

Early-Life Allergen Exposure and Atopy, Asthma, and Wheeze up to 6 Years of Age

Matias Torrent¹, Jordi Sunyer^{2,3}, Raquel Garcia², Jessica Harris⁴, Maria V. Iturriaga¹, Carme Puig⁵, Oriol Vall⁵, Josep M. Anto^{2,3}, Anthony J. Newman Taylor⁴, and Paul Cullinan⁴

¹Menorca Health Area, ib-salut, and Institut Universitari d'Investigació en Ciències de la Salut, Menorca, Spain; ²Centre for Research in Environmental Epidemiology, Institut Municipal Investigació Mèdica, Barcelona, Spain; ³Universitat Pompeu Fabra, Barcelona, Spain; ⁴Department of Occupational and Environmental Medicine, Imperial College, London, United Kingdom; and ⁵Environment and Pediatric Research Unit, Pediatric Service, Hospital del Mar, and Pediatric Department, Barcelona Autònoma University, Barcelona, Spain

Rationale: Although it is widely assumed that the incidence of childhood respiratory allergies to common aeroallergens is directly related to allergen exposure in early life, few longitudinal studies have investigated this issue, and available data are scarce and mainly limited to high-risk groups.

Objectives: To assess, in a prospective manner and in a general population, the role of early life exposures to Der p1 and Fel d1 on the inception of sensitization and asthma.

Methods: Pregnant women and their children were recruited for the Asthma Multicentre Infant Cohort Study. Overall, 1,611 newborns were initially enrolled in three cohorts in the United Kingdom and Spain. Der p1 and Fel d1 allergens were measured in household dust samples at 3 months of age for 1,474 (91.5%) participants, and skin prick tests were performed at 6 years of age on 1,182 (80.2%) participants. Wheeze and diagnosed asthma were reported in yearly questionnaires.

Measurements and Main Results: Exposure to Der p1 early in life was not related to a positive specific prick test or to asthma or persistent wheeze at 6 years of age. Fel d1 showed an association with all these outcomes (third vs. first tertile; odds ratio, 4.43 for positive specific prick test and 2.6 for diagnosed asthma).

Conclusions: Dose-response relationships between allergen exposure and sensitization or asthma may be allergen specific and nonlinear; a minimum threshold level is needed to induce sensitization, but no dose-response relationship exists above this level. The effect of a particular allergen seems to be similar on atopy and asthma inception.

Keywords: wheeze; sensitization; skin prick test; Der p1; Fel d1

Specific sensitization and asthma have traditionally been linked to allergen exposure through inhalation (1, 2). In more recent years, several studies have challenged the assumption that the initiation of asthma is related to allergen exposure (3–5), whereas the available epidemiologic evidence is consistent with a role of allergen exposures in the development of specific sensitization (6–8). Because there is relatively well established evidence that sensitization to common aeroallergens is linked with an increased risk of wheeze and asthma (9), it is a relevant question whether allergen exposure is or is not a risk factor for asthma (10). Several reasons could explain the incongruities found in this respect, including the following: the influence of

AT A GLANCE COMMENTARY

Scientific Knowledge on the Subject

Few longitudinal studies have investigated the role of allergen exposure early in life on the inception of atopy and asthma because data are scarce and mainly limited to high-risk groups.

What This Study Adds to the Field

In prospectively studied representative community samples, we found no effect of house dust mite exposure on specific sensitization or asthma and a nonlinear risk curve for Fel d1 exposure. Effects of allergen exposure approach an “all or nothing” event.

factors that could inhibit atopic sensitization, such as contact with pets, farm environments, or endotoxins (11, 12); geographic differences in relation to different levels of exposure (13); or the influence of susceptibility factors (14, 15) in studies in which the population has been selected from high-risk families (16). Some of these explanations can only be simultaneously tested in prospective studies conducted in different areas with enough variation to exposure, including several allergens, and in nonselected representative populations (4, 17).

Our objective when setting up the Asthma Multicentre Infant Cohort Study was to assess, in a prospective manner and in a general population, the role of early life exposures to two major household aeroallergens (Der p1 and Fel d1) upon the subsequent development of sensitization and asthma. Our findings from three independent European populations, one in the United Kingdom and two in Spain, with different aeroallergen exposure profiles (18, 19), are presented. In subjects who were 4 years of age, we found an effect on sensitization (20) and wheeze (21) to cat allergen but not to house dust mite, although both phenotypes are not considered to be fully expressed at this age (22). Now we have the opportunity to assess the prospective relationship between exposure to aeroallergens in early life and the development of specific sensitization, wheeze, and asthma up to 6 years of age in nonselected populations.

METHODS

Pregnant women and their children were recruited in three concurrent cohorts (Ashford, UK; Menorca Island and Barcelona City, Spain) following the same research protocol. Cohorts were population based in Ashford and Menorca and hospital based in Barcelona. In all centers, women agreed to participate after they were told the objectives of the study; each provided written consent. The study was approved by the ethics committee of the participating centers.

Information collected prospectively included details of the pregnancy, cord blood, a household visit when the infant was 3 months of

(Received in original form July 6, 2006; accepted in final form June 11, 2007)

Supported by Spanish Ministry of Health (Barcelona: grants FIS 95/0314 and FIS 00/0021–01; Menorca: grants FIS 97/0588, FIS 00/0021–02 and FIS G03/176), The COLT Foundation (Ashford), and by the Fifth Framework Program of the European Community (QLK4-CT-2000–00263).

Correspondence and requests for reprints should be addressed to Dr. Matias Torrent, ib-salut Menorca, C/ Barcelona 3, 07703-Mao (Menorca), Spain. E-mail: mtorrent@smen.es

Am J Respir Crit Care Med Vol 176. pp 446–453, 2007

Originally Published in Press as DOI: 10.1164/rccm.200607-916OC on June 15, 2007

Internet address: www.atsjournals.org

age for collection of dust and ambient air NO₂ samples, blood sampling and skin prick testing of the mothers (and of a smaller proportion of fathers), a yearly questionnaire up to the age of 6 years enquiring into respiratory symptoms and diagnosis, household characteristics and exposures (pets, passive smoking, cooking and heating appliances), venipuncture of the children at the 4 years of age, and a skin prick test including *Dermatophagoides pteronyssinus* and cat epithelia allergens at 6 years of age. All data collection methods, household sampling, and prick test technique were standardized between centers (21). A common training was followed by the fieldworkers of the three centers, and the same dust collection appliances and the same prick test technique, including the same equipotent extracts (prepared by Leti Laboratories for the Spanish centers and by ALK-Abello for the U.K. center) and the same 1-mm prick test lancets, was used.

Dust samples were collected from living room and from children's mattresses during the first 3 months of life. Results obtained using living room or bed levels were similar. In this article, living room levels were used for the main analysis because, apart from showing lower levels, bed levels were more affected by nondifferential misclassification (risk estimates tended more to null; data not shown). House dust mite allergen (Der p1) and cat allergen (Fel d1) concentrations were estimated following a standard protocol (18, 19).

Out of 1,611 newborn children initially enrolled, household dust samples were collected for 1,474 (91.5%), and 1,182 (80.2%) children completed skin prick tests at 6 years of age. Dust samples and prick tests were available for about 87% of children in Ashford and Menorca and 59% of children in Barcelona. For information on asthma at 6 years of age, these percentages were 95 and 72%, respectively. Due to differences in loss of follow-up in the three centers, we repeated all analyses after the exclusion of children from Barcelona and tested for heterogeneity between centers. House dust mite and cat allergen concentrations were log transformed because of their skewed distribution. Five outcome variables were defined: a positive (wheal ≥ 3 mm) prick test to Der p1 and to Fel d1 at 5 to 6 years of age, a medical diagnosis of asthma up to 6 years of age as reported by the parents, persistent wheeze (any wheeze between 5 and 6 years of age or having had wheezing between 3 and 4 years of age), and current wheeze at 6 years of age (any wheeze during the sixth year of life). To examine the independent effects of allergen concentrations on the risk of the different outcomes defined, we first used generalized additive models (GAMs), which allow the fitting of a nonlinear exposure–response relationship and give a graphic representation of any association. Second, we fitted multiple regression models for binary data for each of the dependent variables defined. Allergen concentrations were introduced into the models as tertiles of the pooled levels of the three centers. Interactions with allergen exposure, including factors such as gender and parental atopy or asthma, were tested using interaction terms in the regression model; only maternal atopy showed significant interaction, and stratified models by maternal atopic status were constructed. Stratified analysis was also performed for atopic and nonatopic asthma and wheeze. Statistical analyses were conducted using Stata software (version 8.2; Stata Corporation, College Station, TX).

RESULTS

The concentrations of Der p1 and Fel d1 antigens in living room dust are shown in Table 1 separately by center, along with the proportions of children with positive prick tests to these aeroallergens, a diagnosis of asthma by 6 years of age or having wheezed in their fifth or sixth year of life, and those considered to be persistent wheezers. Exposure profiles were very different between the centers, with high levels of Der p1 in Menorca and high levels of Fel d1 in Ashford.

Nonadjusted comparisons of sensitization, diagnosed asthma, persistent wheeze, or wheeze at 6 years of age by tertile of dust allergen exposure (Table 2) showed no associations for house dust mite exposure with any one of the outcomes. For cat allergen exposure, a statistically significant association was observed with wheeze at 6 years of age, and the associations with diagnosed asthma, persistent wheeze, and positive specific prick test to Fel

d1 were at the limit of statistical significance. None of these relationships was modified by center as shown by the heterogeneity tests; nor were there any important changes after exclusion of children from Barcelona. Table 2 also shows the positive associations between sensitization and asthma or wheeze. All these associations were highly significant; the sense of the association was the same for the three centers, although the association between positive specific prick test to Der p1 and wheezing was lower in Barcelona (*P* for heterogeneity between centers < 0.05).

All variables that showed an association with any of the outcomes on bivariate analysis are shown in Table 3. Male gender, high home crowding, maternal or paternal atopy, and maternal or paternal asthma significantly increased the risk of specific sensitization to at least one of the two aeroallergens studied. Male gender, a lower gestational age at birth, lower NO₂ levels, smoking during pregnancy, maternal atopy, maternal and paternal asthma, lower respiratory tract infection during the first year of life, and a shorter breastfeeding period were risk factors for diagnosed asthma or wheeze. Other variables analyzed that did not reach statistical significance for any of the outcomes included birth order, birth weight, cat or dog ownership (or number of them) during the first year of life, household dampness, season in which the dust was collected, and maternal education.

Figure 1 depicts, on a continuous scale, the relationship of Der p1 and Fel d1 exposures with each one of the outcomes studied based on a GAM constructed for each exposure–outcome pair. There was no association between house dust mite exposure and specific sensitization, asthma, or wheeze. In contrast, Fel d1 exposure showed a positive association for all outcomes; in each case the risk seemed to increase at low levels of exposures and to flatten above 1 $\mu\text{g/g}$ of dust. Wide confidence intervals (CIs) at the extremes highlight the low precision of the risk estimates of the GAM at this point.

Multiple regression models for Der p1 exposure (Table 4) confirmed that the level of exposure early in life was not related to subsequent specific sensitization. Male sex, with an odds ratio estimate of 1.85 (95% CI, 1.24–2.76) and an atopic mother (odds ratio, 2.07; 95% CI, 1.38–3.10) were the main risk factors for this outcome. Of the three sites studied, Ashford had the lowest risk of a positive prick test to Der p1, and Barcelona had the highest risk. Fel d1 exposure showed a strong association with the sensitization (Table 4), although no difference in risk was observed between the second and third tertiles of exposure. Other risk factors associated with a positive prick test to cat were high home crowding, maternal atopy, and maternal asthma. By center, the major risk of sensitization to cat was in Barcelona, and the lowest was in Menorca.

Diagnoses of asthma and persistent wheeze were positively related to Fel d1 exposure, although only asthma reached statistical significance. There were no relationships between Der p1 exposure and any of the respiratory outcomes (Table 4). The observed risk factors included in the multivariate models because of a *P* value lower than 0.10 for a diagnosis of asthma and wheeze, with very similar estimates for the Fel d1 and the Der p1 models, were having a positive specific prick test, maternal and paternal asthma, smoking during pregnancy, a lower respiratory tract infection during the first year of life, and being exposed to higher levels of NO₂ early in life, whereas being breastfed for more than 2 weeks seemed to have a protective effect. Children in Ashford had the highest risk of an asthma diagnosis and of wheeze even after adjusting for the relevant variables, but no interaction with center was observed. Similar results were obtained when taking wheeze at 6 years instead of persistent wheeze as the dependent variable.

TABLE 1. DOMESTIC ALLERGEN CONCENTRATIONS IN EARLY LIFE AND SPECIFIC PRICK TEST POSITIVITY, ASTHMA DIAGNOSIS, PERSISTENT WHEEZE, AND WHEEZE AT AGE 6 BY CENTER

	Ashford			Barcelona			Menorca		
	Total n	Geometric Mean (95% CI)		Total n	Geometric Mean (95% CI)		Total n	Geometric Mean (95% CI)	
At 3 mo of age									
House dust mite allergen concentrations, $\mu\text{g/g}$	624	1.36 (1.17–1.59)		369	0.77 (0.65–0.92)		481	9.04 (7.93–10.31)	
Cat allergen concentrations, $\mu\text{g/g}$	623	3.12 (2.55–3.81)		368	0.38 (0.31–0.45)		481	0.43 (0.36–0.51)	
At 1 yr of age									
Cat ownership	627	208	33.17	393	34	8.65	478	8	1.67
At 6 yr of age									
Specific prick to									
House dust mite*	552	39	7.07	256	63	24.61	419	52	12.41
Cat*	551	35	6.35	256	13	5.08	419	3	0.72
Asthma diagnosis	604	118	19.54	312	29	9.29	459	13	2.83
Persistent wheeze†	602	87	14.45	214	23	10.75	478	31	6.49
Wheeze at 6 yr	604	108	17.88	311	43	13.83	481	41	8.52

Definition of abbreviation: CI = confidence interval.

* Positive specific prick test: ≥ 3 mm.

† Defined as wheeze at 5–6 years and at 3 or 4 years.

We looked extensively for possible interactions with exposure to the two aeroallergens of interest, with only maternal atopy suggestive of any such effect. Table 5 shows the models stratified by maternal atopy and the *P* values for interactions for each allergen and outcome model. In children of atopic mothers, exposure to Der p1 seemed to protect against a positive prick

test to house dust mite. For asthma diagnosis, a higher risk was associated with aeroallergen exposure if the mother was atopic; this was especially the case for Fel d1 exposure.

We analyzed different possible associations with aeroallergen exposure depending on whether the child's asthma or wheeze was accompanied by specific sensitization. Similar results were obtained for sensitized and nonsensitized children with no evidence of any interaction by sensitization status.

TABLE 2. PREVALENCE OF POSITIVE SPECIFIC PRICK TEST, DIAGNOSED ASTHMA, PERSISTENT WHEEZE, AND WHEEZE AT AGE 6 BY CATEGORY OF AEROALLERGEN EXPOSURE AND PREVALENCE OF RESPIRATORY OUTPUTS BY SPECIFIC PRICK TEST RESULT

	Positive Specific Prick Test*		Diagnosed Asthma		Persistent Wheeze		Wheeze at Age 6	
	n	%	n	%	n	%	n	%
Allergen in dust								
Der p1 concentration								
<0.83 $\mu\text{g/g}$	48	13.4	66	16.1	55	15.2	74	18.0
0.83–6.46 $\mu\text{g/g}$	53	12.6	56	12.5	44	10.5	59	12.9
≥ 6.46 $\mu\text{g/g}$	52	10.6	30	6.5	37	8.3	49	10.4
<i>P</i> value†		0.617		0.219		0.240		0.193
<i>P</i> heterogeneity‡		0.623		0.854		0.279		0.524
Fel d1 concentration								
<0.25 $\mu\text{g/g}$	5	1.3	27	6.3	34	8.5	48	11.0
0.25–1.39 $\mu\text{g/g}$	18	4.8	44	10.3	35	8.9	49	11.3
≥ 1.39 $\mu\text{g/g}$	27	6.3	82	17.9	67	15.5	86	18.6
<i>P</i> value†		0.060		0.069		0.051		0.036
<i>P</i> heterogeneity‡		0.477		0.587		0.212		0.247
Specific skin prick test								
Der p1								
<3 mm			114	10.7	87	8.8	118	11.0
≥ 3 mm			35	22.7	40	32.8	57	37.0
<i>P</i> value†				<0.001		<0.001		<0.001
<i>P</i> heterogeneity‡				0.259		0.031		0.032
Fel d1								
<3 mm			127	10.9	109	10.2	150	12.8
≥ 3 mm			21	41.2	17	41.5	24	47.1
<i>P</i> value†				<0.001		<0.001		<0.001
<i>P</i> heterogeneity‡				0.479		0.252		0.258

* Positive (≥ 3 mm) specific prick test to Der p1 or Fel d1 according to the specific allergen in dust.

† Adjusted for site.

‡ Minimum of *P* values for heterogeneity between centers.

DISCUSSION

The Asthma Multicentre Infant Cohort Study is the first international birth cohort study designed to investigate the relationship between exposures early in life and childhood sensitization and asthma in a general nonselected population. The main exposures of interest were household aeroallergens, namely house dust mite and cat, of which a wide range of exposures were apparent in the three settings. Our results strongly suggest that the relationship between exposure to inhaled allergens in early life and the development of sensitization and asthma is allergen dependent, with Der p1 and Fel d1 showing different dose–response patterns. In addition, within the range of exposures observed, for the same allergen, the role of aeroallergen exposure is similar on sensitization and on asthma.

With respect to cat allergen exposures, we found a nonlinear relationship between home exposure to Fel d1 in early life and corresponding sensitization by skin prick test and a diagnosis of asthma or wheeze up to 6 years of age. The relationships suggest a steep increase in risk at low levels of exposure and a flattening above 1 $\mu\text{g/g}$ of dust. This result is in agreement with previously published literature, although we did not observe a reduction of risk, as others have, at high exposure levels (9). In our population, we found no effect of cat ownership or of the number of cats owned on the risk of sensitization or on asthma. Such effects have been reported by some authors (12) but not by others (23) or have been reported to be limited to sensitization but not to asthma (5). These inconsistencies may reflect particular behaviors in relation to cat keeping.

We did not find any significant associations between early life domestic Der p1 allergen exposure and any one of the outcomes studied. The lack of any apparent association of house dust mite with sensitization or with asthma up to 6 years of age confirms our preliminary findings concerning wheeze up to 4 years of age (21)

TABLE 3. UNADJUSTED ASSOCIATIONS BETWEEN SPECIFIC PRICK TEST, DIAGNOSED ASTHMA, PERSISTENT WHEEZE, AND WHEEZE AT AGE 6 WITH VARIABLES OF INTEREST

Variables*	Total n	Positive Specific Prick Test [†] to Der p1, n (%)	Positive Specific Prick Test [†] to Fel d1, n (%)	Diagnosed Asthma, n (%)	Persistent Wheeze, n (%)	Wheeze at Age 6, n (%)
Sex						
Female	762	54 (9.3)	19 (3.3)	66 (10.3)	50 (8.5)	73 (11.2)
Male	847	100 (15.5) [‡]	32 (5.0)	94 (12.8)	91 (13.6) [‡]	119 (16.0) [‡]
Birth order						
No firstborn	854	72 (10.8)	25 (3.7)	90 (12.0)	81 (11.7)	104 (13.8)
Firstborn	741	78 (14.2)	26 (4.7)	69 (11.1)	59 (10.4)	86 (13.6)
Gestational age						
≥ 37 wk	1,495	140 (12.3)	46 (4.0)	144 (11.3)	126 (10.7)	175 (13.5)
< 37 wk	103	13 (15.9)	5 (6.1)	15 (16.9)	15 (19.0) [‡]	17 (19.1)
Home crowding						
“Low” (≤2 persons per room)	1,363	136 (12.6)	37 (3.4)	135 (11.2)	123 (11.0)	162 (13.2)
“High” (>2)	127	8 (7.5)	13 (12.3) [‡]	20 (16.4)	15 (13.0)	22 (18.0)
NO ₂						
<10 µg/m ³	702	52 (8.4)	29 (4.7)	91 (13.6)	81 (12.3)	102 (15.0)
≥10 µg/m ³	730	80 (15.0)	21 (3.9)	61 (10.0) [‡]	53 (9.9)	78 (12.5)
Maternal social class						
I–II, Professional/technician	316	35 (13.9)	19 (7.6)	39 (13.7)	33 (12.6)	46 (15.9)
III, Skilled nonmanual	253	15 (7.7) [‡]	8 (4.1)	14 (6.1)	9 (4.2) [‡]	11 (4.8) [‡]
III, Skilled manual	609	64 (13.6)	11 (2.3) [‡]	48 (9.3)	44 (9.2)	71 (13.4)
IV, Unskilled	259	26 (12.3)	9 (4.3)	30 (13.2)	33 (15.6)	38 (16.7)
Maternal smoking at pregnancy						
Never	1,157	114 (12.6)	39 (4.3)	107 (10.6)	94 (10.0)	132 (12.9)
Ever	382	34 (12.1)	12 (4.3)	47 (14.6) [‡]	44 (15.3) [‡]	56 (17.3) [‡]
Maternal atopy [†]						
No	1,094	85 (10.4)	26 (3.2)	101 (10.9)	81 (9.7)	118 (12.6)
Yes	478	62 (16.3) [‡]	24 (6.3) [‡]	56 (13.5)	57 (14.4) [‡]	71 (16.6) [‡]
Paternal atopy [†]						
No	576	43 (9.1)	17 (3.6)	63 (12.3)	61 (13.0)	82 (16.0)
Yes	390	48 (14.9) [‡]	22 (6.8) [‡]	60 (16.7)	40 (11.8)	53 (14.5)
Maternal asthma						
No	1,446	133 (12.1)	39 (3.5)	124 (10.1)	108 (9.6)	155 (12.4)
Yes	163	21 (16.9) [‡]	12 (9.7) [‡]	36 (25.2) [‡]	33 (24.4) [‡]	37 (25.5) [‡]
Paternal asthma						
No	1442	132 (12.0)	43 (3.9)	128 (10.4)	114 (10.1)	163 (13.0)
Yes	143	19 (16.5) [‡]	8 (7.0)	32 (25.2) [‡]	26 (21.8) [‡]	28 (21.7) [‡]
LRTI at year 1						
No	890	97 (13.9)	38 (5.4)	73 (9.3)	65 (9.0)	89 (11.3)
Yes	540	43 (9.7)	13 (2.9)	82 (16.5) [‡]	67 (14.5) [‡]	91 (18.2) [‡]
Birth weight						
≤2,500 g	118	13 (15.7)	5 (6.0)	13 (13.3)	15 (17.4)	17 (17.0)
>2,500 g	1,493	141 (12.3)	46 (4.0)	147 (11.5)	126 (10.7)	175 (13.5)
Breastfeeding						
<2 wk	479	40 (10.7)	20 (5.3)	76 (18.1)	59 (14.8)	71 (16.6)
2–12 wk	417	43 (13.4)	11 (3.4)	39 (10.9)	31 (9.7)	48 (13.3)
>12 wk	563	61 (13.3)	19 (4.2)	37 (7.3)*	42 (8.9)	61 (11.8)

Definition of abbreviation: LRTI = lower respiratory tract infection.

* Variables included are those with $P < 0.10$ for any one of the outcomes.

[†] Positive specific prick test ≥3 mm.

[‡] $P < 0.05$.

and IgE sensitization at 4 years of age (20) and is in agreement with the results observed in another highly exposed population in Michigan (24). These findings are in apparent contradiction to other published literature from observational studies that suggest that sensitization to house dust mite is directly related to levels of early allergen exposure. These surveys, however, are generally derived from small studies of selected populations from atopic or asthmatic families (16), from surveys based on asthmatic children alone (25), or from studies in settings where exposures to house dust mite were extremely low (3, 26). In this sense, our finding of an interaction effect of maternal atopy on the relationship between allergen exposure and sensitization or asthma is especially relevant, indicating that results from studies in descendants of high-risk families may not apply to the general population. Other studies have also pointed out the importance of parental history as a modifying independent variable in the relationship between early dust mite exposure and atopic outcomes (24),

although the effect modification has not always been shown to work in the same direction.

Our findings suggest that, for a given allergen, the effects on the inception of specific sensitization and of asthma are similar, a consistency that remained after stratification by maternal atopy. This consistency is in contrast with previous studies (3–5) and resolves an incongruity in the understanding of the exposure–atopy–asthma relationship. The difference between our results and those from previous studies may be partially due to the fact that we have covered a wide range of exposure levels, but only a few subjects were exposed to very low levels of allergens, especially for Der p1, and the different effect on sensitization and on asthma may only occur at this very low level. In fact, in the Ashford cohort, the only one of our three cohorts with a sufficient number of children exposed to low levels of house dust mite allergen, a weak, nonsignificant risk at very low levels of exposure with attenuation thereafter was found (27).

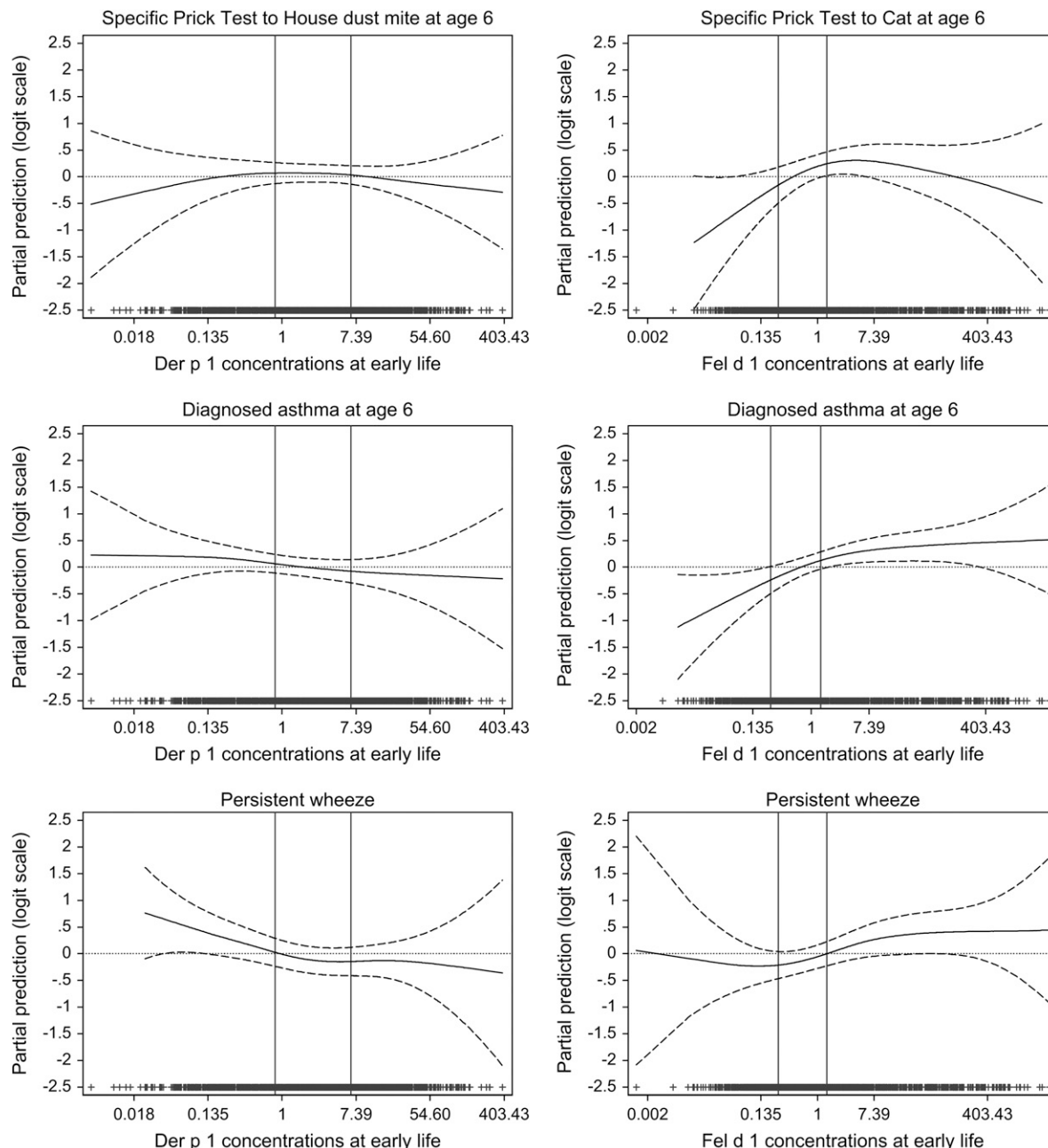


Figure 1. Probability (logit scale) of skin prick test positivity and diagnosis of asthma and wheeze up to 6 years of age according to exposure to house dust mite (Der p1) and cat (Fel d1) allergens in early life. Vertical lines indicate tertile cut points (0.83 and 6.46 μg per gram of dust for Der p1 and 0.25 and 1.39 μg per gram of dust for Fel d1).

We have extensively analyzed possible confounding variables, and none of them showed any significant effect on the relationship between exposure and sensitization or asthma. The main variables found to be associated with sensitization to both allergens were maternal atopy and maternal asthma, whereas high home crowding increased the risk of sensitization to Fel d1 and male subjects showed a higher risk of Der p1 sensitization. For asthma, the main predictor variables were having a positive prick test, maternal and paternal asthma, and having lower respiratory tract infections during the first year of life. A mother smoking during pregnancy or a child being exposed to high levels of NO_2 during early life showed a 50% increase in the risk of asthma or wheeze up to the 6 years of age, although this was not statistically significant. The only protective factor observed was breastfeeding

for at least 2 weeks. An important residual effect by center remained after adjusting for all relevant variables collected.

We have also examined many potential interactions within our data. Only maternal atopy showed any effect. Other authors have reported that any effect of allergen exposure on atopy and asthma is restricted to children of atopic or asthmatic parents and especially children of atopic or asthmatic mothers (28, 29). For diagnosed asthma, we obtained some indication of a restricted effect of allergen exposure, specifically of Fel d1, in children of atopic mothers; this interaction was not seen with maternal asthma. For specific skin prick test positivity, we observed a protective effect of Der p1, especially at very high levels of exposure, among children of atopic mothers, as if a tolerance effect appeared. This effect has not previously been described for

TABLE 4. MULTIVARIATE MODELS FOR POSITIVE PRICK TEST, DIAGNOSED ASTHMA, AND PERSISTENT WHEEZE IN RELATION TO Der p1 AND Fel d1 EXPOSURE

	Positive Specific Prick Test* (n = 1,059)		Diagnosed Asthma (n = 911)		Persistent Wheeze (n = 859)	
	OR	95% CI	OR	95% CI	OR	95% CI
Allergen in dust						
Der p1 concentration†						
0.83–6.46 µg/g	0.88	0.53–1.46	0.67	0.40–1.12	0.59	0.32–1.08
≥6.46 µg/g	0.74	0.42–1.32	0.68	0.37–1.25	0.74	0.38–1.46
	(n = 1,058)		(n = 911)		(n = 858)	
Fel d1 concentration						
0.25–1.39 µg/g	4.09	1.14–14.67	1.59	0.75–3.36	0.73	0.34–1.54
≥1.39 µg/g	4.43	1.22–16.07	2.6	1.27–5.34	1.56	0.79–3.08
Atopy at 6 yr of age‡			3.32	1.94–5.67	5.61	3.24–9.70
Male	—§		—		1.14	0.70–1.87
Firstborn	0.98	0.51–1.89	—		—	
High home crowding	2.65	1.18–5.97	—		—	
Maternal social class						
III, Skilled nonmanual	0.56	0.22–1.42	0.56	0.26–1.21	0.49	0.19–1.26
III, Skilled manual	0.45	0.20–1.03	0.74	0.42–1.30	0.94	0.49–1.79
IV, Unskilled	0.59	0.25–1.42	0.75	0.40–1.43	1.52	0.76–3.03
Maternal atopy	1.84	0.95–3.58	—		1.21	0.72–2.04
Maternal asthma	2.08	0.94–4.58	2.81	1.60–4.96	3.28	1.74–6.18
Paternal asthma	—		2.66	1.51–4.70	2.38	1.25–4.53
LRTI during first year	—		1.82	1.15–2.88	1.47	0.89–2.43
NO ₂ , 10 µg/m ³	—		1.53	0.87–2.70	—	
Smoking during pregnancy	—		1.46	0.85–2.49	1.59	0.89–2.82
Breastfeeding						
2–12 wk	—		0.52	0.29–0.92	0.52	0.28–1.00
>12 wk	—		0.56	0.32–0.98	0.74	0.41–1.33
Barcelona	2.12	0.90–4.96	0.45	0.19–1.06	0.49	0.18–1.38
Menorca	0.25	0.07–0.89	0.22	0.10–0.47	0.79	0.42–1.49

Definition of abbreviations: CI = confidence interval; LRTI = lower respiratory tract infection; OR = odds ratio.

* Prick test to Der p1 or Fel d1 according to the specific allergen in dust.

† Each column presents two multivariate regression models, one for Der p1 and the other for Fel d1, both adjusted for confounding variables. For Der p1, the confounding variables are not shown because odds ratios were similar to those of the Fel d1 model.

‡ Positive skin prick test to any allergen.

§ Not included due to $P > 0.10$.

Der p1, maybe because lower levels of exposure had been observed. Among those with no maternal atopy, the association of allergen exposure with each one of the outcomes studied is similar for Der p1 and Fel d1; the different role of these two allergens seems to be restricted to children with an atopic mother.

The results presented in this article constitute, to our knowledge, the first prospective report on the role of domestic allergen exposures in representative community samples covering a wide range of exposures in different international settings. Although the three environments had different household exposure profiles and incidence rates of atopy and asthma, the role of the allergens studied was similar in the three settings, lending important strength to the generalizability of the results obtained.

After accounting for allergen levels and other relevant variables, the risk of developing sensitization varied greatly between locations, from an odds ratio of 0.25 for Menorca to 2.12 for Barcelona, taking Ashford as reference. The risk of asthma also remained different between locales after multivariate adjustment, although differences were not of the same magnitude and were not parallel to those of sensitization. These differences are especially important when looking at the diagnosis of asthma and less so if looking at wheeze, probably indicating different diagnostic criteria in the three settings. Nonetheless, in the analysis of risk factors, the results are virtually the same when considering asthma or wheeze, giving higher consistency to our findings.

Prospective cohort study designs are the most robust epidemiologic method for assessing the risk of exposures that occur long before the onset of disease, allowing the collection of relevant data at the appropriate time. The main threats to validity are loss to follow-up or missing data. In our study, we had performed prick tests at 6 years of age for 80% of the children for whom early life household allergen measurements were available; 89% had information on diagnosis of asthma at this age. Loss of follow-up was marked in one of the three centers, but we obtained similar results when restricting the analysis to the other two, whose combined follow-up rates were 87% for skin prick test and 95% for a diagnosis of asthma, making it improbable that any important selection bias was introduced in this manner. We recognize the limitations of using a single measure of exposure that may not accurately reflect the total effective relevant exposure; there are, however, data to suggest that allergen levels are consistent over time within households (30). A further limitation is our measurement in dust of only Der p1 (and not Der f1); however, given the high levels of Der p1 found in our study, any effect of mite exposure should have become apparent taking into account Der p1 alone, at least in the United Kingdom center, where Der f1 levels are known to be very low. Very low correlations have been found between Der p1 and Der f1 levels in the European Community Respiratory Health Survey II (31), which allows us to conclude that the relationship observed for Der p1 alone is not confounded by the unmeasured levels of

TABLE 5. ADJUSTED ASSOCIATION BETWEEN Der P1 AND Fel D1 ALLERGEN EXPOSURE AND SPECIFIC PRICK TEST, DIAGNOSED ASTHMA, AND PERSISTENT WHEEZE STRATIFIED BY MATERNAL ATOPY

	Maternal Atopy		No Maternal Atopy		P Value
	OR*	95% CI	OR*	95% CI	
Der p1 concentration					
Positive specific prick test					
0.83–6.46 µg/g	0.45	0.20–1.02	1.26	0.64–2.48	0.061
≥6.46 µg/g	0.28	0.11–0.70	1.44	0.67–3.09	0.003
Diagnosed asthma					
0.83–6.46 µg/g	1.48	0.63–3.48	0.48	0.25–0.91	0.044
≥6.46 µg/g	0.71	0.25–2.02	0.55	0.26–1.17	0.606
Persistent wheeze					
0.83–6.46 µg/g	0.59	0.25–1.41	0.54	0.25–1.15	0.798
≥6.46 µg/g	0.39	0.15–1.03	0.88	0.38–2.05	0.321
Fel d1 concentration					
Positive specific prick test					
0.25–1.39 µg/g	2.61	0.49–13.99	1.23	0.35–4.38	0.852
≥1.39 µg/g	2.34	0.44–12.37	1.82	0.53–6.30	0.852
Diagnosed asthma					
0.25–1.39 µg/g	5.76	0.66–50.68	1.32	0.60–2.88	0.224
≥1.39 µg/g	18.43	2.20–154.0	1.55	0.71–3.41	0.037
Persistent wheeze					
0.25–1.39 µg/g	1.47	0.49–4.38	0.55	0.23–1.34	0.144
≥1.39 µg/g	2.49	0.88–7.07	1.37	0.61–3.08	0.269

Definition of abbreviations: CI = confidence interval; OR = odds ratio.

* Odds ratio adjusted for the same variables as in Table 4.

Der f1; our conclusions in relation to mite exposure refer to Der p1, but, as far as we know, there is no evidence of a differential causal relationship between the different types of mite allergens with atopy or respiratory outcomes. Negative confounding by some unmeasured and potentially protective factor—perhaps endotoxin exposure—is one possibility that we cannot explore with our data.

In summary, we did not find any association between house dust mite exposure and specific sensitization or with asthma or wheeze. For Fel d1 exposure, there was a nonlinear risk curve for all these outcomes. We suggest that the effects of allergen exposure on the inception of atopy or asthma may approach an “all or nothing” event; the different exposure–effect curve observed for the two allergens studied could be explained by the different exposure ranges observed: above the threshold for house dust mite and including the threshold for cat allergen exposure. In addition, we found that the effects of a particular allergen on the inception of atopy and on asthma are very similar. Our data did not include the reported incongruity in the exposure–atopy–asthma relationship (i.e., that exposure would be related to atopy but not to asthma), whereas atopy and asthma are clearly associated. The different effect observed in other studies may be explained by the range of exposures covered, which might include the threshold level for one of the outcomes but not for the other, or could be due to a dominant inclusion of high-risk families. If we are correct, then any exposure thresholds for sensitization or asthma seem to be low, making it improbable that intervention to reduce domestic allergen levels alone will have a major impact on the incidence of sensitization or asthma in childhood.

Conflict of Interest Statement: None of the authors has a financial relationship with a commercial entity that has an interest in the subject of this manuscript.

Acknowledgment: The authors thank the children and their families and all the health professionals and teachers who made this study possible and the nurses who were responsible for the fieldwork and whose personal involvement greatly facilitated the continuing involvement of the cohort families: Carol White, Pamela Mills, Susan Moffat in Ashford, Victoria Estraña and Mireia Garcia in Menorca, and the members of the AMICS study in Barcelona: Gonçal Figueras, Oscar Garcia, and Cecilia Figueras.

References

1. Lau S, Falkenhofst G, Weber A, Werthmann I, Lind P, Buettner-Goetz P, Wahn U. High mite-allergen exposure increases the risk of sensitization in atopic children and young adults. *J Allergy Clin Immunol* 1989;84:718–725.
2. Sporik R, Holgate ST, Platts-Mills TA, Cogswell JJ. Exposure to house-dust mite allergen (Der p 1) and the development of asthma in childhood: a prospective study. *N Engl J Med* 1990;323:502–507.
3. Lau S, Illi S, Sommerfeld C, Niggemann B, Bergmann R, von Mutius E, Wahn U. Multicentre Allergy Study Group. Early exposure to house-dust mite and cat allergens and development of childhood asthma: a cohort study. *Lancet* 2000;356:1392–1397.
4. Pearce N, Douwes J, Beasley R. Is allergen exposure the major primary cause of asthma? *Thorax* 2000;55:424–431.
5. Ingram JM, Sporik R, Rose G, Honsinger R, Chapman MD, Platts-Mills TA. Quantitative assessment of exposure to dog (Can f1) and cat (Fel d1) allergens: relation to sensitisation and asthma among children living in Los Alamos, New Mexico. *J Allergy Clin Immunol* 1995; 96:449–456.
6. Custovic A, Woodcock A. Exposure and sensitisation in infants and children. *Curr Opin Allergy Clin Immunol* 2001;1:133–138.
7. Murray CS, Woodcock A, Custovic A. The role of indoor allergen exposure in the development of sensitization and asthma. *Curr Opin Allergy Clin Immunol* 2001;1:407–412.
8. Wahn U, Lau S, Bergmann R, Kulig M, Forster J, Bergmann K, Bauer CP, Guggenmoos-Holzmann I. Indoor allergen exposure is a risk factor for sensitization during the first three years of life. *J Allergy Clin Immunol* 1997;99:763–769.
9. Platts-Mills TA, Vaughan JW, Squillace S, Woodfolk J, Sporik R. Sensitization, asthma, and a modified Th2 response in children exposed to cat allergen: a population-based cross-sectional study. *Lancet* 2001;357: 752–756.
10. Anto JM. The causes of asthma: the need to look at the data with different eyes. *Allergy* 2004;59:121–123.
11. Almqvist C, Egmar AC, Hedlin G, Lundqvist M, Nordvall SL, Pershagen G, Svartengren M, van Hage-Hamsten M, Wickman M. Direct and indirect exposure to pets: risk of sensitisation and asthma at 4 years in a birth cohort. *Clin Exp Allergy* 2003;33:1190–1197.
12. Ownby DR, Johnson CC, Peterson EL. Exposure to dogs and cats in the first year of life and risk of allergic sensitisation at 6 to 7 years of age. *JAMA* 2002;288:963–972.
13. Sporik R, Platts-Mills TA. Allergen exposure and the development of asthma. *Thorax* 2001;56:ii58–ii63.
14. Gore C, Custovic A. Can we prevent allergy? *Allergy* 2004;59:151–168.

15. Kulig M, Bergmann R, Niggemann B, Burow G, Wahn U; Tehe Mas Study Group. Prediction of sensitization to inhalant allergens in childhood: evaluating family history, atopic dermatitis and sensitization to food allergens. *Clin Exp Allergy* 1998;28:1397–1403.
16. Munir AK, Kjellman NI, Björkstén B. Exposure to indoor allergens in early infancy and sensitization. *J Allergy Clin Immunol* 1997;100:177–181.
17. Wickens K, Pearce N, Siebers R, Ellis I, Patchett K, Sawyer G, Stone L, Tohill S, Kennedy J, Slater T, *et al.* Indoor environment, atopy and the risk of the asthma in children in New Zealand. *Pediatr Allergy Immunol* 1999;10:199–208.
18. Atkinson W, Harris J, Mills P, Moffat S, White C, Lynch O, Jones M, Cullinan P, Newman Taylor AJ. Domestic aeroallergen exposures among infants in an English town. *Eur Respir J* 1999;13:583–589.
19. Basagana X, Torrent M, Atkinson W, Puig C, Barnes M, Vall O, Jones M, Sunyer J, Cullinan P. Domestic aeroallergen levels in Barcelona and Menorca (Spain). *Pediatr Allergy Immunol* 2002;13:412–417.
20. Torrent M, Sunyer J, Muñoz L, Cullinan P, Iturriaga MV, Figueroa C, Vall O, Newman Taylor AJ, Anto JM. Early life domestic aeroallergen exposure and IgE sensitization at age four. *J Allergy Clin Immunol* 2006;118:742–748.
21. Polk S, Sunyer J, Muñoz L, Barnes M, Torrent M, Figueroa C, Harris J, Vall O, Antó JM, Cullinan P. A prospective study of Fel d1 and Der p1 exposure in infancy and childhood wheezing. *Am J Respir Crit Care Med* 2004;170:273–278.
22. Morgan WJ, Stem DA, Sherrill DL, Guerra S, Holberg CJ, Guilbert TW, Taussig LM, Wright AL, Martinez FD. Outcome of asthma and wheezing in the first 6 years of life: follow-up through adolescence. *Am J Respir Crit Care Med* 2005;172:1253–1258.
23. Hölcher B, Frye C, Wichmann HE, Heinrich J. Exposure to pets and allergies in children. *Pediatr Allergy Immunol* 2002;13:334–341.
24. Johnson CC, Ownby DR, Havstad SL, Peterson EL. Family history, dust mite exposure in early childhood, and risk for pediatric atopy and asthma. *J Allergy Clin Immunol* 2004;114:105–110.
25. Huss K, Adkinson NF Jr, Eggleston PA, Dawson C, Van Natta ML, Hamilton RG. House dust mite and cockroach exposure are strong risk factors for positive allergy skin test responses in the Childhood Asthma Management Program. *J Allergy Clin Immunol* 2001;107:48–54.
26. Wickman M, Swartengren M. Allergen exposure and asthma. *Lancet* 2001;357:1042.
27. Cullinan P, MacNeill SJ, Harris JM, Moffat S, White C, Mills P, Newman Taylor AJ. Early allergen exposure, skin prick responses, and atopic wheeze at age 5 in English children: a cohort study. *Thorax* 2004;59:855–861.
28. Brussee JE, Smit HA, van Strien RT, Corver K, Kerkhof M, Wijga AH, Aalberse RC, Postma D, Gerritsen J, Grobbee DE, *et al.* Allergen exposure in infancy and the development of sensitization, wheeze and asthma at 4 years. *J Allergy Clin Immunol* 2005;115:946–952.
29. Celedon JC, Litonjua AA, Ryan L, Platts-Mills T, Weiss ST, Gold DR. Exposure to cat allergen, maternal history of asthma, and wheezing in the first 5 years of life. *Lancet* 2002;360:781–782.
30. Heinrich J, Holscher B, Douwes J, Richter K, Koch A, Bischof W, Fahlbusch B, Kinne RW, Wichmann NE. Reproducibility of allergen, endotoxin and fungi measurements in the indoor environment. *J Expo Anal Environ Epidemiol* 2003;13:152–160.
31. Zock J-P, Heinrich J, Jarvis D, Verlato G, Norba D, Plana E, Sunyer J, Chinn S, Olivieri M, Soon A, *et al.* for the Indoor Working Group of the European Community Respiratory Health Survey II. Distribution and determinants of house dust mite allergens in Europe: the European Community Respiratory Health Survey II. *J Allergy Clin Immunol* 2006;118:682–690.