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Associations Between Asthma and Sensitization to Pet or Pollen Allergens in Young Swedish Twins – The STOPPA Study

Cecilia Lindemalm,[1] Björn Nordlund,[1,2,3] Anne K. Örtqvist,[1] Cecilia Lundholm,[1] Marianne van Hage,[4] Tong Gong,[1] and Catarina Almqvist[1,2]

[1]Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, Stockholm, Sweden
[2]Pediatric Allergy and Pulmonology Unit at Astrid Lindgren Children's Hospital, Karolinska University Hospital, Stockholm, Sweden
[3]Department of Women's and Children's Health, Karolinska Institutet, Stockholm, Sweden
[4]Department of Medicine Solna, Immunology and Allergy Unit, Karolinska Institutet and University Hospital, Stockholm, Sweden

Background: An association between childhood asthma and IgE sensitization has been established, but our understanding of the genetic and environmental contribution to it is incomplete. Our aim was to estimate the associations and dose-response relationship between asthma and sensitization to airborne allergens in Swedish 9- to 14-year-old twins. Additionally, we aimed to explore the importance of familial confounding from shared genes and environment using co-twin controls.
Methods: In the STOPPA cohort, 752 same-sex twin children were screened with Phadiatop® (Thermo Fisher Scientific; Pharmacia, Uppsala, Sweden); if positive further analysis of IgE antibodies to airborne allergens of pets (cat, horse, dog), pollens (birch, timothy, mugwort), mites, and mold were performed. The associations between asthma and airborne allergens were assessed with generalized estimating equations. The co-twin control analysis was performed by conditional logistic regression.
Results: Children with positive Phadiatop® had more than doubled odds of asthma (*OR* 2.53, 95% CI [1.74, 3.70]). Sensitization to pet allergens was associated with increased odds of asthma; for example, cat *OR* 4.15 (95% CI [2.67, 6.45]), with similar estimates for pollens; for example, birch *OR* 3.22 (95% CI [2.12, 4.91]). Associations persisted with sensitization as a categorical variable and for trend, indicating a dose-response relationship. Results remained in the co-twin analyses; for example, cat OR 4.75 (95% CI [1.62, 14.0]) and birch OR 5.00 (95% CI [1.45, 17.3]).
Conclusion: The association between childhood asthma and sensitization to airborne allergens remains in co-twin analyses, indicating they are not due to confounding from shared environmental or genetic factors.

■ **Keywords:** asthma, epidemiology, immunoglobulin E, pediatrics

Allergic sensitization, an immunoglobulin E (IgE) antibody response to common environmental allergens, is associated with asthma (Craig, 2010; Johansson et al., 2004). Furthermore, sensitization to airborne allergens has been shown to be linked to asthma severity, particularly in children (Carroll et al., 2006; Craig, 2010; Simpson et al., 2005). However, not all sensitized individuals develop asthma and, conversely, asthma patients are not always sensitized (Illi et al., 2001). Clinical and epidemiological studies have demonstrated considerable variability in the associations between asthma and total serum IgE (Stromgaard et al., 2011), between asthma and a variety of specific IgEs (sIgEs; Almqvist et al., 2007; Arroyave et al., 2013; Ghunaim et al., 2006; Gruchalla et al., 2005; Hoffmann-Petersen et al., 2013; Lévesque et al., 2005; Rotsides et al., 2010; Simpson et al., 2005), and between asthma and total or sIgEs in geographically disparate cohorts (Weinmayr et al., 2007).

Both environmental factors and genetic effects may play a part in these associations. Asthma heritability studies have provided evidence of a substantial genetic contribution, with estimates varying from 50% to 90% (Thomsen

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ADDRESS FOR CORRESPONDENCE: Professor Catarina Almqvist MD, PhD, Pediatrician, Department of Medical Epidemiology and Biostatistics, PO Box 281, Karolinska Institutet, SE 171 77 Stockholm, Sweden. E-mail: catarina.almqvist@ki.se

et al., 2010; Ullemar et al., 2016). For sensitization, studies have confirmed a genetic influence, albeit with considerable modification by environmental effects (Duffy et al., 1998; Strachan et al., 2001).

To examine the influences of genes and environment, twins as a research population offer unique opportunities. Specifically, a co-twin control design on disease-discordant twin pairs is a powerful methodology when exploring associations between exposure and outcome as confounding — by sex, age, unmeasured shared familial factors, and to some extent genes — can be controlled for (Boomsma et al., 2002). Further, stratification by zygosity may indicate the relative importance of confounding by genetic or familial environmental factors, since dizygotic (DZ) twins share on average 50% of their segregating genes, and monozygotic twins (MZ) share 100%.

Based on the STOPPA cohort (the Swedish Twin study On Prediction and Prevention of Asthma; Almqvist et al., 2015), the aim of this study was to investigate the associations and dose-response relationship between asthma and sensitization to airborne allergens in twin children. Additionally, we aimed to explore the importance of familial confounding from shared genes and environment through a co-twin control analysis.

Methods

Study Design and Recruitment

The STOPPA twins were recruited from the Childhood and Adolescent Twin Study in Sweden cohort (CATSS), a study initiated in 2004 that included all twins born from July 1992 and onwards ($N \sim$23,900 children), identified through the Swedish Twin Registry (Anckarsater et al., 2011). Based on questions validated through the International Study of Asthma and Allergies in Childhood (ISAAC; Asher et al., 1995), on asthma ever (yes/no) and wheezing (current or after three years of age), present in the CATSS interview material, an algorithm was created to identify twins aged 9 to 14 years discordant or concordant for asthma. Since most twin pairs in the CATSS cohort were healthy concordant, a larger proportion of the asthma concordant and asthma discordant pairs were invited to obtain equal-sized categories. In the final STOPPA study population, 31% was classified as asthma concordant, 38% as asthma discordant, and 31% as healthy concordant, according to the recruitment algorithm of asthma status. This has been described in detail elsewhere (Almqvist et al., 2015).

The study was approved by the Regional Ethical review board in Stockholm, Sweden. Informed consent for the study was obtained from all the participants and their parents.

Data Collection

Questionnaires. All 752 twins and their parents completed questionnaires. The parental questionnaire included questions on the parents' background, lifestyle, and medical history, followed by sections on each twin's lifestyle, general health status, medical history with primary focus on respiratory diseases, symptoms and medication, physical activity, stress, and socio-economic factors. The child questionnaire included questions on physical activity, respiratory symptoms, asthma, other allergic diseases, puberty, and stress.

Zygosity. Data on zygosity were obtained from the CATSS study. A majority (82%, 618 children) had their zygosity determined by DNA analysis, with the remaining pairs assessed through an algorithm of five questions on twin similarity, a technique validated to determine zygosity with at least 95% accuracy (Anckarsater et al., 2011).

National health registers. Additional data were drawn from the National Patient Register (NPR) and the Swedish Prescribed Drug Register (SPDR). The NPR contains primary and secondary diagnoses according to the International Classification of Diseases (ICD), from the medical records of all hospitalizations since 1987, and approximately 80% of outpatient specialist care since 2001 in Sweden. The SPDR contains all medications dispensed by pharmacies in Sweden since July 1, 2005, classified according to the Anatomical Therapeutic Chemical classifications system (ATC). The diagnosis code of asthma (ICD-10: J45), inhalations of glucocorticoids (ATC: R03BA), selective beta2-adrenoreceptor agonists (ATC: R03AC), fixed combinations of beta2-agonists and glucocorticoids (ATC: R03AK) and leukotriene receptor antagonists (ATC: R03DC) were the items of interest.

Blood samples. More than 90% of the participants (n = 699) completed blood sampling, analyzed with Phadiatop® (Thermo Fisher Scientific, Uppsala, Sweden), a screening test for common inhalant allergens, registered as positive if $\geq$0.35 kU/l. When so, sera were subsequently analyzed for IgE antibodies to the single allergens of cat-, dog-, and horse-epithelium, birch, timothy, mugwort, mites (*Dermatophagoides pteronyssinus* [*Der p*] and *Dermatophagoides pharinae* [*Der f*]), and mold (*Cladosporium herbarum*), and reported as continuous values, from <0.1 to >100. Cat, dog, and horse allergens were classified as pet allergens; timothy, birch, and mugwort as pollen allergens.

All samples were analyzed at the Department of Clinical Immunology and Transfusion Medicine at the Karolinska University Hospital Solna, Sweden.

Variables

Asthma outcomes. Two asthma outcomes were defined. The first, 'parent-reported asthma', was based on the updated information on the children's morbidity status obtained from the STOPPA parental questionnaire, with positive outcome defined by the questions 'Does your child have

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asthma?' (yes) and/or 'Has your child wheezed in the past 12 months?' (yes). The second, 'register-based asthma', was defined as a physician's diagnosis of asthma recorded in the NPR and/or dispensed medication data from the SPDR. In order not to include one-time asthma medication users, the medication criteria were set to more than two dispenses of glucocorticoids, fixed combinations of glucocorticoids and beta-2-agonists, or leukotriene receptor antagonists independent of time between distributions, or more than three of the aforementioned drugs, with the addition of selective beta-2-agonists, within a 12-month-period (Ortqvist et al., 2013).

Phadiatop and specific (s) IgE. For all sIgEs, binary and categorical variables were created, the latter with ranges <0.35, 0.35–0.69, 0.7–3.4, 3.5–17.4, ≥17.5 kU/l. Continuous sIgE measurements below the level of quantification were assigned the value 0.09 kU/l, and measurements reported as >100 were set to 100 kU/l.

Although sIgE measurements only existed for study participants with positive Phadiatop® tests, it can be inferred that individuals testing negative would not have levels of sIgE ≥0.35 kU/l. Hence, these participants were assigned the value 0.09 kU/l for each sIgE.

Statistical Analyses

Generalized estimating equations (GEE) models with the logit link and exchangeable correlation matrix within twin pairs were used to obtain odds ratios (ORs) with 95% confidence intervals (CIs) for all asthma outcomes, with sensitization levels analyzed as binary, categorical (with test for linear trend), and continuous variables for the all-twins analyses. To investigate possible modification of results by gender or age (9–11 vs. 12–15 years), analyses were repeated with the introduction of interactions terms between these variables and sensitization levels. For the analysis of associations within twin pairs, a co-twin control analysis was conducted among the pairs discordant for both asthma outcome and sensitization, as binary, categorical (with tests for linear trend) and continuous sIgE, using conditional logistic regression models. Here, twin pairs with sensitization data missing for either of the two twins (41 individuals) were automatically excluded. For all the analyses, we used 5% significance level. Statistical analyses were performed using Stata Statistical Software, Release 13 (Stata Corp, 2013).

Results

In the final study population ($n = 752$; 376 pairs) 410 twins were MZ (54%) and 342 DZ (46%). For the two asthma outcomes, the distributions of zygosity, gender, and year of birth were similar to the total study population, except that a larger proportion of MZ twins had register-based asthma. In total, 699 children (93%) were tested for Phadiatop® of whom 277 (40%) screened positive (Table 1).

Table 2 displays the associations between sensitization and parent-reported asthma, obtained by all-twin analyses. Children sensitized to any airborne allergen (≥0.35 kU/l) had a more than two-fold increased odds ratio of parent-reported asthma (OR 2.53, 95% CI: 1.74–3.70) compared to non-sensitized children. There were significantly increased odds for the association between parent-reported asthma and sensitization (binary variable) to all pet and pollen allergens, ranging from OR 4.15 (95% CI [2.67, 6.45]) to OR 6.16 (95% CI [3.52, 10.8]) for pet allergens, and from OR 1.94 (95% CI [1.14, 3.32]) to OR 3.22 (95% CI [2.12, 4.91]) for pollen allergens. The associations persisted when we performed analyses with sensitization as a categorical variable, indicating a dose-response relationship. For the lowest category of sensitization (0.35–0.69 kU/l) odds ratios were, with few exceptions, statistically non-significant, but with higher categories (0.7–3.4, 3.5–17.5 and ≥17.5 kU/l) significant associations were seen between parent-reported asthma and all pet allergens and birch, compared to non-sensitization (all p values for trend <.05). With sensitization as a continuous variable, we observed a 4% (95% CI [1.02, 1.07]) odds increase per kU/l increase in sIgE level for cat, and a corresponding 2% (95% CI [1.01, 1.03]) increased odds per kU/l for birch.

With the outcome register-based asthma, the associations for sensitization (binary variable and trend analysis) for all pets and pollen allergens remained, although with slightly smaller estimates (Table S1). Statistically significant increases in odds were obtained for sensitization as a continuous variable for cat, horse, and birch allergens, ranging from 2% (birch, CI: [1.02, 1.03]) to 4% (horse, CI: [1.02, 1.06]) per kU/l.

The associations between sensitization to any sIgE and any of the asthma outcomes were not significantly modified by age and gender (data not shown).

Results from the co-twin control analyses for the association between sensitization and parent-reported asthma are displayed in Table 3. Compared to the cohort analyses, point estimates remained in the co-twin analyses, although with wider confidence intervals. There was an association between sensitization and parent-reported asthma within all twin pairs (*OR* 2.75, 95% CI [1.22, 6.18]), MZ (*OR* 2.67, 95% CI [0.71, 10.0]), and DZ (*OR* 2.80, 95% CI [1.01, 7.77]), similar to the point estimate in the cohort analyses (2.53). Similarity was also observed for the associations between parent-reported asthma and sensitization to cat within all twin pairs (OR 4.75, 95% CI [1.62, 14.0]), MZ: (OR 4.00) and DZ (*OR* 5.50) as well as for horse allergens for all twin pairs (OR 5.33, 95% CI [1.55, 18.3]), MZ (*OR* 5.00), and DZ (OR 6.00).

Estimates for MZ and DZ asthma-discordant twins exhibited wider confidence intervals, but they were still in line with the odds ratios from the all-twins analyses (Figure 1).

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TABLE 1

Characteristics of the STOPPA Study Cohort

			Parent-reported asthma				Register-based asthma			
	Total	(%)	Yes	(%)	No	(%)	Yes	(%)	No	(%)
	752		177		575		142		610	
MZ	410	(55)	98	(55)	312	(54)	86	(61)	324	(53)
DZ	342	(45)	79	(45)	263	(46)	56	(39)	286	(47)
Male	396	(53)	97	(55)	299	(52)	80	(56)	316	(52)
Female	356	(47)	80	(45)	276	(48)	62	(44)	294	(48)
Age at examination										
9	16	(2)	7	(4)	9	(2)	4	(3)	12	(2)
10	134	(18)	35	(20)	99	(17)	19	(13)	115	(19)
11	142	(19)	45	(25)	97	(17)	30	(21)	112	(18)
12	144	(19)	17	(10)	127	(22)	18	(13)	126	(21)
13	182	(24)	43	(24)	139	(24)	45	(32)	137	(22)
14	122	(16)	26	(15)	96	(17)	19	(13)	103	(17)
15	12	(2)	4	(2)	8	(1)	7	(5)	5	(1)
Serology: Phadiatop										
Performed	699	(93)	161	(91)	538	(94)	126	(89)	573	(94)
Negative (<0.35 kU/l)	422	(56)	69	(39)	353	(61)	51	(36)	371	(61)
Positive (≥0.35 kU/l)	277	(37)	92	(52)	185	(32)	75	(53)	202	(33)
Not performed	53	(7)	16	(9)	37	(6)	16	(11)	37	(6)
Cat (e1) negative	574	(82)	102	(63)	472	(88)	80	(63)	494	(86)
Positive	125	(18)	59	(37)	66	(12)	46	(37)	79	(14)
Dog (e5) negative	587	(84)	101	(63)	486	(90)	78	(62)	509	(89)
Positive	112	(16)	60	(37)	52	(10)	48	(38)	64	(11)
Horse (e3) negative	625	(89)	117	(73)	508	(94)	94	(75)	531	(93)
Positive	74	(11)	44	(27)	30	(6)	32	(25)	42	(7)
Birch (t3) negative	581	(83)	110	(68)	471	(88)	88	(70)	493	(86)
Positive	118	(17)	51	(32)	67	(12)	38	(30)	80	(14)
Timothy (g6) negative	537	(77)	103	(64)	434	(81)	81	(64)	456	(80)
Positive	162	(23)	58	(36)	104	(19)	45	(36)	117	(20)
Mugwort (w6) negative	635	(91)	138	(86)	497	(92)	108	(86)	527	(92)
Positive	64	(9)	23	(14)	41	(8)	18	(14)	46	(8)
Mite (d1) negative	589	(84)	127	(79)	462	(86)	98	(78)	491	(86)
Positive	110	(16)	34	(21)	76	(14)	28	(22)	82	(14)
Mite (d2) negative	589	(84)	130	(81)	459	(85)	100	(79)	489	(85)
Positive	110	(16)	31	(19)	79	(15)	26	(21)	84	(15)
Mold (m2) negative	687	(98)	150	(93)	537	(100)	118	(94)	569	(99)
Positive	12	(2)	11	(7)	1	(0)	8	(6)	4	(1)

Note: Negative IgE <0.35 kU/L; positive IgE ≥0.35 kU/L. MZ = monozygotic twins, DZ = dizygotic twins.

For register-based asthma, a larger proportion of the estimates in the co-twin control analyses were not statistically significant (Table S2). However, the associations for sensitization to all pet and pollen allergens remained with increasing odds, except for horse and mugwort sensitization (trend analyses) where the estimates were smaller.

Discussion

In this cohort of twin children with asthma status determined through both questionnaires, medical records and prescribed medications, combined with objective markers of sensitization, we have demonstrated statistically significant associations between childhood asthma and sensitization to airborne allergens. The results largely remain in co-twin analyses, which adjust for factors shared within twin pairs, indicating they are not due to confounding from shared environmental or genetic factors. We have also found a dose-response relationship between asthma and increasing IgE levels to pollen and pets, which mostly remained in the co-twin control analyses.

Our cohort analyses highlighted significant associations between sensitization to pet and pollen allergens and asthma morbidity, which is in line with several previous studies (Lévesque et al., 2005; Simpson et al., 2005; Simpson et al., 2015). The novelty of our study is that many of these associations remain within twin pairs, although with wider confidence intervals for both MZ and DZ pairs. This has not been shown before, and highlights even further the strength of the associations. Simpson et al. (2015), who in their study on 11-year-old children tested allergen components grouped into categories, found the category including all domestic pet allergens to be most strongly associated with asthma morbidity. Lévesque et al. (2005) found significant differences in sensitization to cat in children with and without asthma, whereas none were observed for dog and timothy sensitization. In accordance with Ghunaim et al. (2006), we noted higher estimates for the associations for birch pollen and asthma than for timothy.

Sensitization patterns and prevalence vary with geographical location (ISAAC, 1998). Prevalence of mold sensitization was low in our cohort. As for mite antibodies, the

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TABLE 2

All Twins Analyses: Odds Ratios and 95% Confidence Intervals for the Association Between Sensitization (Exposure) and Parent-Reported Asthma (Outcome) in the STOPPA Twins

	Binary variable		Categorical variable 0.35–0.69 kU/l		0.7–3.4 kU/l		3.5–17.4 kU/l		≥17.5 kU/l		Trend*		Continuous variable	
Phadiatop	2.53 *277*	[1.74, 3.70]												
Cat (e1)	4.15 *125*	[2.67, 6.45]	1.20 *25*	[0.43, 3.37]	3.51 *33*	[1.77, 6.96]	6.37 *34*	[2.98, 13.6]	7.63 *33*	[3.32, 17.5]	1.74	[1.48, 2.06]	1.04	[1.02, 1.07]
Dog (e5)	5.33 *112*	[3.26, 8.71]	1.53 *27*	[0.58, 3.97]	4.77 *47*	[2.53, 8.98]	19.0 *28*	[7.12, 50.9]	13.4 *10*	[3.69, 48.7]	2.27	[1.81, 2.84]	1.24	[0.99, 1.56]
Horse (e3)	6.16 *74*	[3.52, 10.8]	3.79 *17*	[1.50, 9.56]	4.20 *26*	[1.80, 9.78]	9.20 *19*	[3.30, 25.6]	21.4 *12*	[5.05, 90.4]	2.14	[1.70, 2.69]	1.06	[0.97, 1.16]
Birch (t3)	3.22 *118*	[2.12, 4.91]	0.43 *9*	[0.07, 2.84]	2.09 *38*	[1.05, 4.19]	3.84 *25*	[1.71, 8.60]	5.36 *46*	[2.87, 10.0]	1.52	[1.32, 1.75]	1.02	[1.01, 1.03]
Timothy (g6)	2.37 *162*	[1.59, 3.54]	2.50 *15*	[0.95, 6.56]	2.89 *60*	[1.63, 5.14]	2.37 *45*	[1.23, 4.56]	1.70 *42*	[0.86, 3.38]	1.27	[1.11, 1.45]	1.00	[0.99, 1.02]
Mugwort (w6)	1.94 *64*	[1.14, 3.32]	1.45 *24*	[0.62, 3.38]	1.52 *25*	[0.65, 3.52]	4.45 *15*	[1.65, 12.0]	— *0*	—	1.47	[1.14, 1.88]	1.19	[0.96, 1.49]
Mite1 (d1)	1.64 *110*	[1.01, 2.65]	3.09 *22*	[1.42, 6.73]	1.14 *29*	[0.48, 2.75]	1.69 *31*	[0.82, 3.49]	1.24 *28*	[0.51, 3.04]	1.12	[0.95, 1.32]	1.00	[0.98, 1.02]
Mite2 (d2)	1.45 *110*	[0.90, 2.32]	1.55 *13*	[0.52, 4.68]	1.27 *32*	[0.55, 2.94]	1.87 *23*	[0.84, 4.15]	1.33 *42*	[0.63, 2.78]	1.11	[0.95, 1.30]	1.00	[0.99, 1.02]
Mold (m2)	31.2 *12*	[5.81, 168]	8.35 *5*	[2.02, 34.6]	— *6*	—	— *1*	—	— *0*	—	12.6	[4.02, 39.5]	—	—

Note: Numbers in italics indicate number of cases with positive (≥0.35 kU/l) serology for the binary variables, and within cut-offs for the categorical variables. *Linear trend across the categories.

TABLE 3

Co-Twin Control Analysis: Associations Between Sensitization (Exposure) and Parent-Reported Asthma (Outcome) in the STOPPA Twins

	OR (95% CI)													
	Binary				Trend*				Continuous variable					
	All		MZ		DZ		All		MZ		DZ		All		MZ		DZ	
Phadiatop	2.75	[1.22, 6.18]	2.67	[0.71, 10.0]	2.80	[1.01, 7.77]	1.51	[1.17, 1.95]	1.61	[1.03, 2.53]	1.46	[1.07, 1.98]	1.02	[1.00, 1.04]	1.06	[1.00, 1.13]	1.01	[0.99, 1.03]
	60		*22*		*38*		*98*		*38*		*60*		*166*		*74*		*92*	
Cat (e1)	4.75	[1.62, 14.0]	4.00	[0.85, 18.8]	5.50	[1.22, 24.8]	1.85	[1.23, 2.78]	1.70	[0.99, 2.95]	2.01	[1.09, 3.72]	1.12	[1.00, 1.25]	1.08	[0.97, 1.19]	1.33	[0.93, 1.89]
	46		*20*		*26*		*56*		*26*		*30*		*82*		*44*		*38*	
Dog (e5)	2.88	[1.29, 6.43]	2.75	[0.88, 8.64]	3.00	[0.97, 9.30]	2.08	[1.32, 3.27]	1.92	[1.07, 3.46]	2.29	[1.14, 4.60]	2.98	[1.12, 7.91]	2.27	[0.80, 6.48]	4.49	[0.79, 25.6]
	62		*30*		*32*		*66*		*32*		*34*		*82*		*42*		*40*	
Horse (e3)	5.33	[1.55, 18.3]	5.00	[1.20, 22.8]	6.00	[0.72, 49.8]	2.43	[1.22, 4.82]	2.16	[1.05, 4.45]	3.92	[0.60, 25.7]	4.32	[0.71, 26.3]	2.70	[0.68, 10.8]	–	–
	38		*24*		*14*		*36*		*22*		*14*		*50*		*26*		*24*	
Birch (t3)	5.00	[1.45, 17.3]	2.00	[0.37, 10.9]	11.0	[1.42, 85.2]	1.81	[1.20, 2.73]	1.67	[0.88, 3.15]	1.91	[1.10, 3.29]	1.06	[1.01, 1.11]	1.16	[0.86, 1.56]	1.05	[1.00, 1.10]
	36		*12*		*24*		*50*		*22*		*28*		*68*		*34*		*34*	
Timothy (g6)	2.83	[1.12, 7.19]	1.33	[0.30, 5.96]	4.33	[1.23, 15.2]	1.22	[0.90, 1.65]	0.96	[0.55, 1.68]	1.35	[0.93, 1.95]	1.00	[0.98, 1.02]	0.97	[0.84, 1.12]	1.00	[0.98, 1.02]
	46		*14*		*32*		*62*		*18*		*44*		*88*		*36*		*52*	
Mugwort (w6)	2.00	[0.50, 8.00]	—	—	6.00	[0.72- 49.8]	1.64	[0.73, 3.66]	0.47	[0.07, 3.25]	2.65	[0.77, 9.04]	1.15	[0.72, 1.83]	1.37	[0.44, 4.26]	1.10	[0.67, 1.83]
	18		*4*		*14*		*20*		*6*		*14*		*46*		*18*		*28*	

Note: Numbers in italics indicate number of exposure- and disease-discordant children. Some OR estimates were not possible to calculate due to co-variation in data; in all of those disease-discordant twin pairs, the twin with asthma also had the highest rate of IgE. *Linear trend across the categories. MZ = monozygotic twins, DZ = dizygotic twins.

prevalence was on par with the levels of pets and pollen antibodies. Still, we only observed a weak association between sensitization to *Der p* as a binary variable and parent-reported asthma. Previous studies have published conflicting results on the associations between asthma and mite sensitization (Custovic et al., 2015; Gruchalla et al., 2005; Ronmark et al., 2003; von Hertzen & Haahtela, 2009). In keeping with previous studies we found a distinct dose-response relationship, with increasing odds of asthma with increasing sIgE levels of cat, dog, horse, birch, and mugwort (Arroyaye et al., 2013; Gruchalla et al., 2005), but not of timothy (Rotsides et al., 2010).

The point estimates of our co-twin control analyses were similar to those in the all-twins analyses, in particular for cat and horse sensitization. Moreover, we found comparable estimates for MZ and DZ twins, although with wide confidence intervals. Thus, we found no indication of genetic confounding. Only a few previous twin studies have explored the genetic and environmental factors regulating asthma phenotypes. Wu et al. (2010) showed that genetic effects accounted for a substantial part of the variation in asthma phenotypes such as total IgE, sIgEs, and skin prick tests, whereas Strachan et al. (2001) found that genetic factors influence susceptibility to allergic disease and sensitization to airborne allergens, although with a considerable modifying role for environmental factors.

Although many epidemiological studies have focused on the connection between asthma and sensitization, only a fraction of these have deployed the powerful methodologies of twin designs, and the ones that have (Duffy et al., 1998; Strachan et al., 2001; Wu et al., 2010), have exclusively studied adult populations. Our study based on the STOPPA twins is, to our knowledge, the first on twin children. Its strengths are the relatively large sample size, combined with validated data from questionnaires, national health registers and objective markers of allergic sensitization, collected in a standardized manner, thus ensuring commensurable results.

Still, the results of this study should be assessed together with its limitations. Recall bias as well as seasonal variability of asthma symptoms may have influenced questionnaire data. To mitigate the latter we chose the time frame of the preceding year when inquiring about respiratory symptoms. As for recall bias it is noteworthy that our results based on questionnaire data were confirmed by those based on data from external sources. An additional limitation was insufficient power in the final co-twin control analyses, resulting in wide confidence intervals of the odds ratios. However, the results largely support the findings from the cohort analysis, indicating positive associations between asthma and sensitization to pet and birch allergens. Furthermore, objections toward the assumptions inherent in twin models, such as absence of gene-environment interaction and their generalizability to non-twin populations have been brought forward. Twins differ from singletons in several aspects,

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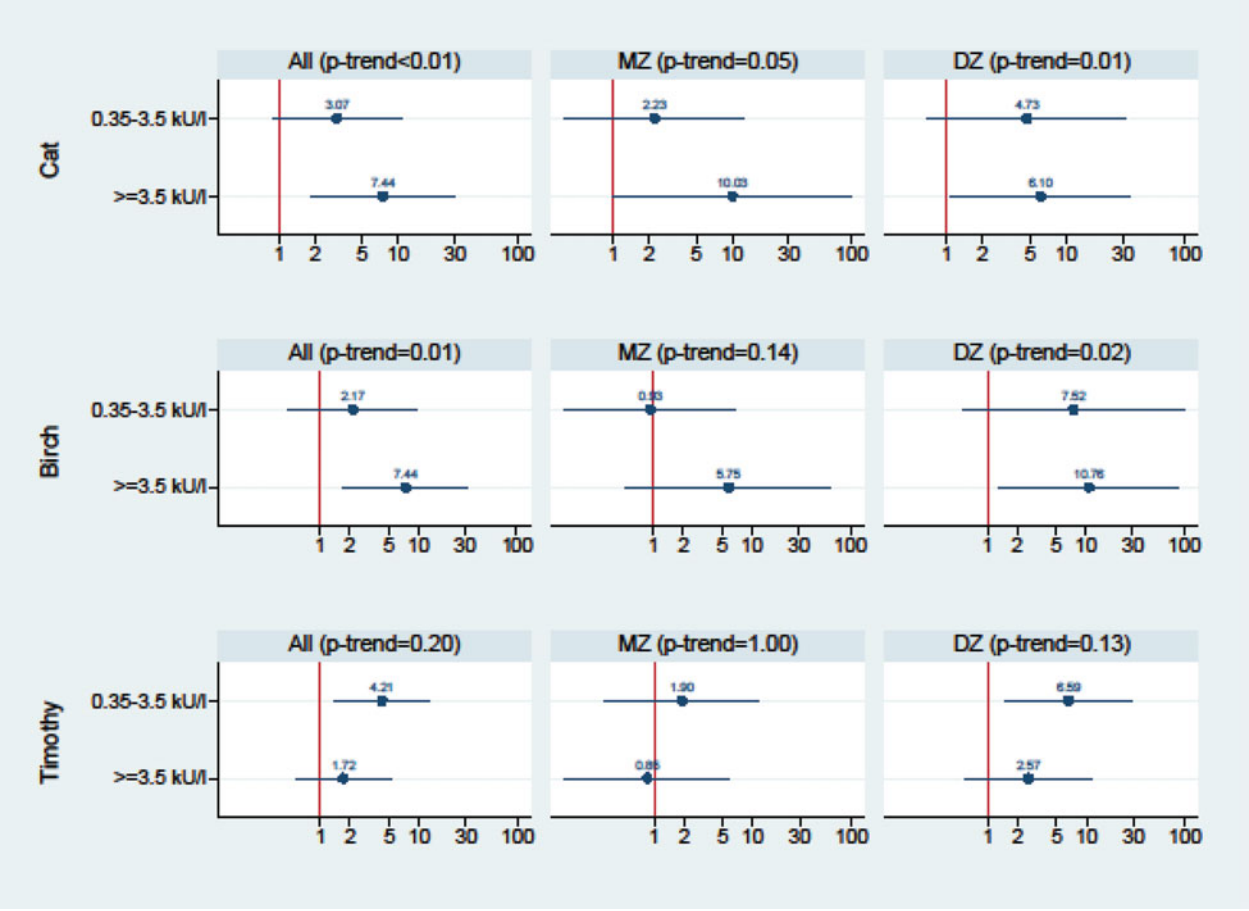

FIGURE 1

(Colour online) Linear trend for the association between parent-reported asthma and IgE-sensitization (0.35–3.5 kU/L and ≥3.5 kU/L) to cat, birch, and timothy within all twin pairs, MZ and DZ twins.

such as birth weight and neonatal respiratory morbidity. Regardless, several studies (Thomsen et al., 2008; Ullemar et al., 2015) have concluded that results from twin studies on asthma can be generalized to the general population.

In conclusion, we believe the insights obtained in this paper add to the knowledge on how to interpret sensitization among children, suggesting that sIgE levels to pet and pollen allergens are strongly associated with asthma morbidity. Since a single diagnostic test currently does not exist for asthma, a more detailed understanding of the sensitization patterns that pose the higher risk of disease may provide valuable addition to the models of asthma prediction and prognosis. Emerging therapeutic interventions in allergic disorders emphasize the need to identify the correct patient groups.

Authors' Contributions

This study was initiated by CA and designed by CLi, CLu, TG and CA. BN, AÖ, MvH and CLi participated in data acquisition, and CLi performed the data analysis under supervision of TG, CA, and CLu. All authors participated in interpretation of data. CLi drafted the article and all authors revised the paper critically for important intellectual content and approved the final version of the manuscript.

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Conflict of Interest

Marianne van Hage has received lecture fees from Thermo Fisher Scientific.

Supplementary material

To view supplementary material for this article, please visit https://doi.org/10.1017/thg.2017.48

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