994 and 1 May 1995 or between 1 November 1995 and 30 April 1996 and the family had not moved and had agreed to a second visit, we visited the home a second time 6–8 months after the first home visit. If the family moved during the study, we visited the new home. Table 1 shows the number of homes and visits made for the study. At every home visit, we collected four dust samples and administered a detailed questionnaire about home characteristics that included questions about type of building, use of humidifier and dehumidifier, carpeting, dampness and water damage, mold, presence of pets, frequency of cleaning, children's bedroom environment, other home characteristics, and parents' education and income.

We used a Eureka Mighty-Mite vacuum cleaner (Model 3621; Eureka Co., Bloomington, IN) modified to hold 19 × 90 mm cellulose extraction thimbles to collect house dust. We collected four dust samples in each home. In the bedroom, we vacuumed 2 m^2 of the bedroom floor surrounding the baby's crib for 5 min. For bed dust, we vacuumed for 5 min all layers of bedding in the baby's crib, or the parents' bed if the index child slept there more than 50% of the time. In the family room, we vacuumed the seat cushion, arms, and back of the upholstered chair where the baby spent the most time (for 2.5 min) along with 2 m^2 of the surrounding floor (for 2.5 min). In the kitchen, we vacuumed for 5 min the edges of the floor under cabinets, around the refrigerator, and under the sink. Within 24 hr after collection, we weighed and sifted the dust through a 425-µm mesh sieve. We reweighed the fine dust and made aliquots for various analyses–allergens, culturable fungi, and endotoxin. An assay for endotoxin was done only if sufficient dust remained after all other assays had been performed.

We collected air samples in a subgroup of homes (166 of 499 homes; 33%) between April 1995 and April 1997. The subgroup consisted of homes in which the family agreed to allow an air-sampling pump to be left and in which the field staff considered it safe to leave the samplers to be fetched later. The subgroup was identified among the homes scheduled for a second home visit and among the homes first visited during the last year of subject enrollment. We sampled in the family room for an average of 1.5 days using a Gilian pump (model HFS 513A; Gilian Instrument Corp., West Caldwell, NJ) attached to a filter cassette assembled with a 0.4-mm preweighted polycarbonate filter. Each assembly was precalibrated at a flow rate of 2 L/min with a Gilian soap bubble flowmeter (P/N 800286; Gilian Instrument Corp.) before and after sampling. After sampling, we weighed the filters for total suspended particulate (TSP) analysis with an electrobalance (model Cahn 21; Cahn Instrument Inc., Cerritos, CA) at 65–75°F, 35–45% relative humidity, and then assayed them for endotoxin.

We also prepared 98 blank filters for quality control of air sampling (on average, 4 blank filters/month). Blank filters were subjected to all the procedures of precalibration, postcalibration, storage, and assay. If the blank filters indicated possible contamination during calibration, shipping, and storage, we excluded the air samples collected between one day before the date the contaminated blank was collected and the day of next clean blank. We excluded 50 air filters from data analysis because of contamination that occurred during calibration when the soap solution in the calibrator was contaminated with gram-negative bacteria. Table 2 shows the final number samples of airborne endotoxin available for data analysis; the number of samples includes five duplicate

Table 1. Number of homes and visits.

Home[a]	Visit 1	Visit 2	Total
1	499	198	697
2	103	1	104
3	10	0	10
Total	612	199	811

[a]Home 1: the original home the family lived in during the study period; home 2 or 3: the second or third home occupied by families that moved during the study period.

Table 2. Number of airborne endotoxin samples available.

Home[a]	Visit 1	Visit 2	Total
1	41	66	107
2	8	0	8
3	1	0	1
Total	50	66	116

[a]Home 1: the original home the family lived in during the study period; home 2 or 3: the second or third home occupied by families that moved during the study period.

Table 3. Airborne endotoxin samples matched with family room dust endotoxin samples.

Home[a]	Visit 1	Visit 2	Total
1	36	50	86
2	6	0	6
3	1	0	1
Total	43	50	93

[a]Home 1: the original home the family lived in during the study period. Home 2 or 3: the second or third home occupied by families that moved during the study period.

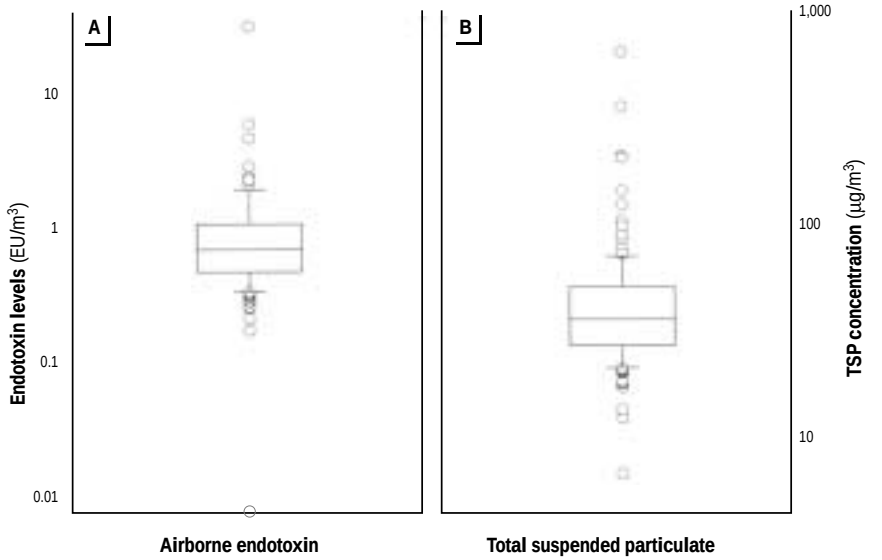

Figure 1. Box plots of (*A*) airborne endotoxin and (*B*) total suspended particulate (TSP) concentration. The lower and upper boundaries of the box indicate the 25th and 75th percentiles, respectively. The line within the box marks the median, and whiskers above and below the box indicate the 90th and 10th percentiles. Circles are outliers. The family room median airborne endotoxin is 0.72 EU/m^3 (n = 116) and median TSP is 38 µg/m^3 (n = 159). The number of samples includes five duplicate measurements from both visit 1 and 2 for five homes.

measurements from both visit 1 and 2 for five homes. Table 3 shows the number of samples of airborne endotoxin matched with the family-room dust endotoxin. We did not obtain sufficient family-room dust to assay for endotoxin from 23 homes where we collected air samples (Table 3).

Endotoxin assay. We measured the endotoxin activity in dust and air samples by the kinetic Limulus assay with resistant-parallel-line estimation (KLARE) method, described previously (*25*,*26*). We obtainted Limulus amebocyte lysate (LAL) from BioWhittaker (Walkersville, MD), reference standard endotoxin from the United States Pharmacopoeia, Inc. (Rockville, MD), and control standard endotoxin from Associates of Cape Cod (Woods Hole, MA). All glassware was heated to 270°C for 30 min before use. Control and reference standards and field samples were serially diluted for endotoxin analysis in a standard buffer (0.01% triethylamine and 0.05 M potassium phosphate).

For dust samples, we placed 25 mg of sifted dust in endotoxin-free borosilicate tubes with 5 mL of buffer and bath sonicated for 1 hr with vortexing at 15-min intervals. An initial 1:25 dilution of dust extracts with suspended particulate was made before the start of the serial dilutions used in the assay. For air samples, we added 5 mL of standard buffer to endotoxin-free test tubes with the filter sample and bath sonicated it for 1 hr. We made no initial dilution before serial dilution for air samples. We also assayed four serial dilutions (1:6) for both dust and air samples.

We placed duplicate 50-µL aliquots of the initial dilution, four serial dilutions of dust or filter extracts, and a control standard endotoxin in an endotoxin-free 96-well, flat-bottomed polystyrene microplate (Associates of Cape Cod); 50 µL of LAL was added, and the microplate was agitated. We recorded the optical density (OD) of each well at 405 nm every 30 sec for 120 min during incubation at 37°C. The response parameter for the LAL reaction was the maximum rate of OD change (V_{max}). We computed the log potency and its variance using the method developed by Milton et al. (*25*). Results were reported in endotoxin units (EU) with reference to EC5 or EC6 reference standard endotoxin (U.S. Pharmacopoeia, Inc.; 1 ng EC5 and EC6 = 10 EU).

In a previous report (*26*), we showed that the sensitivity of LAL to environmental endotoxin was different by lots within the same manufacturing company. Therefore, we tested and compared the sensitivity of 12 LAL lots used in the study and adjusted the final EU of samples that were assayed with the LAL lots significantly different from the lot used to assay the largest number of samples.

Data analysis. Home endotoxin, TSP, and mass of dust collected showed right-skewed distributions. Therefore, we performed log transformation of those measurements to obtain symmetrical, approximately Gaussian distributions and used the log-transformed data in all analyses. We constructed continuous, binary, or categorical variables for home characteristics on the basis of the questionnaire results.

We computed both Pearson and Spearman correlation coefficients to examine the correlation of airborne endotoxin with dust endotoxin measurements, TSP, and total amount of fine dust collected at home. We present only the Pearson correlation because log-transformed airborne endotoxin and other measurements were Gaussian (Shapiro-Wilk normality test) or approximately Gaussian, and the two methods (Pearson and Spearman) gave similar results.

We used fixed-effect models (*27*) in univariate and multivariate regression analyses of potential associations of airborne endotoxin level with home characteristics (SAS Proc GLM, SAS Institute Inc., Cary, NC). To examine seasonal effects on airborne endotoxin level, we used univariate and multivariate fixed-effect models, controlling for some important home characteristics. We matched all home characteristics examined for association with airborne endotoxin by visit number and defined them as discrete variables (yes/no or category) except for endotoxin measurements (EU per milligram); total amount of fine dust collected (milligrams per home); and number of people per room, which were continuous. In these regression analyses, we dropped two influential outliers, the highest and lowest measurements of airborne endotoxin.

We developed a predictive model for airborne endotoxin, which we selected by manual stepwise backward elimination. At each step, we eliminated the regressor with the highest *p*-value for its coefficient. We chose the final model using internal cross-validation by finding the model with the smallest predicted residual error sum of squares (PRESS statistic) (*28*,*29*). The candidate predictors were the individual home characteristics that had been significant in univariate models ($p < 0.1$) or home characteristics that we suspected, on the basis of published reports, might contribute to airborne endotoxin. We tested the linearity of the relationship between airborne endotoxin and endotoxin in family-room dust in the final model by a generalized additive model (S-plus; MathSoft, Seattle, WA) with cubic splines (cross-validation) after controlling for all other covariates.

Results

Distribution and correlation of airborne endotoxin with other measurements. Airborne endotoxin and TSP concentrations are best described by a log-normal distribution (Figure 1). The geometric mean (GM) of the indoor airborne endotoxin level was 0.77 EU/m^3 (GSD = 2.3, n = 116; range, 0.01–30.23 EU/m^3). The GM of the TSP concentration was 40 µg/m^3 (GSD = 1.8, n = 159; range, 7.4–639.0 µg/m^3). The GM of the endotoxin level in family-room dust available from all homes and all visits was 83 EU/mg (GSD = 2.0, n = 589; range, 2.1–2,405 EU/mg). The GM of the endotoxin level in family room dust matched with airborne endotoxin samples was 96 EU/mg (GSD = 2.1, n = 93; range, 28–1,945 EU/mg).

Airborne endotoxin in the family room was weakly correlated with endotoxin level in all four house dust endotoxin samples but was significantly associated with endotoxin level only in family-room dust (Table 4). A plot of airborne endotoxin and endotoxin in family-room dust is shown in Figure 2. The total amount of fine dust collected at home was also significantly correlated with TSP concentration and level of airborne endotoxin (Table 4). Correlation between airborne endotoxin and TSP was weak but highly significant ($p < 0.005$).

Table 4. Correlation coefficients[a] between airborne endotoxin and other measurements.

TSP and total endotoxin level	TSP	Airborne[b] endotoxin	Total house dust collected[c]
TSP [µg/m^3]	—	0.29*	0.27*
	(159)[d]	(112)	(159)
Airborne endotoxin [EU/m^3]	—	—	0.20*
		(116)	(116)
Bed dust [EU/mg]	–0.48	0.29	—
	(11)	(10)	
Bedroom floor dust [EU/mg]	–0.12	0.23	—
	(90)	(67)	
Kitchen floor dust [EU/mg]	0.06	0.11	—
	(59)	(43)	
Family-room dust [EU/mg]	0.08	0.21*	—
	(127)	(93)	

Abbreviations: EU, endotoxin units; TSP, total suspended particulate. [a]Pearson correlation coefficient: *$p < 0.05$. [b]Airborne endotoxin was sampled in family room. [c]Total house dust collected [log(mg/home)]: sum of collected amount of fine dust over all four rooms, which may represent overall dirtiness of home. [d]Number of samples.

Effect of season on airborne endotoxin level and TSP. We did not observe a significant seasonal variation in level of airborne endotoxin. Neither a univariate model nor a multivariate fixed-effect model controlling for home characteristics suggested a significant seasonal effect on level of airborne endotoxin. We did not observe a seasonal effect on TSP concentration.

Home characteristics and airborne endotoxin level. We tested 49 home characteristics to determine whether they were predictors of level of airborne endotoxin. The univariate R^2 for tested individual home characteristics ranged between < 0.001 and 0.13. Table 5 presents the significant univariate predictors ($p < 0.1$) and all candidate covariates selected for use in developing a multivariate model. Level of airborne endotoxin was significantly and positively correlated with current presence of a dog (the strongest predictor of increased level of airborne endotoxin), past presence of a dog, any sign of mice, water damage in the past year, and amount of total house dust collected. Dehumidifier use was significantly but negatively associated with airborne endotoxin. Cool-mist humidifier use and a concrete floor in the family room were not significant univariate predictors (Table 5), although they appeared to contribute to the predictive power of the multivariate model (Table 6). Cigarette smoke was not associated with elevated endotoxin, but smoking was uncommon in our study homes.

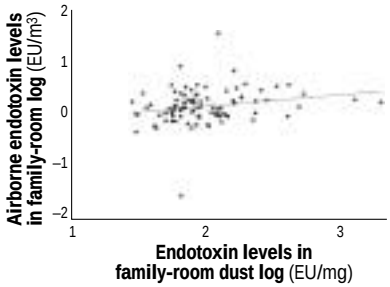

Figure 2. Scatter plot of airborne endotoxin and endotoxin in family-room dust with regression line (R^2 = 0.04, slope = 0.25).

Development of predictive models for airborne endotoxin level. We used endotoxin measured in family-room dust as our primary surrogate for airborne endotoxin because airborne endotoxin was sampled in the family room. We then developed a multivariate model using stepwise backward elimination starting with a model including all candidate variables listed in Table 5, as described in "Methods." The final model selected had R^2 of 0.42, adjusted R^2 of 0.34, and *p*-values for each estimated coefficient smaller than 0.2, and minimized the predicted residual sum of square (PRESS) statistic.

Table 6 shows the magnitude of effects associated with the parameters and the partial R^2 for each predictor after adjusting for other variables in the final model. Current presence of a dog in the home remained the strongest predictor in the multivariate model. Endotoxin level in family-room dust remained a significant, but still weak, predictor for airborne endotoxin. The amount of total fine dust collected, water damage in the past year, concrete floor in the family room, and cool-mist humidifier use in the past year had marginal positive associations with airborne endotoxin but were retained as predictors contributing to predictive power of the multivariate model. Dehumidifier use had a similar magnitude and a significantly negative association with airborne endotoxin levels in univariate and multivariate models. The relationship between airborne endotoxin and endotoxin in family-room dust did not deviate significantly from linearity after adjustment for other covariates (*p*-value of test for linearity = 0.24).

Table 5. Univariate predictors[a] of airborne endotoxin level.

Home characteristics	Level	*n*	Mean[b] (EU/m^3)	Percent difference[c]	95% CI LL	95% CI UL	Model R^2 (%)
Total house dust collected	Cont	114	—	30	11	53	8.4
Dust endotoxin in family room	Cont	91	—	18	2	37	5.3
Number of people/room	Cont	114	—	9	–5	26	1.3
Dog in home	Current	16	1.28	96	42	169	15.2
	Past	25	0.92	41	8	84	
	Never	73	0.65				
Use of dehumidifier in home	Y	22	0.56	–34	–50	–12	6.5
	N	92	0.84				
Any sign of mice, past year	Y	19	1.10	53	13	108	6.4
	N	95	0.72				
Water damage in building, past year	Y	43	0.93	32	4	68	4.7
	N	67	0.70				
Mold/mildew in bedroom	Y	3	1.24	61	–22	233	1.5
	N	109	0.77				
Cool-mist humidifier use in home	Y	52	0.85	16	–8	46	1.4
	N	60	0.73				
Continuously burning pilot flame	Y	35	0.86	16	–11	49	1.1
	N	71	0.74				
Any sign of cockroach, past year	Y	13	0.65	–17	–43	20	0.9
	N	100	0.79				
Concrete floor on family room	Y	7	0.93	24	–23	101	0.8
	N	103	0.75				
Ever used carpet freshener in home	Past year	11	0.78	3	–31	54	0.8
	Past month	12	0.91	20	–18	77	
	Never	90	0.76				
Smoking inside home	Y	6	0.84	8	–36	83	0.1
	N	107	0.77				
Stuffed animals in room > 4	Y	62	0.76	–3	–24	22	0.1
	N	52	0.79				
Living in apartment (≥ 3 homes)	Y	30	0.78	1	–22	32	0.0
	N	84	0.77				

Abbreviations: Cont, continuous; LL, lower limit; UL, upper limit; Y, yes; N, no.
[a]Significant univariate predictors and other candidate predictors for developing multivariate predictive model. Two influential outliers were excluded in the data analysis. [b]Mean airborne endotoxin concentration (EU/m^3) at each level of categorical variables. [c]Percent change in airborne endotoxin level relative to "no" category for categorical variable, and corresponding to an interquartile difference (0.37 EU/mg for log dust endotoxin, 0.44 mg/home for log total fine dust, and 0.25/room for number of persons per room) for continuous variable.

Discussion

Correlation with dust endotoxin and seasonal variation of airborne endotoxin. The finding that airborne endotoxin was only weakly correlated with endotoxin level in family-room dust, where the air samples were collected, suggests that measurement of house-dust endotoxin alone may be only a weak surrogate measure for airborne endotoxin. House-dust endotoxin as a measure of exposure to home endotoxin has been used in several epidemiologic studies (*14–17*). Measurement of dust endotoxin may represent cumulative endotoxin levels, but measurement of airborne endotoxin has the advantage of possibly being a direct measure of inhalation exposure. If so, the use of house-dust endotoxin alone as a surrogate for inhaled exposure may attenuate the measure of association of home exposure to endotoxin with health outcomes in epidemiologic studies (*30*). Thus, we developed a predictive model estimating level of airborne endotoxin using both dust endotoxin level and home characteristics.

We did not observe significant seasonal variation in indoor airborne endotoxin levels. In our longitudinal study of 20 homes (*31*), we also did not observe strong evidence of a seasonal pattern of indoor airborne endotoxin. The lack of a seasonal pattern indoors in the presence of significant seasonal

variation in outdoor airborne endotoxin level described in the previous study (*31*) may suggest that indoor sources are important and that indoor airborne endotoxin level is driven by time invariant indoor factors.

Airborne endotoxin: home characteristics and predictive models. Both current and past presence of a dog were strong positive predictors of airborne endotoxin in the home, remaining strong and significant factors in multivariate models (Table 6), which implied that the presence of a dog was not confounded by other home characteristics. Cats and other pets were not significantly associated with airborne endotoxin. Recently dogs were implicated as important carriers of contaminants from the outdoor environment into the indoor living space (*32*). However, it remains to be determined whether the endotoxin associated with dogs is from bacteria they bring in from outdoors or from bacteria that originate from the dogs themselves. Analysis of the 3-hydroxy fatty acids in dust from homes with and without dogs would allow determination of whether dogs are associated with increased amounts of LPS or with changes in the types of LPS present in the home (*33*,*34*).

Cool-mist humidifier use, concrete floors in the family room, and water damage in the past year were positively associated with airborne endotoxin. All of these factors appear to be related to dampness in the home. On the other hand, dehumidifier use was a negative predictor in both univariate and multivariate models. The presence of a dehumidifier may decrease multiplication of bacteria in dust or other reservoirs by taking moisture out of the environment.

Dampness in the home has been recognized as a strong predictor of children's respiratory disease or severity of respiratory symptoms (*4*,*35*–*37*) and is probably a surrogate measure for exposure to biologic agents such as endotoxin, fungal allergens, and toxins or other allergens (e.g., house dust mites). Reported mold/mildew has been used as an indicator of home dampness (*4*, *37*), but our data did not show that reported mold/mildew was an important predictor of airborne endotoxin (Table 6)—possibly because of lack of power (n = 3). However, water damage was significantly and positively correlated with airborne endotoxin in a univariate model and also was selected as a predictor in our multivariate model. The lack of an association between cigarette smoke and endotoxin may reflect the low rates of smoking in our study. Others have found endotoxin in cigarette smoke (*38*).

The primary objective of this study was to develop a predictive model for airborne endotoxin for this cohort; the second was to investigate determinants or sources of endotoxin in the homes. The small number of homes with certain characteristics in our study, e.g., mold/mildew in the bedroom (n = 3) and smoking inside the home (n = 6), limited the power to determine significance of these factors as contributors to airborne endotoxin. Also, one should be cautious about generalizing from our model to different climates and residential environments, although our predictive model is informative for our study and identifies home characteristics that should be considered in future studies.

Our major findings are that dust endotoxin alone is only a weak surrogate of airborne endotoxin and that the multiple regression model, which included home characteristics in addition to dust endotoxin level, predicted airborne endotoxin level more efficiently than did dust endotoxin alone. Presence of a dog was the strongest predictor of airborne endotoxin levels. In addition, home characteristics related to humidity (dehumidifier or cool-mist humidifier use, water damage, increased amounts of settled fine dust, and concrete floor in the family room) contributed significantly to the predictive power of a multivariate model for level of airborne endotoxin. The multivariate regression model explained up to 42% of the variance in airborne endotoxin levels and may be useful in applying measurement error methods to the estimation of airborne endotoxin levels from dust endotoxin and home characteristics in our study.

If levels of airborne endotoxin provide a more precise measure of endotoxin exposure than do levels of endotoxin in dust, epidemiologic studies (*14*–*17*) that use dust endotoxin as the only index of exposure might underestimate the association of endotoxin exposure and respiratory disease (*30*). Thus, in new epidemiologic studies of endotoxin exposure and respiratory disease it may be advantageous to evaluate not only dust endotoxin but also airborne endotoxin levels. However, the choice of endotoxin measurement in the future studies will need to consider whether to measure current exposure or long-term average exposure. Our recent longitudinal study of endotoxin in a small set of homes (*31*) suggests that bed-dust endotoxin may give the best ability to discriminate between homes when the range of exposure is narrow and long-term average exposure is of interest. Those data would also suggest that when short-term exposure, such as exposure in the first month of life, is of interest then airborne levels would be preferable. Obviously, the most important factor in these study designs will be the overall range of exposures in the study population. Thus, when current airborne exposure is the measurement of interest, an internal validation study and development of predictive models such as ours may prove useful for estimation of airborne endotoxin levels and their relation to respiratory outcomes.

Table 6. Multivariate regression model for predicting level of airborne endotoxin with endotoxin in family-room dust and home characteristic variables.

Predictors in the model	Variable type	Parameter estimates[a] Percent difference	95% CI LL	95% CI UL	*p*-Value	Partial R^2 (%)
Dog currently in home	Binary	72	26	136	< 0.01	12.8
Dehumidifier use in home	Binary	–31	–49	–6	0.02	6.4
Dog previously in home	Binary	35	2	78	0.04	5.5
Log_{10} (family-room dust endotoxin)	Continuous	15	1	31	0.04	5.3
Total fine dust collected at home	Continuous	17	–2	40	0.08	3.8
Concrete floor in family room	Binary	62	–6	180	0.09	3.7
Water damage in home	Binary	22	–3	54	0.09	3.6
Cool mist humidifier use in home, past yr	Binary	19	–6	49	0.15	2.7

Abbreviations: LL, lower limit; UL, upper limit.

[a]Percent change in airborne endotoxin level relative to "no" category for binary variable and corresponding to interquartile difference of log of dust endotoxin (0.37 EU/mg) and log of total fine dust (0.40 mg/home) for continuous variable, after adjusting for other covariates in the model (n = 91 without two influential outliers); 95% CI: LL and UL of 95% CI for percent difference; p-value of two-sided t-test adjusted for other covariates in the model. Total R^2 of this final model is 42%.

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