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ORIGINAL ARTICLE

Childhood Asthma and Indoor Aeroallergens and Endotoxin in Palestine: A Case-Control Study

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This study was carried out to evaluate the relationship between wheezing or sensitization and concentrations of mites, cat and dog allergens, and bacterial endotoxin in Palestine. A nested case-control involved analysis of mattress and floor dust from a 110 children's houses with reported wheezing and without wheezing. We found no consistent associations between allergen levels and either wheeze or specific atopic sensitization. Furthermore, no clear associations between mattress endotoxin levels and wheeze or atopy were found. Endotoxin in floor dust was inversely associated with atopic sensitization and wheeze, statistically significant only for atopic wheeze. Finally, a nonsignificant inverse association was observed between living room endotoxin and atopy within the non-wheezing control group. In conclusion, although our study found mostly negative results, it does suggest that endotoxin on living room floors might protect against atopic wheeze. However, this finding should be interpreted with caution due the relatively small sample size of the study and requires further confirmation.

Keywords asthma, aeroallergens, children, endotoxin, Palestine

INTRODUCTION

Several studies have documented that sensitization to indoor allergens, such as those originating from house dust mites, cockroaches, cats and dogs, is an important risk factor for childhood asthma (1, 2). Nevertheless, the impact of indoor allergen levels on asthma development is still controversial and studies have shown contradictory findings (3–5). Similarly, there is uncertainty about the role and importance of domestic exposure to endotoxin (6). Some studies showed that indoor endotoxin exposure was associated with asthma exacerbation in both adults and children (7, 8). Conversely, studies in farming environments showed that house dust endotoxin exposure was *inversely* related to the occurrence of hay fever, atopic asthma, and atopic sensitization in children (9). Also, an inverse association between atopic asthma and occupational exposure to endotoxin was found in adult farmers (10). Most of these studies were carried out in industrialized countries, and very little information is available from developing countries.

As part of the International Study for Asthma and Allergies in Childhood (ISAAC), we have previously shown, in a cross-sectional study in the Ramallah district in Palestine, that the prevalence of wheeze in the past 12 months amounted to 8.2%, 7.2%, and 12.6% for children residing in villages, cities and refugee camps, respectively (11). Nested in this study, we carried out a case-control study to explore the associations between a number of traditional risk factors and asthma (12). We found, among other things, that a positive skin prick test (SPT) against house dust mites and cockroaches was positively associated with wheeze and asthma; the presence of specific immunoglobulin (Ig)E against house dust mites and cat allergens was associated with an increased risk for wheeze. Previously, we also assessed the extent to which the degree of contamination by allergens and endotoxin depended on the location of the houses (cities, refugee camps, or villages), and various home characteristics (13).

In the current study, we collected dust from the homes of a subsample of the children, approximately half of whom had symptoms of wheeze. Four allergens (from house dust mite, Der p1 and Der f1; cat, Fel d1; and dog, Can f1), as well as bacterial endotoxin were measured in dust collected from the child's mattress and the living room floor of 110 houses (63 cases and 47 control subjects). In this paper we present the association between indoor levels of domestic aeroallergens and endotoxin and wheeze in the past 12 months and atopy.

MATERIAL AND METHODS

Study Design and Protocols

The initial prevalence and case-control studies have been described previously in detail (11, 12). In brief, based on a parent-administered standardized questionnaire in 3,382 children 6 to 12 years of age 298 reported wheezing in the previous 12 months (cases). Of those, 273 children had approval of their guardians to participate in the subsequent studies, and they were matched by school location, class, and sex, with an equal number of controls, i.e., children without reported wheezing (ever or in the previous 12 months) and without physician-diagnosed asthma. Of these 546 children, 420 children had approval of their guardian to participate in

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the full case-control study. Eventually, this study comprised 191 cases and 184 control subjects who completed the full study protocol (12). Procedures used in the current study were based on the International Study for Asthma and Allergies in Childhood (ISAAC) phase II study protocols (14). These procedures entailed skin prick tests (SPT) with eight allergens (Der p1, Der f1, *Alternaria tenuis*, tree pollen mix, grass pollen mix, Fel d1, Can f1, olive tree pollen, cockroach; Soluprick SQ, ALK, Hørsholm, Denmark), and venipuncture for the measurement of total IgE and allergen-specific IgE for the same allergens using an enzyme-linked fluorescent assay (Vidas, bioMérieux, sa, Lyon, France).

For dust collection, due to our limited resources, we could not collect environmental samples for all cases and controls in the study; however, we randomly selected 132 children for a home visit (66 cases and 66 controls). Since we had found in the baseline survey that the prevalence of wheezing differed among cities, villages, and refugee camps, we randomly selected only 44 children (22 cases, 22 controls) from each location. However, we were able to collect samples in only 110 houses (63 cases and 47 controls, 21 cases from each location, and 15, 17, and 15 controls from city, village and camp, respectively). The study team were not permitted to collect samples from 3 cases' and 19 controls' houses because those children's families refused to have the team in their houses, even though at the beginning of the study they signed a consent form in which they accepted to participate in the full study stages. Furthermore, due to a technical error we lost one mattress sample of one case; analyses regarding mattress dust therefore relate to only 109 observations.

Field work procedures, house dust extraction, and allergen and endotoxin assays have been described in detail previously (13). Briefly, we collected two dust samples: one from the index child's mattress and one from the floor of the living room. Following an internationally standardized protocol (15), the whole surface (2 minutes) of the child's mattress, and 1 m^2 (2 minutes) of wall-to-wall carpet or rug, or 2 m^2 (4 minutes) from the smooth floor of the living room were vacuumed using a vacuum cleaner–equipped with a special nozzle (ALK filter and mouthpiece, ALK, Hørsholm, Denmark) to collect dust on glass fiber filters (Schleicher & Schuell, Gent, Belgium). Dust and filter were stored at -70°C and then transferred on dry ice for transport to the laboratory for analysis (Institute for Risk Assessment Sciences, Utrecht University, The Netherlands).

Endotoxin and allergens were extracted from the dust as described previously (15, 16). Briefly, the dust and the filter were extracted for 2 hours by shaking in pyrogen-free water containing 0.05% Tween-20. The volume taken for the endotoxin analysis (10% of total volume) was substituted with 10-fold (10 M) concentrated phosphate-buffered saline, and extraction was continued in PBS-Tween-20 for allergen analysis.

Dust extracts were analyzed for allergens from house dust mite (Der p1 and Der f1), cat (Fel d1), and dog (Can f1) using a monoclonal enzyme-linked immunosorbent assay (ELISA) with standards (Indoor Biotechnologies, Clwyd, UK) (17). Since Der f1 concentrations were extremely low (geometric mean <0.08 μg/g; see reference 13) compared to Der p1 values, we decided not to present results related to Der f1 concentrations. Two Der p1 samples were lower than the detection limit of 4.3 ng/mL, 6 samples of Fel d1 were lower than the detection limit of 0.6 ng/mL, and 4 Can fL measurements were less than the detection limit of 2.7 ng/mL. These levels were assigned a value of two thirds of the relevant detection limit. Endotoxin extracts were diluted 1:500 and analyzed using a chromogenic kinetic Limulus amebocyte lysate (LAL) assay (Kinetic-QCL 50-650 U; Bio Whittaker, Walkersville, MD) as described previously (15). The average detection limit, expressed in endotoxin unit per milliliter (EU/mL), of all assays was 0.041 EU/mL. Endotoxin and allergen levels were expressed per gram of sampled dust.

Ethical approval by the institutional review board was obtained before to the study (12).

Data Analyses. Student's unpaired *t* test was used to compare log-transformed endotoxin and allergen levels between cases and control subjects.

As this study is a cross-sectional case-control study we assessed the association between exposures and wheeze (in the past 12 months) by calculating prevalence odds ratios (ORs), using logistic regression analysis. For this purpose we created three exposure groups (low, medium, and high) based on tertiles of the exposure distribution of the entire data set (See Table 1 for cut points). In those analyses where we stratified for location of residence (only for analyses regarding endotoxin) we could not use tertiles because of the lower numbers (approximately 40 per subgroup); therefore, to compare "low" with "highly" exposed subjects, we used median values, i.e., 25,722 and 48,851 EU/g for mattress and living room floor concentrations, respectively.

Adjusted odds ratios (aOR) are presented with 95% confidence intervals (95% CI). Analyses were adjusted for sex, place of residence (city, village and refugee camp), and where applicable (see tables) atopy (defined on the basis of either any positive SPT or any specific IgE level >0.35 kIU/l) (12). In the multivariate analyses we also adjusted for all other measured exposures. In addition to 12 months wheeze as an outcome variable (cases), we also assessed the association between exposure and wheeze using two alternative case definitions, i.e., atopic cases ($n = 33$) and non-atopic cases ($n = 29$) (non-atopic control subjects were used as the reference group ($n = 24$).

Moreover, we did the same analysis comparing atopic ($n = 23$) with non-atopic controls. Also, we studied the association between endotoxin exposure and atopy. However, since we used a case-control design based on symptoms of wheeze, we adjusted the latter analyses for case status (i.e., wheeze in past 12 months).

Previously, we showed that the prevalence of wheezing was higher in refugee camps than in cities or villages (11). Moreover, we showed that there were differences between contaminant levels between locations, notably lower mattress endotoxin concentrations in the city compared to the two other locations (13). However, since the study sample was matched on location, we could not investigate to what extent differences in wheeze between locations were related to the contaminants studied.

All analyses were performed using SPSS version 1.0 (Chicago, IL, USA) and SAS for Windows version 8.2 (SAS Institute, Cary, NC).

TABLE 1.—Associations of wheezing with exposure to Der p1, Fel d1, and Can f1 allergens, and bacterial endotoxin.

	Mattress			Living room floor		
	Total ($N = 109$)	Case ($n = 62$)	aOR (95% CI)*	Total ($N = 110$)	Case ($n = 62$)	aOR (95% CI)*
Der p1 (μg/g)						
High (>9.37 μg/g)	37	25	3.37 (0.77–14.7)	37	17	0.82 (0.17–3.97)
Medium (3.35–9.37 μg/g)	36	16	0.87 (0.23–3.22)	37	21	0.76 (0.21–2.70)
Low (<3.35 μg/g)	36	21	1.00	36	25	1.00
Fel d1 (μg/g)						
High (>0.41 μg/g)	37	23	2.64 (0.73–9.46)	37	19	0.73 (0.20–2.55)
Medium (0.22–0.41 μg/g)	36	16	1.52 (0.45–5.08)	37	21	1.09 (0.32–3.71)
Low (<0.22 μg/g)	36	17	1.00	36	23	1.00
Can f1						
High (>0.52 μg/g)	37	19	0.27 (0.05–1.40)	37	15	0.40 (0.08–1.91)
Medium (0.25–0.52 μg/g)	36	18	0.48 (0.12–1.92)	37	23	1.08 (0.31–3.74)
Low (<0.25 μg/g)	36	25	1.00	36	25	1.00
Endotoxin (EU)						
High (>41,754 EU/g)	37	19	0.50 (0.15–1.63)	37	19	0.56 (0.15–2.00)
Medium (16,021–41,754 EU/g)	36	19	0.61 (0.19–1.94)	37	17	0.41 (0.12–1.39)
Low (<16,021 EU/g)	36	24	1.00	36	27	1.00

Exposure groups were created on the basis of tertiles of concentrations of allergens or endotoxin in dust.
*Reference category: non-wheezing children.
n: number of wheezing children exposed; N: total number of children.
aOR (95% CI): adjusted odds ratio (95% confidence interval). Analyses were adjusted for all other measured exposures, atopic sensitization (any skin prick test positivity or any specific IgE ≥0.35 KIU/L), sex, and place of residence (city, village, and refugee camp).

RESULTS

Figure 1(A–D) presents the geometric mean concentrations of Der p1, Can f1, Fel d1, and bacterial endotoxin in mattress and floor dust for all cases and controls, and with stratification by location of residence. Der p1, Can f1, and endotoxin levels on living room floors appeared somewhat higher in controls but these differences were not statistically significant.

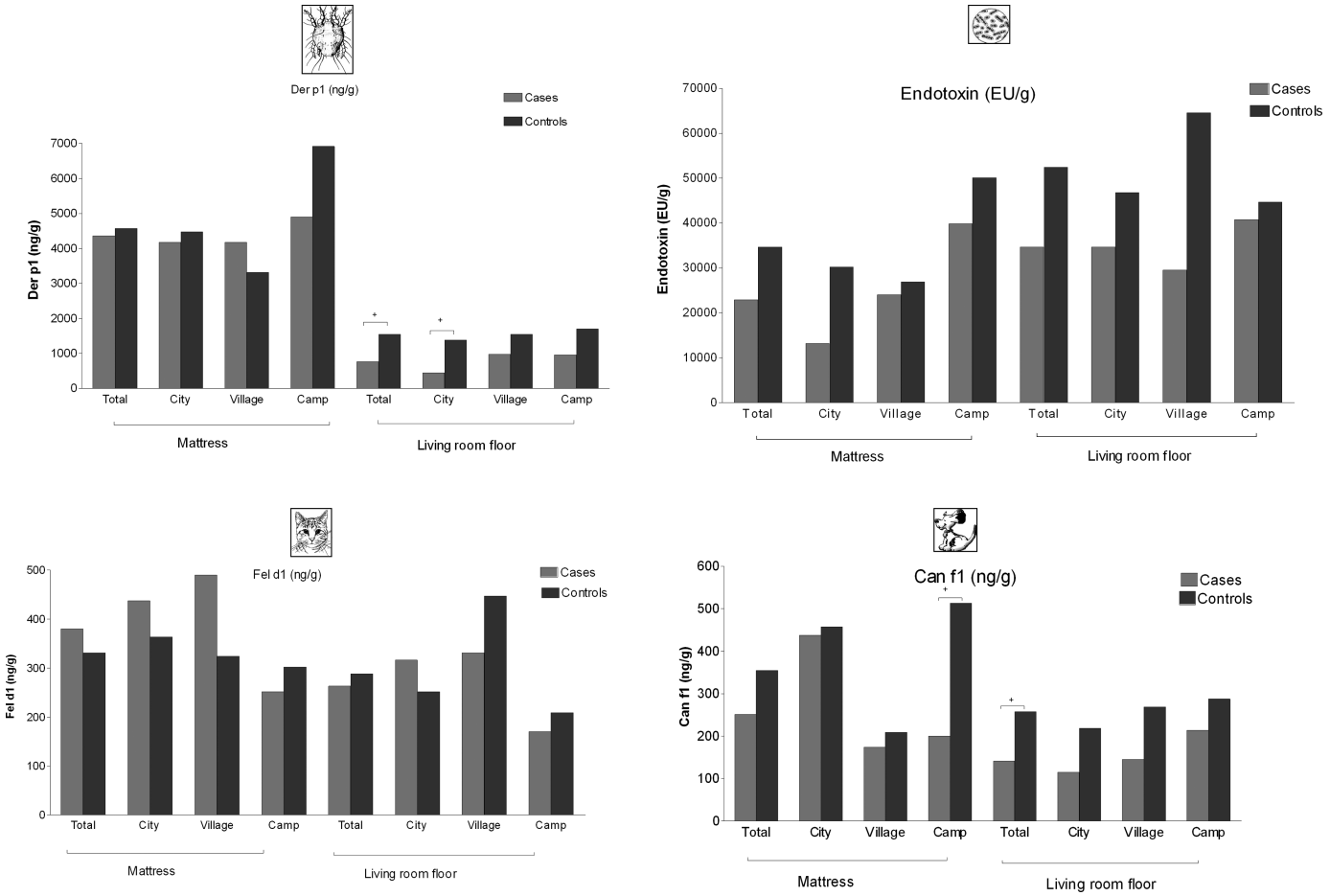

FIGURE 1.—Geometric means of allergens and endotoxin concentrations in dust collected from houses of "cases" and "controls" in cities, villages and refugee camps. Cases ($n = 63$) are children with reported wheezing in the previous 12 months; controls ($n = 47$), are children without wheezing. Number of children in cities = 36, villages = 38, and refugee camps = 36.

TABLE 2.—Associations of sensitization (any skin prick test positivity or any specific IgE ≥0.35 KIU/L) with bacterial endotoxin in dust.

	Mattress			Living room floor		
	Total ($N = 109$)	Case ($n = 56$)	aOR (95% CI)*	Total ($N = 110$)	Case ($n = 56$)	aOR (95% CI)*
Endotoxin						
High	37	18	0.90 (0.27–2.96)	37	16	0.32 (0.08–1.25)
Medium	36	20	2.05 (0.61–6.91)	37	17	0.32 (0.08–1.19)
Low	36	18	1.00	36	24	1.00

Exposure groups were created on the basis of tertiles of endotoxin concentrations in dust.
*Reference category: non-sensitized children.
n: number of sensitized children; N: total number of children.
aOR (95% CI): adjusted odds ratio (95% confidence interval). Analyses were adjusted for all other measured exposures, case status (wheeze in the past 12 months), sex, and place of residence (city, village and refugee camp). Significant odds ratios are in bold.

Der p1 and Fel d1 in mattress dust appeared associated with an increased risk for wheezing, however, these associations did not reach statistical significance (Table 1). Also, no associations with living room floor concentrations were found for these allergens. Can f1 and bacterial endotoxin in mattresses and living room floor dust showed an inverse association with wheeze in the past 12 months; However, these associations were also not statistically significant (Table 1). Of the 110 children in the study, 57 children were sensitized to at least one of the allergens tested (55% of cases and 49% of controls). Endotoxin on living room floors was inversely associated (not significant) with atopy (defined as any positive skin prick test or IgE test ≥0.35 KIU/L), after adjusting for other exposures, sex, location of residence, and case-control status (Table 2). No clear association with mattress dust levels was found. We also analyzed the association between specific mite sensitization and mite allergen concentration in floor and mattress dust using median values. However, no significant associations were found (data not shown). We did not carry out the analysis for pet allergens because the numbers of children sensitized to cat or dog allergens were too small ($n < 15$).

Endotoxin concentrations were consistently higher among non-atopic children regardless of their wheezing condition, and this was statistically significant for endotoxin concentrations in mattresses (data not shown). In multivariate analyses we found a strong inverse association between endotoxin and wheeze when non-atopic controls served as the reference group (Table 3). The strongest associations were found with floor endotoxin concentrations comparing atopic cases with non-atopic control subjects. The effect of endotoxin appeared less pronounced when comparing non-atopic cases with non-atopic controls, suggesting that endotoxin may protect more against atopy than against wheeze. Endotoxin also appeared inversely associated with atopy within the control group (Table 3). The effects were most consistent for living room floor levels.

Table 4 shows the crude associations between endotoxin and wheeze/atopy by location using only two exposure groups. The differences in odds ratios between locations were modest for floor dust, indicating that endotoxin in floor dust is consistently associated with a lower prevalence of wheeze in the past 12 months and a lower prevalence of atopy (statistically significant only for wheezing in the village population and the total pooled population; Table 4). The associations with mattress concentrations were less consistent (Table 4). This is in agreement with the results presented in Tables 1 to 3 in which the same associations were assessed for the total group of children using three instead of two exposure groups.

DISCUSSION

In this case-control study we found no consistent associations between allergen levels and either wheeze or specific atopic sensitization. Also, no clear associations between mattress endotoxin levels and wheeze or atopy were found. However, we did find an inverse association between bacterial endotoxin in living room floor dust and wheeze, statistically significant only for atopic wheeze. Also, among non-wheezing controls a negative association between living

TABLE 3.—Associations between endotoxin and wheezing using two case definitions (sensitized vs non-sensitized) or sensitization in the non-wheezing controls.

	Controls-non-sensitized* $N = 24$		Cases- sensitized $N = 33$		Cases- non sensitized $N = 29$		Controls-sensitized $N = 23$	
	N	n	n	aOR (95% CI)	n	aOR (95% CI)	n	aOR (95% CI)
Mattress								
High	37	9	9	0.69 (0.10–4.83)	10	0.25 (0.05–1.26)	9	0.60 (0.10–3.34)
Medium	36	11	14	1.53 (0.21–10.8)	5	**0.13 (0.02–0.71)**	6	0.23 (0.03–1.44)
Low	36	4	10	1.00	14	1.00	8	1.00
Living floor								
High	37	10	8	**0.02 (0.002–0.26)**	11	0.23 (0.03–1.67)	8	0.17 (0.02–1.57)
Medium	37	12	9	**0.04 (0.005–0.35)**	8	0.17 (0.01–1.02)	8	0.28 (0.03–2.48)
Low	36	2	17	1.00	10	1.00	7	1.00

Exposure groups were created on the basis of tertiles of endotoxin concentrations in dust.
*Reference category: non-sensitized control children; n: number children in each exposure category according to the case definition; N: total number of children in each exposure category.
aOR (95% CI): Adjusted odds ratio (95% confidence interval). Analyses were adjusted for sex and place of residence (city, village and refugee camp). Significant odds ratios are in bold.

TABLE 4.—Unadjusted associations between endotoxin and wheeze and sensitization by location of residence.

	City $n = 36$ OR (95% CI)	Village $n = 38$ OR (95% CI)	Camps $n = 36$ OR (95% CI)	Combined $n = 110$ OR (95% CI)
Mattress				
12 months wheezing	0.27 (0.06–1.08)	0.84 (0.23–3.05)	0.75 (0.18–3.03)	0.57 (0.26–1.22)
sensitization	0.85 (0.18–3.93)	0.77 (0.21–2.80)	1.28 (0.29–5.55)	0.77 (0.36–1.64)
Sensitization in cases	1.81 (0.16–20.7)	1.12 (0.19–6.41)	1.19 (0.19–7.45)	0.90 (0.33–2.48)
Sensitization in controls	0.40 (0.03–5.15)	0.48 (0.06–3.35)	1.71 (0.13–22.5)	0.65 (0.20–2.09)
Living room floor				
12 months wheezing	0.50 (0.13–1.92)	**0.16 (0.04–0.68)**	0.37 (0.09–1.48)	**0.31 (0.14– 0.69)**
sensitization	0.19 (0.03–1.12)	0.63 (0.17–2.31)	0.65 (0.16–2.58)	0.51 (0.24–1.10)
Sensitization in cases	0.40 (0.05–3.12)	0.57 (0.07–4.12)	0.20 (0.02–1.42)	0.44 (0.15–1.25)
Sensitization in controls	†	0.47 (0.05–3.99)	†	0.64 (0.19–2.15)

Exposure groups were created on the basis of median values of endotoxin concentrations in dust.
n: total number of children in each location; Cases: children with wheezing in the previous 12 months; Controls: Children without wheezing in the previous 12 months.
Significant odds ratios are in bold.
†No odds ratio could be calculated since all sensitized controls living in cities and camps were exposed to endotoxin levels greater than the median.

room levels of endotoxin and atopy was observed (not statistically significant).

Potential Limitations

One important aspect of the study is that cases and controls were matched for study location. As a result, differences in wheeze between study locations could not be investigated in relation to biocontaminant exposure. This is important because wheeze and endotoxin (particularly in mattress dust) were lowest in the city and highest in the camps, suggesting a positive relation on the "ecologic" scale, whereas the individual analyses presented here suggest an inverse association (for endotoxin in floor dust). Since our previously reported larger case-control study (12) was also matched on location, we were unable to directly investigate the extent to which differences in wheeze between locations were related to the various risk and/or protective factors that were investigated in this and the previous case-control study. Furthermore, the limited number of cases and controls for whom we collected the environmental samples affected the statistical power of the results in this study.

Also, equal numbers of cases were enrolled from each of the three locations of residence. In the original study (11), there were three times more cases in the camps, and twice as many cases in the villages than in the city, so that in the current analyses, the subjects from the city carry most weight. The pooled analysis presented here effectively gives equal weight to the three locations and can thus not be directly extrapolated to the source population in which there were different numbers in the three locations. However, this will only be an issue if the odds ratios differ between the three locations. As shown in Table 4 the ORs for endotoxin and wheeze and atopy calculated separately for each location were only moderately different, suggesting that this is only a minor issue.

The pooled analysis focusing on atopic sensitization needed to take the case-control design of the study into account, assuming homogeneity of associations observed in cases and controls. Results presented in Tables 3 and 4 show that the direction of the effect of endotoxin on atopy is the same for cases and controls; therefore, despite the difference in strength and magnitude of the associations it appears that the assumption of homogeneity is not seriously violated.

Finally, we may not have measured the most important indoor allergens or pollutants. For example, we did not measure cockroach allergens in dust, whereas cockroach sensitization was one of the significant determinants of wheezing in our first case-control analysis (12). Similarly, we did not measure $(1 \rightarrow 3)$-ß-*D*-glucans or other markers of fungal contamination, and yet dampness was a significant determinant of wheezing in our previous analysis. Also, changes associated with westernization may play a role, involving an increase in asthma susceptibility rather than an increase in exposure to "established" asthma risk factors such as indoor allergens (18). Thus the differences in asthma prevalence among cities, villages, and refugee camps may not only be due to differences in indoor allergen and endotoxin levels, but other factors, whether environmental or related to lifestyle, may also be important.

Indoor Endotoxin and Allergen Exposure and Wheezing and Sensitization

Our results are in concordance with previous studies that have suggested a protective effect of endotoxin on atopy and asthma in both animals (19, 20) and humans (9, 10, 21–23). Bacterial endotoxin has strong immunomodulatory capacities and has been suggested to inhibit the atopic Th_2 response through IL-12 and IFN-γ production in antigen-presenting cells (24, 25). In our study, endotoxin was inversely associated with atopy, but this did not reach statistically significance (Table 2). We also found an inverse association ($p < 0.05$) between endotoxin and atopic wheeze (Table 3), thus suggesting that bacterial endotoxin may protect against atopic wheeze, possibly by inhibiting the atopic immune response. In addition, we showed that this protective effect was not only observed in villages where 45% of the houses had animals (a well known source of endotoxin) within three meters of the house (13), but also in a non-rural environment. However, our finding that endotoxin in floor dust was inversely associated with atopic wheeze should, be interpreted with caution since it was based on a relatively small sample size and could therefore be due to a chance finding. Also, there is evidence that inhaled endotoxin can induce non-atopic inflammatory responses, airflow obstruction, and non-allergic symptoms in both normal and asthmatic subjects (6, 26). Furthermore, a recent birth cohort study indicated that early exposure to high endotoxin levels in mattresses might increase the risk for atopic reactions to inhalant allergens at the age of 2 years (27). The evidence that endotoxin may protect against atopy and asthma is thus mixed.

In our baseline prevalence study, children in refugee camps had the highest prevalence of wheeze compared to children in cities or villages (11). However, as mentioned earlier, we found the highest endotoxin concentration in mattresses of children living in refugee camps. Also, floor concentrations (expressed per g dust) were very similar among the three locations (with slightly higher levels in villages and refugee camps). Therefore, indoor endotoxin levels may not play a major role in explaining the differences in asthma prevalence between these locations (13). We have no explanation for this (nonsignificant) finding, other than that other environmental factors present in the refugee camps may be involved. It is also not clear why results were more consistent for living room floor dust than for mattress dust.

Several studies have shown dose-response relationships between exposure to mite and pet allergens and sensitization (28, 29). Many studies, however, failed to show a consistent or direct dose-response relationship between allergen exposure and asthma development (5). Moreover, studies in countries with high exposure to dust mites showed similar asthma prevalence rates to those with relatively low levels of mite allergen (5). In our study, Der p1 and Fel d1 levels in mattresses appeared positively associated with wheeze (not significant); however, no associations were found with living room floor concentrations. A previous study in refugee camps in the coastal location of the Gaza Strip in Palestine showed that houses of asthmatic children were highly contaminated with mites, which was strongly associated with diagnosed asthma (30). This confirms (to a certain extent) our findings for Der p1 levels in mattresses. Surprisingly, we could not demonstrate a consistent relationship between allergen levels and specific sensitization. However, this could be due to the relatively small population size that does not allow such detailed analyses. The reason why we found an inverse association between Can f1 and wheeze (not statistically significant) is not clear and may be a chance finding.

We have expressed endotoxin and allergen concentrations per gram of dust, as is commonly done in indoor studies. However, it has been argued that concentrations expressed per meter square may better reflect the actual exposure (31). In our study, we found comparable results when concentrations were expressed per meter square (data not shown).

Conclusions

Our study is one of the few where the concentrations of four different domestic aeroallergens and endotoxin have been measured in a relatively large number of houses. Even in case-control studies from more affluent countries, such as Sweden, the numbers of investigated homes rarely exceeded 200 (23). Moreover, the survey and data collection were done in difficult circumstances due to the war situation in the occupied territories. The environment of Palestine enabled us not only to compare a rural and urban environment, which has not often been investigated in other studies, but also to compare these two environments with a unique other environment, i.e., refugee camps. Our findings related to endotoxin exposure and its potential protective effect on atopy and atopic disease extend the validity of some conclusions derived from studies in Western countries to a non-Western location and a type of population where the epidemiology of childhood asthma has only rarely been studied.

In conclusion, this case-control study in children in Palestine demonstrated that dust mite and pet allergens in house dust were not consistently associated with wheeze and specific sensitization. However, there was, some indication that endotoxin on living room floors might protect against atopic wheeze. This finding should, however, be interpreted with caution due the relatively small sample size of the study and requires further confirmation.

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