

Predictive value of food sensitization and filaggrin mutations in children with eczema

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Background: It was reported that in infants with eczema and food sensitization, the presence of a filaggrin (*FLG*) null mutation predicts future asthma with a specificity and positive predictive value of 100%.

Objectives: We sought to evaluate the predictive value of food sensitization and food allergy, *FLG* haploinsufficiency, and their combination in infants with early-onset eczema for persistent eczema and childhood asthma.

Methods: The German Infant Nutritional Intervention (GINI) and Influence of Lifestyle-related Factors on the Immune System and the Development of Allergies in Childhood (LISA)

birth cohorts, as well as a collection of 65 cases of early-onset eczema with and without food allergy were investigated. **Results:** The risk for asthma was significantly increased by food sensitization (positive diagnostic likelihood ratios [PLRs] of 1.9 [95% CI, 1.1-3.4] in the GINI cohort and 5.5 [95% CI, 2.8-10.8] in the LISA cohort) and the presence of an *FLG* mutation (PLRs of 2.9 [95% CI, 1.2-6.6] in the GINI cohort and 2.8 [95% CI, 1.0-7.9] in the LISA cohort) with a rather high specificity (79.1% and 92.9% in the GINI cohort and 89.0% and 91.7% in the LISA cohort, respectively) but low sensitivity (40.0% and 39.3% in the GINI cohort and 31.6% and 23.5% in the LISA cohort, respectively). Likewise, the risk for persistent eczema was increased. In the clinical cases neither food allergy nor *FLG* mutations had a significant effect. The combination of both parameters did not improve prediction and reached positive predictive values of 52.3% (GINI cohort), 66.9% (LISA cohort), and 30.6% (clinical cases), assuming an asthma prevalence in children with early eczema of 30%.

Conclusion: Early food sensitization and the presence of an *FLG* mutation in infants with early eczema increase the risk for later asthma, but the combination of the 2 factors does not represent a clinically useful approach to reliably identify children at risk. (J Allergy Clin Immunol 2011;128:1235-41.)

Key words: Eczema, atopic dermatitis, asthma, food sensitization, food allergy, filaggrin, prediction

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Atopic dermatitis (eczema) is the most common inflammatory skin disease in childhood, with prevalence estimates of up to 20%.¹ Eczema is commonly associated with increased levels of total serum IgE antibodies and aberrant IgE-mediated responses to environmental agents. However, up to 50% of infants and children with eczema do not exhibit detectable specific IgE (sIgE) antibodies.²⁻⁴ In those with early sensitization, the spectrum shifts from common food allergens (hen's egg, cow's milk, wheat, and soy) at age 1 year to aeroallergens at age 6 years.⁵ Notably, subjects can have allergic sensitization to food allergens, as determined by skin prick testing or sIgE measurement, without having clinical symptoms on exposure to those foods.⁶ Therefore diagnosis of IgE-mediated food allergy requires both the presence of sensitization and the development of specific signs and symptoms on exposure (ie, noneczematous immediate-type anaphylactic symptoms, eczematous delayed-type reactions, or combinations

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Abbreviations used

DBPCFC:	Double-blind, placebo-controlled food challenge
FLG:	Filaggrin
GINI:	German Infant Nutritional Intervention
LISA:	Influence of Lifestyle-related Factors on the Immune System and the Development of Allergies in Childhood
MAS:	Multicenter Allergy Study
NPV:	Negative predictive value
PLR:	Positive diagnostic likelihood ratio
PPV:	Positive predictive value
RR:	Relative risk
sIgE:	Specific IgE

of both). Through not yet understood mechanisms, the time course of food allergy resolution in children varies by food and mostly occurs in early school-aged years.⁷

Eczema runs a chronic fluctuating course; about two thirds of children with early-onset eczema show spontaneous remission until early adolescence, whereas up to 20% have persistent eczema, and a further 17% show intermittent symptoms by age 7 years. Known risk factors for persisting eczema include severe disease, early allergic sensitization, and 2 or more first-degree relatives with atopic disease.⁸

Up to 50% of patients with childhood eczema go on to have asthma later in life.^{5,9,10} However, it has been suggested that not eczema *per se* but rather early-onset eczema with specific allergic sensitization constitutes a risk for asthma.^{8,11} In particular, early sensitization to hen's egg^{12,13} and persistent (>1 year) sensitization to food allergens in children with early eczema¹⁴ were reported to increase the risk for subsequent asthma and rhinitis. Findings from the German Multicenter Allergy Study (MAS) imply that allergic rhinitis until the age of 5 years is associated with wheezing between the ages of 5 and 13 years, yet this association is not attributable to eczema.¹⁵

To date, null mutations in the filaggrin gene (*FLG*) are the strongest and most widely replicated risk factor for eczema, particularly early-onset and persistent eczema with allergic sensitizations, and asthma in the context of eczema.¹⁶⁻¹⁸ A very recent work indicated that *FLG* mutations might also be of relevance for peanut allergy independently from the presence of eczema.¹⁹

On the basis of observations made in the MAS cohort, it was further suggested that the combination of eczema, sensitization to food allergens, and *FLG* haploinsufficiency predicts childhood asthma with a specificity and a positive predictive value (PPV) of as much as 100%.²⁰ This is of great interest because it would mean that every infant with this combination will have asthma, that these infants could be identified early in life with the help of simple and rather easily measurable parameters, and that they could be subjected to early interventions and targeted prevention measures.²¹ Overestimation of absolute risk, however, might lead to inadequate or inappropriate intervention in subjects whose asthma risk is actually lower than anticipated. Therefore we were interested to validate these observations and also to evaluate the usefulness of food sensitization and *FLG* mutation status for the prediction of the course of eczema. To this end, we investigated the German GINI and LISA birth cohorts, as well as a collection of carefully phenotyped children with eczema with or without confirmed food allergy.

METHODS**Study populations and phenotyping****German Infant Nutritional Intervention birth cohort.**

For the German Infant Nutritional Intervention (GINI) birth cohort, a total of 5991 full-term newborn infants were recruited from 2 regions of Germany (Munich and Wesel) between 1995 and 1998. The cohort is composed of intervention (n = 2252) and nonintervention (n = 3739) groups. For the current study, the intervention group was used. Briefly, the intervention study was a prospective, randomized double-blind trial designed primarily to examine the effect of different hydrolyzed infant formulas on the development of allergic diseases. Children with a positive family history of allergic disease were enrolled and invited to clinical examinations at 1, 4, 8, 12, and 36 months of age. For the first, second, and third years, diagnosis of eczema was made by using an algorithm of diagnostic criteria, as described previously.²² sIgE levels against cow's milk, casein, α -lactalbumin, β -lactoglobulin, egg white, and soy allergen were measured at ages 12 and 36 months (Pharmacia and Upjohn Diagnostics AB, Uppsala, Sweden). Sensitization was defined as levels of 0.70 kU/L or greater (RAST class ≥ 2) against at least 1 of these allergens at 12 or 36 months. The whole birth cohort was followed up to now 10 years by self-administered questionnaires. At 6 and 10 years, a visit to the study center for clinical examination and blood sampling was offered. In participants of the 10-year follow-up visit, sIgE levels against aeroallergens (*Dermatophagoides pteronyssinus*, *Dermatophagoides farinae*, German cockroach, cat dander, mixed molds, timothy grass, mugwort, English plantain, ribwort, wall pellitory, and birch pollen) were determined. Sensitization was defined as an sIgE concentration of 0.35 kU/L or greater against at least 1 of the allergens.

Details of the design, recruitment, and follow-up of the study have been described previously.²²⁻²⁵ The outcomes presented here are based on parental reports in the 10-year follow-up questionnaire. Asthma and eczema were defined as present if any physician had diagnosed asthma or eczema during the last 4 years, if the child was treated for asthma (eczema) in the last 12 months, or both. In the present analysis we included subjects with a clinical diagnosis of eczema in the first 3 years, known status of food sensitization, and available *FLG* genotypes who participated in the 10-year follow-up by questionnaire.

Influence of Lifestyle-related Factors on the Immune System and the Development of Allergies in Childhood birth cohort.

For the Influence of Lifestyle-related Factors on the Immune System and the Development of Allergies in Childhood (LISA) birth cohort, 3097 newborns were initially recruited between 1997 and 1999 from 4 German cities: Munich, Leipzig, Wesel, and Bad Honnef. Data on the children's health and on lifestyle factors were collected by using repeated parent-completed questionnaires at regular time intervals during the first 2 years of life (6, 12, 18, and 24 months) and at 4, 6, and 10 years of age. Early eczema was defined if 1 positive answer was given to the following question: "Has a doctor diagnosed your child with allergic or atopic eczema in the past 6 months?"

Blood samples were drawn at 2, 6, and 10 years of age. At 2 years of age, total IgE and sIgE levels against a set of common food allergens (egg white, cow's milk, wheat, peanut, soybean, and codfish) were measured by using the CAP-RAST FEIA system (Pharmacia Diagnostics, Freiburg, Germany). Allergic sensitization against food allergens was defined as a specific serum IgE concentration of 0.7 kU/L or greater against at least 1 of the allergens. The design and objectives of this prospective birth cohort study were described in detail elsewhere.²⁶ Total IgE and sIgE levels against aeroallergens were measured at 10 years of age. The definition of outcome variables at age 10 years is identical to that of the GINI cohort.

Clinical case series. Of children with eczema aged 0 to 2 years who had been recruited in Munich for genetic studies between 2004 and 2006 (for details, see Weidinger et al¹⁶) and whose parents had given informed consent to be recontacted for follow-up, we randomly selected and reinvited 50 children with well-defined food allergy, as well as 50 children without any detectable sensitization and no history of adverse reactions to food. sIgE data from an enzyme immunoassay (CAP-FEIA; Pharmacia, Uppsala, Sweden) for a panel of food allergens (egg white, cow's milk, α -lactalbumin, β -lactoglobulin, peanut, soybean, wheat, codfish, and casein) and aeroallergens (mixed grass pollen, *D pteronyssinus*, *D farinae*, birch pollen, mugwort pollen, cat

TABLE I. Characteristics of early-onset eczema cases at recruitment and follow-up stratified by food allergy/sensitization

	GINI cohort		LISA cohort		Clinical cases	
	Food sensitization (at 3 y)		Food sensitization (at 2 y)		Food allergy*	
	Yes (n = 44)	No (n = 144)	Yes (n = 28)	No (n = 212)	Yes (n = 38)	No (n = 23)
Sex (male/female)	26/18	68/76	19/9	106/106	30/8	8/15
Mean SCORAD score (SD)	24.2 (10.4)	18.3 (7.7)	—	—	34.5 (18.7)	27.1 (8.4)
Geometric mean of total IgE (kU/L [SD])	121.5 (3.5)	27.1 (3.3)	109.9 (3.0)	18.2 (3.3)	244.7 (6.3)	36.6 (5.2)
<i>FLG</i> mutation carriers, n/N	7/29 (24.1%)	8/82 (9.8%)	5/18 (27.8%)	10/121 (8.3%)	10/38 (26.3%)	8/23 (34.8%)
Follow-up						
Persistent eczema, n/N	16/43 (37.2%)	38/139 (27.3%)	9/28 (32.1%)	39/210 (18.6%)	23/38 (60.5%)	16/23 (69.6%)
Mean SCORAD score (SD)	—	—	—	—	29.5 (14.26)	23.0 (9.45)
Asthma, n	10 (22.7%)	15 (10.4%)	11 (39.3%)	17 (8.0%)	18 (47.4%)	6 (26.1%)
Rhinitis, n/N	15/43 (34.9%)	32/140 (22.9%)	10/28 (35.7%)	37/207 (17.9%)	16/38 (42.1%)	10/23 (43.5%)
Geometric mean of total IgE (kU/L [SD])	247.2 (3.6)	82.3 (5.0)	244.7 (5.0)	66.7 (5.0)	473.4 (4.9)	154.5 (4.4)
Food sensitization†/allergy*, n/N	8/31 (25.8%)	15/104 (14.4%)	8/22 (36.4%)	19/141 (13.5%)	25/38 (65.8%)	0
Sensitization against aeroallergens, n/N	25/31 (80.6%)	57/104 (54.8%)	20/22 (90.9%)	79/141 (56.0%)	36/38 (94.7%)	15/23 (65.2%)

Percentages refer to subjects with available data.

*Clinical case series: diagnosis confirmed by means of DBPCFC.

†RAST class ≥ 2 .

dander, dog dander, and *Cladosporium herbarum*) were available from these children.

Food allergy was defined by (1) a convincing history of reactions to egg white and/or cow's milk and/or peanut and/or other food; (2) detectable IgE antibodies against the suspected food in serum (positively defined as ≥ 0.7 kU/L); and (3) positive results on double-blind, placebo-controlled food challenges (DBPCFCs) performed at the time of recruitment, as described previously²⁷ and in accordance with national guidelines.²⁸

Parents were invited to answer a mail-in questionnaire, which included items on respiratory and allergic diseases taken from the European Community Respiratory Health Survey and International Study of Asthma and Allergies in Childhood questionnaires^{29,30} to collect information on health outcomes, allergic symptoms, a physician's diagnosis of allergic diseases, and covariates, such as children's nutrition, environmental tobacco smoke exposure, and pet ownership. Overall, 75 (75%) subjects returned the completed questionnaire, and of those, 65 agreed to take part in clinical examinations at the local study center between 2009 and 2010. The respective children underwent a skin examination by experienced dermatologists and venous blood sampling. Both total IgE and sIgE levels against the same panel of food allergens and aeroallergens were measured (CAP-FEIA).

Subjects were classified as having asthma or allergic rhinitis when they reported a history of a physician's diagnosis of asthma or rhinitis at follow-up, whereas eczema was diagnosed on the basis of the skin examination using the UK diagnostic criteria for eczema.

All study methods followed the Declaration of Helsinki protocols and were approved by the ethics committee of the Bavarian Medical Chamber.

Genotyping

Genotyping for *FLG* mutations (R501X and 2282del4 in the GINI and LISA cohorts and R501X, 2282del4, R2447X, and S3247X in the case series) was performed as described previously.^{17,31}

Statistical analysis

Descriptive statistics for quantitative and qualitative values are presented as means $\pm$ SDs and relative frequencies or absolute numbers, respectively.

For the comparison of baseline data between groups, standard tests, such as *t* tests or the Fisher exact test, were used, and for the comparison of follow-up data, relative risks (RRs) with 95% CIs are given. Standard measures of diagnostic tests, such as sensitivity, specificity, and positive diagnostic likelihood ratio (PLR; calculated as sensitivity/[1 – specificity]), are given to evaluate

the prediction of a trait by risk factors and their combinations. PPVs and negative predictive values (NPVs) with 95% CIs were calculated for various possible proportions of the disease in populations. Statistical analyses were done with the statistical software SAS for Windows, version 9.2 (SAS Institute, Inc, Cary, NC). In the case series descriptive analyses were performed with the software R2.12.0.³²

RESULTS

Characteristics of the study population

The study populations comprised 188 GINI children, 240 LISA children, and 65 clinical cases of early eczema (eczema within the first 2 or 3 years of life). At baseline, the median objective SCORAD score was 18 (minimum, 7.1; maximum, 52) in the GINI cohort and 22 (minimum, 7.1; maximum, 75.5) in the clinical cases; the SCORAD score was not determined in the LISA cohort.

In the GINI and LISA cohorts the children were followed up to age 10 years, whereas in the clinical case series the observation period was shorter (mean, 8 years). Table I provides an overview of the clinical characteristics of the children stratified by food sensitization/allergy. The study population comprised 44 and 28 children with and 144 and 212 children without food sensitization in the GINI and LISA cohorts, respectively. *FLG* genotypes were available from 121 children in the GINI cohort and 139 children in the LISA cohort. The *FLG* mutation carrier frequencies in these children with early eczema were considerably higher in those displaying a food sensitization (24.1% vs 9.8% in the GINI cohort and 27.8% vs 8.3% in the LISA cohort). The clinical collection of early eczema cases comprised 38 children with and 23 children without food allergy. The frequency of *FLG* mutations in the clinical case series did not differ significantly between patients with and without food allergy (Table I).

Prediction of asthma

In the GINI and LISA study populations, at follow-up, physician-diagnosed asthma was reported by 13.6% and 11.2% of the children with early eczema, with significantly higher rates in early

TABLE II. Basic description of early-onset eczema cases stratified by *FLG* genotype

	GINI cohort		LISA cohort		Clinical cases	
	<i>FLG</i> mutation		<i>FLG</i> mutation		<i>FLG</i> mutation	
	Yes (n = 19)	No (n = 118)	Yes (n = 15)	No (n = 134)	Yes (n = 20)	No (n = 45)
Mean SCORAD score	24.2 (12.3)	19.2 (7.7)	—	—	38.3 (12.9)	29.9 (16.4)
Geometric mean of total IgE (kU/L [SD])	40.4 (5.0)	33.1 (4.5)	3.3 (1.1)	3.1 (1.4)	99.5 (5.5)	109.9 (8.2)
Food sensitization*/allergy,† n/N	7/15 (46.7%)	22/96 (22.9%)	5/15 (33.3%)	13/124 (10.5%)	10/18 (55.1%)	28/43 (65.1%)
Follow-up						
Persistent eczema, n/N	6/18 (33.3%)	33/116 (28.4%)	4/15 (26.7%)	23/133 (17.3%)	14/20 (70.0%)	28/45 (62.2%)
Mean SCORAD score (SD)	—	—	—	—	43.07 (11.58)	32.52 (17.09)
Asthma, n	6 (31.6%)	13 (11.0%)	4 (26.7%)	13 (9.7%)	6 (30.0%)	18 (40.0%)
Rhinitis, n/N	6/19 (31.6%)	32/115 (27.8%)	4/15 (26.7%)	24/130 (18.5%)	12/20 (60.0%)	16/45 (35.6%)
Geometric mean of total IgE (kU/L [SD])	221.4 (3.7)	121.5 (4.5)	5.0 (2.0)	4.3 (1.6)	221.4 (4.5)	330.3 (5.5)
Sensitization against aeroallergens, n/N	10/14 (71.4%)	57/91 (62.6%)	10/11 (90.9%)	67/113 (59.3%)	17/20 (85.0%)	38/45 (84.4%)

Percentages refer to subjects with available data.

*RAST class ≥ 2 .

†Clinical case series: diagnosis confirmed by means of DBPCFC.

TABLE III. Prediction of asthma at age 7 to 10 years in children with early eczema from the GINI and LISA cohorts

	No. not missing predictors, N	Frequency of positive predictors (%)	TP, n	FP, n	TN, n	FN, n	Sensitivity (% [95% CI])	Specificity (% [95% CI])	PLR	PPV for prevalence of 30%	NPV for prevalence of 30%
Predictor: food sensitization											
GINI cohort	188	23.4	10	34	129	15	40.0 (21.1-61.3)	79.1 (72.1-85.1)	1.9 (1.1-3.4)	45.1 (31.8-59.1)	75.5 (68.9-81.1)
LISA cohort	240	11.7	11	17	195	17	39.3 (21.5-59.4)	92.9 (88.5-96.0)	5.5 (2.8-10.8)	70.3 (54.8-82.3)	78.1 (72.6-82.8)
Predictor: <i>FLG</i> mutation											
GINI cohort	137	13.9	6	13	105	13	31.6 (12.6-56.6)	89.0 (81.9-94.0)	2.9 (1.2-6.6)	55.2 (34.8-74.9)	75.2 (69.0-80.6)
LISA cohort	149	10.1	4	11	121	13	23.5 (6.8-49.9)	91.7 (85.6-95.8)	2.8 (1.0-7.9)	54.8 (30.3-77.2)	73.7 (68.1-78.4)
Predictor: <i>FLG</i> mutation and food sensitization											
GINI cohort	111	6.3	2	5	91	13	13.3 (1.7-40.6)	94.8 (88.3-98.3)	2.6 (0.5-12.0)	52.3 (18.9-83.8)	71.8 (67.5-75.8)
LISA cohort	139	3.6	2	3	119	15	11.8 (1.5-36.4)	97.5 (93.0-99.5)	4.8 (0.9-26.6)	66.9 (26.8-91.8)	72.1 (68.4-75.5)

FN, False negative; FP, false positive; TN, true negative; TP, true positive.

food-sensitized children (GINI cohort: RR, 2.2; 95% CI, 1.1-4.5; LISA cohort: RR, 4.8; 95% CI, 2.6-9.4) and in the presence of an *FLG* mutation (GINI cohort: RR, 2.9; 95% CI, 1.2-6.6; LISA cohort: RR, 2.7; 95% CI, 1.0-7.4). Twenty-four (36.9%) of the clinical eczema cases had developed asthma up to the follow-up examination. Children with food allergy at recruitment more often had asthma than children without food allergy (47.4% vs 26.1%; RR, 1.8; 95% CI, 0.8-7.9), whereas there was no clear difference between *FLG* mutation carriers and noncarriers (Tables I and II).

For food sensitization, we observed PLRs of 1.9 (95% CI, 1.1-3.4) and 5.5 (95% CI, 2.8-10.8), and for *FLG* mutations, we observed PLRs of 2.9 (95% CI, 1.2-6.6) and 2.8 (95% CI, 1.0-7.9) in the GINI and LISA cohorts, respectively. If both predictors were used, PLRs did not increase significantly (Table III). In the GINI cohort the best predictor was *FLG* status, whereas in the LISA cohort and in the clinical cases the best predictor was food allergy/sensitization. Logistic regression models confirmed the results, also after adjustment for sex, parental education, and atopy (see Table E1 in this article's Online Repository at www.jacionline.org). Assuming a prevalence of asthma in children with early eczema of 30% (as observed in MAS), the PPV of food sensitization in combination with the presence of an

FLG mutation was estimated to be 52.3% (95% CI, 18.9% to 83.8%) and 66.9% (95% CI, 26.8% to 91.8%) in the GINI and LISA cohorts, respectively. In the clinical cases food allergy plus an *FLG* mutation reached a PPV of 30.6% (95% CI, 12.2% to 58.4%; Table IV and see Fig E1 in this article's Online Repository at www.jacionline.org). Specificity was rather high (94.8%, 97.5%, and 83.8% in the GINI cohort, the LISA cohort, and the clinical cases, respectively), whereas sensitivity was very low (13.3%, 11.8%, and 16.7%, respectively; Tables III and IV).

Results for wheezing as the outcome were similar to those for asthma (see Table E2 in this article's Online Repository at www.jacionline.org).

Risk for persistent eczema

In the GINI and LISA cohorts persistent eczema was defined if parents reported a physician's diagnosis of eczema in the 7th to 10th year or treatment of eczema in the 10th year. In the case series we used report of "itchy rash during the previous 12 months" plus a clinical diagnosis of eczema during the skin examination at follow-up.

In total, 30.3% of the GINI children, 20.6% of the LISA children, and 64.6% of the children with clinical cases of early

TABLE IV. Prediction of asthma and persistent eczema in clinical eczema cases

Predictor	No. not missing predictors, N	Frequency of positive predictors (%)	TP, n	FP, n	TN, n	FN, n	Sensitivity (% [95% CI])	Specificity (% [95% CI])	PLR	PPV for prevalence of 30%	NPV for prevalence of 30%
Outcome: asthma											
Food allergy	61	62.3	18	20	17	6	75.0 (53.3-90.2)	46.0 (29.5-63.1)	1.4 (0.9-2.0)	37.3 (29.9-46.5)	81.1 (66.4-90.3)
<i>FLG</i> mutation	65	30.8	6	14	27	18	25.0 (9.8-46.7)	65.9 (49.4-79.9)	0.7 (0.3-1.7)	23.9 (12.2-41.5)	67.2 (59.8-73.8)
Food allergy + <i>FLG</i> mutation	61	16.4	4	6	31	20	16.7 (4.7-37.4)	83.8 (71.0-93.7)	1.0 (0.3-3.3)	30.6 (12.2-58.4)	70.1 (65.1-74.7)
Outcome: persistent eczema											
Food allergy	61	62.3	23	15	7	16	59.0 (42.1-74.4)	31.8 (13.9-54.9)	0.9 (0.6-1.3)	27.0 (20.1-35.3)	64.4 (46.9-78.8)
<i>FLG</i> mutation	65	30.8	14	6	17	28	33.3 (19.6-49.6)	73.9 (51.6-89.8)	1.3 (0.6-2.9)	35.4 (19.6-55.1)	72.1 (65.2-78.1)
Food allergy + <i>FLG</i> mutation	61	16.4	8	2	20	31	20.5 (9.3-36.5)	90.9 (70.8-98.9)	2.3 (0.5-9.7)	49.1 (18.3-80.6)	72.7 (68.4-76.6)

FN, False negative; FP, false positive; TN, true negative; TP, true positive.

TABLE V. Prediction of persistent eczema at age 7 to 10 years in children with early eczema from the GINI and LISA cohorts

	No. not missing predictors, N	Frequency of positive predictors (%)	TP, n	FP, n	TN, n	FN, n	Sensitivity (% [95% CI])	Specificity (% [95% CI])	PLR	PPV for prevalence of 30%	NPV for prevalence of 30%
Predictor: food sensitization											
GINI cohort	183	24.0	17	27	101	38	30.9 (19.1-44.8)	78.9 (70.8-85.6)	1.5 (0.9-2.5)	38.6 (27.2-51.3)	72.7 (68.6-76.5)
LISA cohort	238	11.8	9	19	171	39	18.8 (9.0-32.6)	90.0 (84.8-93.9)	1.9 (0.9-3.9)	44.6 (28.0-62.5)	72.1 (69.1-74.9)
Predictor: <i>FLG</i> mutation											
GINI cohort	133	12.8	6	11	83	33	15.4 (5.9-30.5)	88.3 (80.0-94.0)	1.3 (0.5-3.3)	36.1 (18.3-58.6)	70.9 (67.6-73.9)
LISA cohort	148	10.1	4	11	110	23	14.8 (4.4-33.7)	90.9 (84.3-95.4)	2.8 (1.0-7.9)	41.1 (19.4-66.9)	71.3 (67.8-74.6)
Predictor: <i>FLG</i> mutation and food sensitization											
GINI cohort	108	5.6	2	4	74	28	6.7 (0.8-22.1)	94.9 (87.4-98.6)	1.3 (0.25-6.7)	36.0 (9.8-74.4)	70.4 (68.0-72.6)
LISA cohort	138	3.6	2	3	111	22	8.3 (1.0-27.0)	97.4 (92.5-99.5)	3.2 (0.6-17.9)	57.8 (19.3-88.6)	71.3 (68.6-73.7)

FN, False negative; FP, false positive; TN, true negative; TP, true positive.

eczema still had eczema at follow-up. In the GINI and LISA cohorts persistent eczema was observed slightly more often in children who had displayed early food sensitization and in those carrying an *FLG* mutation; however, this difference was not statistically significant. In the clinical cases no significant difference between children with and without food allergy was observed, and the presence of an *FLG* mutation slightly but not significantly increased risk for persistent eczema (Tables I and II).

In the birth cohorts a specificity of 78.9% or greater and 88.3% or greater and a sensitivity of 30.9% or less and 15.4% or less was observed for food sensitization and *FLG* mutations as diagnostic criteria to predict eczema persistence, whereas sensitivity for both predictors was low (6.7% and 8.3%). In the GINI and LISA cohorts PLRs were 1.5 (95% CI, 0.9-2.5) and 1.9 (95% CI, 0.9-3.9) for food sensitization and 1.3 (95% CI, 0.5-3.3) and 2.8 (95% CI, 1.0-7.9) for *FLG* mutations, respectively (Table V). No clear effects were observed in the clinical case series (Table IV). If both predictors were used in combination, only a very moderate improvement was observed in the clinical cases and the LISA cohort (Tables IV and V). For an estimated prevalence of persistent eczema in 7- to 10-year-old children with a history of early-onset eczema of 30%, the PPV of the combination of food sensitization/allergy and the presence of an *FLG* mutation for persistent eczema was 36.0% (GINI cohort), 57.8% (LISA cohort), and 49.1% (clinical cases; Tables IV and V).

Sensitization patterns

In the 2 birth cohorts, total IgE levels were significantly higher in those sensitized to food allergens at baseline both at recruitment and follow-up ($P < .001$). Specific sensitizations against aeroallergens at follow-up were present in 80.6% and 90.9% of the children with early eczema and food sensitization versus 54.8% and 56.0% of the children with early eczema without food sensitization at recruitment ($P = .008$ and $P = .002$), respectively. In the clinical case series at follow-up, specific sensitizations against aeroallergens were observed in 36 (94.7%) compared with 15 (65.2%, $P = .003$), with a median number of sensitizations against 10 and 2 allergens, respectively. Twenty-five (65.8%) of the children with food allergy at baseline reported persistence of this food allergy at follow-up (Table I), with the majority of them having allergy to egg (see Table E3 in this article's Online Repository at www.jacionline.org). Geometric mean total IgE levels did not differ by *FLG* mutation status neither at recruitment nor at follow-up in any of the sets.

DISCUSSION

In this study we analyzed the effect of early food sensitization or food allergy and *FLG* haploinsufficiency in children with early eczema on the course of eczema and the risk for asthma. To this end, we used a complementary approach and investigated 2 large birth

cohorts (the GINI and LISA cohorts) and a well-characterized series of tertiary care patients who had undergone DBPCFCs.

Although in general children with early eczema, food sensitization, and *FLG* mutations more often had asthma than children with early eczema and no food sensitization or *FLG* mutations, our observations do not confirm the results from a previous report from the MAS cohort,²⁰ which found that the combination of early eczema, sensitization to food allergens within the first 3 years of life, and the presence of an *FLG* mutation predicts the development of asthma with a PPV of 100%. In fact, using comparable criteria in the GINI and LISA cohorts, we estimated PPVs of 52.3% and 66.9% for sensitization and the presence of an *FLG* mutation. Notably, the combination of *FLG* null alleles and sensitization to food allergens did not substantially improve the prediction of asthma beyond the single factors.

The observations made in MAS relied on a subgroup of high-risk children, which was further enriched for children with infantile eczema ($n = 180$ with complete data) and finally comprised 10 children with the hypothesized combination of risk factors.²⁰ Food sensitization in the previous study, as well as in our birth cohorts, was defined as the presence of an sIgE level of 0.70 kU/L or greater. Although the absence of sIgE reliably excludes the presence of IgE-mediated food allergy, high cutoff levels are needed to correctly identify children as having food allergy.^{33,34} To specifically investigate food allergy as trait of interest, we therefore examined also a collection of 38 infants with early eczema and symptomatic IgE-mediated food allergy, as confirmed by means of DBPCFCs, and 23 infants with early eczema but without food allergy. Also in this collection the PPV of food allergy plus an *FLG* mutation for asthma was only 30.6%, and no improved prediction was observed for the combination of the parameters.

Across the 3 samples, the specificity of asthma prediction by food sensitization and the presence of an *FLG* mutation was rather high (83.3% to 97.5%), whereas the sensitivity was very low (11.8% to 16.7%), indicating a high proportion of false-negative predictions; that is, a considerable proportion of children at risk for asthma are missed.

In this context also the technical requirements for a conclusive *FLG* test result and the potential for variability between different *FLG* genotyping methods have to be considered. Although a small typing error rate is tolerable for rapid large-scale genotyping of large collections, for routine use in clinical practice, where clinical decisions have to be made with confidence, a thorough analytic validation of testing methods together with a high standard of quality assurance would be critical to develop an accurate and reliable *FLG* mutation test assay.

Notably, PPV, NPV, sensitivity, and specificity reflect population characteristics and do not easily translate to individual patients. Likelihood ratios for a test result are a more useful way of expressing the power of diagnostic tests in estimating how much a test result will change the odds of having the condition. PLRs of greater than 10 are generally considered conclusive, indicating high accuracy. In our study the PLRs for the combination of food sensitization or food allergy and the presence of an *FLG* mutation were rather small (GINI cohort, 2.6; LISA cohort, 4.8; and clinical cases, 1.0) and not high enough for clinical purposes. In addition, the PLRs of the combined parameters were not superior of those of each parameter considered in isolation.

When interpreting our results compared with those of the previous study, it should be considered that whereas in MAS

asthma was defined as an episode of wheeze during the previous year, a definition that might be overly sensitive, we used a parental report of physician-diagnosed asthma, which is the International Study of Asthma and Allergies in Childhood definition used in many large-scale epidemiologic and genetic studies of childhood asthma. However, in our sets, results for wheeze in the past 12 months were comparable to those for parental report of asthma. As in the original report, in our analyses only a limited number of children displayed the risk factors of interest. Finally, the methods of follow-up assessment were different across our cohorts. However, we think that the use of 3 independent sets (ie, a high-risk birth cohort [GINI], a population-representative birth cohort [LISA], and a clinical collection of children with eczema with and without food allergy) allowed us to critically assess the previously reported results. We found that in line with previous studies,^{8,35} *FLG* mutations, more severe disease (as reflected by our clinical case series), and early food sensitization each increase the risk for persistent eczema and the development of inhalational allergies. However, the predictive value of the combination of food sensitization and *FLG* mutations with regard to asthma is considerably lower than proposed, and their clinical utility to predict asthma is further limited by a very low sensitivity. We suggest that our results reflect the complexity of atopic diseases, their still insufficiently understood interrelationship, and the yet limited knowledge on their pathophysiology. It appears unlikely that a reliable prediction of asthma can be achieved by evaluating a few risk factors only, but we suggest that a better understanding of different risk factors and predictors might aid in the definition of patient subpopulations for more effective modes of care and the development of more targeted sets of preventive strategies. From a public health perspective, our results are important and demonstrate the need for other risk prediction scores that include specific biomarkers that are sufficiently uncorrelated with established clinical risk factors and achieve clinical utility.

Key messages

- Early sensitization to food and the presence of an *FLG* mutation in infants with early-onset eczema each increase the risk for subsequent asthma.
- Early sensitization to food and the presence of an *FLG* mutation in infants with early-onset eczema each increase the risk for persistent eczema.
- The combined determination of sensitization status and *FLG* genotype in infants with early-onset eczema does not allow a reliable prediction of asthma.

REFERENCES

1. Bieber T. Atopic dermatitis. *N Engl J Med* 2008;358:1483-94.
2. Novak N, Bieber T. Allergic and nonallergic forms of atopic diseases. *J Allergy Clin Immunol* 2003;112:252-62.
3. Williams H, Flohr C. How epidemiology has challenged 3 prevailing concepts about atopic dermatitis. *J Allergy Clin Immunol* 2006;118:209-13.
4. Flohr C, Johansson SG, Wahlgren CF, Williams H. How atopic is atopic dermatitis? *J Allergy Clin Immunol* 2004;114:150-8.
5. Kulig M, Bergmann R, Klettke U, Wahn V, Tacke U, Wahn U. Natural course of sensitization to food and inhalant allergens during the first 6 years of life. *J Allergy Clin Immunol* 1999;103:1173-9.
6. Eller E, Kjaer HF, Host A, Andersen KE, Bindslev-Jensen C. Food allergy and food sensitization in early childhood: results from the DARC cohort. *Allergy* 2009;64:1023-9.

7. Wood RA. The natural history of food allergy. *Pediatrics* 2003;111:1631-7.
8. Illi S, von Mutius E, Lau S, Nickel R, Gruber C, Niggemann B, et al. The natural course of atopic dermatitis from birth to age 7 years and the association with asthma. *J Allergy Clin Immunol* 2004;113:925-31.
9. Kapoor R, Menon C, Hoffstad O, Bilker W, Leclerc P, Margolis DJ. The prevalence of atopic triad in children with physician-confirmed atopic dermatitis. *J Am Acad Dermatol* 2008;58:68-73.
10. Ricci G, Patrizi A, Baldi E, Menna G, Tabanelli M, Masi M. Long-term follow-up of atopic dermatitis: retrospective analysis of related risk factors and association with concomitant allergic diseases. *J Am Acad Dermatol* 2006;55:765-71.
11. Lowe AJ, Abramson MJ, Hosking CS, Carlin JB, Bennett CM, Dharmage SC, et al. The temporal sequence of allergic sensitization and onset of infantile eczema. *Clin Exp Allergy* 2007;37:536-42.
12. Nickel R, Kulig M, Forster J, Bergmann R, Bauer CP, Lau S, et al. Sensitization to hen's egg at the age of twelve months is predictive for allergic sensitization to common indoor and outdoor allergens at the age of three years. *J Allergy Clin Immunol* 1997;99:613-7.
13. Warner JO. A double-blinded, randomized, placebo-controlled trial of cetirizine in preventing the onset of asthma in children with atopic dermatitis: 18 months' treatment and 18 months' posttreatment follow-up. *J Allergy Clin Immunol* 2001;108:929-37.
14. Kulig M, Bergmann R, Tacke U, Wahn U, Guggenmoos-Holzmänn I. Long-lasting sensitization to food during the first two years precedes allergic airway disease. The MAS Study Group, Germany. *Pediatr Allergy Immunol* 1998;9:61-7.
15. Rochat MK, Illi S, Ege MJ, Lau S, Keil T, Wahn U, et al. Allergic rhinitis as a predictor for wheezing onset in school-aged children. *J Allergy Clin Immunol* 2010;126:1170-5.e2.
16. Weidinger S, Illig T, Baurecht H, Irvine AD, Rodriguez E, Diaz-Lacava A, et al. Loss-of-function variations within the filaggrin gene predispose for atopic dermatitis with allergic sensitizations. *J Allergy Clin Immunol* 2006;118:214-9.
17. Weidinger S, O'Sullivan M, Illig T, Baurecht H, Depner M, Rodriguez E, et al. Filaggrin mutations, atopic eczema, hay fever, and asthma in children. *J Allergy Clin Immunol* 2008;121:1203-9.e1.
18. Rodriguez E, Baurecht H, Herberich E, Wagenpfeil S, Brown SJ, Cordell HJ, et al. Meta-analysis of filaggrin polymorphisms in eczema and asthma: robust risk factors in atopic disease. *J Allergy Clin Immunol* 2009;123:1361-70.e7.
19. Brown SJ, Asai Y, Cordell HJ, Campbell LE, Zhao Y, Liao H, et al. Loss-of-function variants in the filaggrin gene are a significant risk factor for peanut allergy. *J Allergy Clin Immunol* 2011;127:661-7.
20. Marenholz I, Kerscher T, Bauerfeind A, Esparza-Gordillo J, Nickel R, Keil T, et al. An interaction between filaggrin mutations and early food sensitization improves the prediction of childhood asthma. *J Allergy Clin Immunol* 2009;123:911-6.
21. Sly PD, Boner AL, Bjorksten B, Bush A, Custovic A, Eigenmann PA, et al. Early identification of atopy in the prediction of persistent asthma in children. *Lancet* 2008;372:1100-6.
22. von Berg A, Koletzko S, Grubl A, Filipiak-Pittroff B, Wichmann HE, Bauer CP, et al. The effect of hydrolyzed cow's milk formula for allergy prevention in the first year of life: the German Infant Nutritional Intervention Study, a randomized double-blind trial. *J Allergy Clin Immunol* 2003;111:533-40.
23. von Berg A, Filipiak-Pittroff B, Kramer U, Link E, Bollrath C, Brockow I, et al. Preventive effect of hydrolyzed infant formulas persists until age 6 years: long-term results from the German Infant Nutritional Intervention Study (GINI). *J Allergy Clin Immunol* 2008;121:1442-7.
24. von Berg A, Koletzko S, Filipiak-Pittroff B, Laubereau B, Grubl A, Wichmann HE, et al. Certain hydrolyzed formulas reduce the incidence of atopic dermatitis but not that of asthma: three-year results of the German Infant Nutritional Intervention Study. *J Allergy Clin Immunol* 2007;119:718-25.
25. Filipiak B, Zutavern A, Koletzko S, von Berg A, Brockow I, Grubl A, et al. Solid food introduction in relation to eczema: results from a four-year prospective birth cohort study. *J Pediatr* 2007;151:352-8.
26. Zutavern A, Brockow I, Schaaf B, Bolte G, von Berg A, Diez U, et al. Timing of solid food introduction in relation to atopic dermatitis and atopic sensitization: results from a prospective birth cohort study. *Pediatrics* 2006;117:401-11.
27. Zuberbier T, Edenharter G, Worm M, Ehlers I, Reimann S, Hantke T, et al. Prevalence of adverse reactions to food in Germany—a population study. *Allergy* 2004;59:338-45.
28. Werfel T, Erdmann S, Fuchs T, Henzgen M, Kleine-Tebbe J, Lepp U, et al. Approach to suspected food allergy in atopic dermatitis. Guideline of the Task Force on Food Allergy of the German Society of Allergology and Clinical Immunology (DGAKI) and the Medical Association of German Allergologists (ADA) and the German Society of Pediatric Allergology (GPA). *J Dtsch Dermatol Ges* 2009;7:265-71.
29. Weiland SK, von Mutius E, Hirsch T, Duhme H, Fritzsche C, Werner B, et al. Prevalence of respiratory and atopic disorders among children in the East and West of Germany five years after unification. *Eur Respir J* 1999;14:862-70.
30. Burney PG, Luczynska C, Chinn S, Jarvis D. The European Community Respiratory Health Survey. *Eur Respir J* 1994;7:954-60.
31. Cramer C, Link E, Horster M, Koletzko S, Bauer CP, Berdel D, et al. Elder siblings enhance the effect of filaggrin mutations on childhood eczema: results from the 2 birth cohort studies LISAPlus and GINIplus. *J Allergy Clin Immunol* 2010;125:1254-60.e5.
32. Team RDC. R: a language and environment for statistical computing. Vienna (Austria): Team RDC; 2009. Available at: <http://www.R-project.org>. Accessed October 20, 2011.
33. Ahrens B, Lopes de Oliveira LC, Schulz G, Borres MP, Niggemann B, Wahn U, et al. The role of hen's egg-specific IgE, IgG and IgG4 in the diagnostic procedure of hen's egg allergy. *Allergy* 2010;65:1554-7.
34. Sampson HA. Utility of food-specific IgE concentrations in predicting symptomatic food allergy. *J Allergy Clin Immunol* 2001;107:891-6.
35. Weidinger S, Rodriguez E, Stahl C, Wagenpfeil S, Klopp N, Illig T, et al. Filaggrin mutations strongly predispose to early-onset and extrinsic atopic dermatitis. *J Invest Dermatol* 2007;127:724-6.

APPENDIX 1

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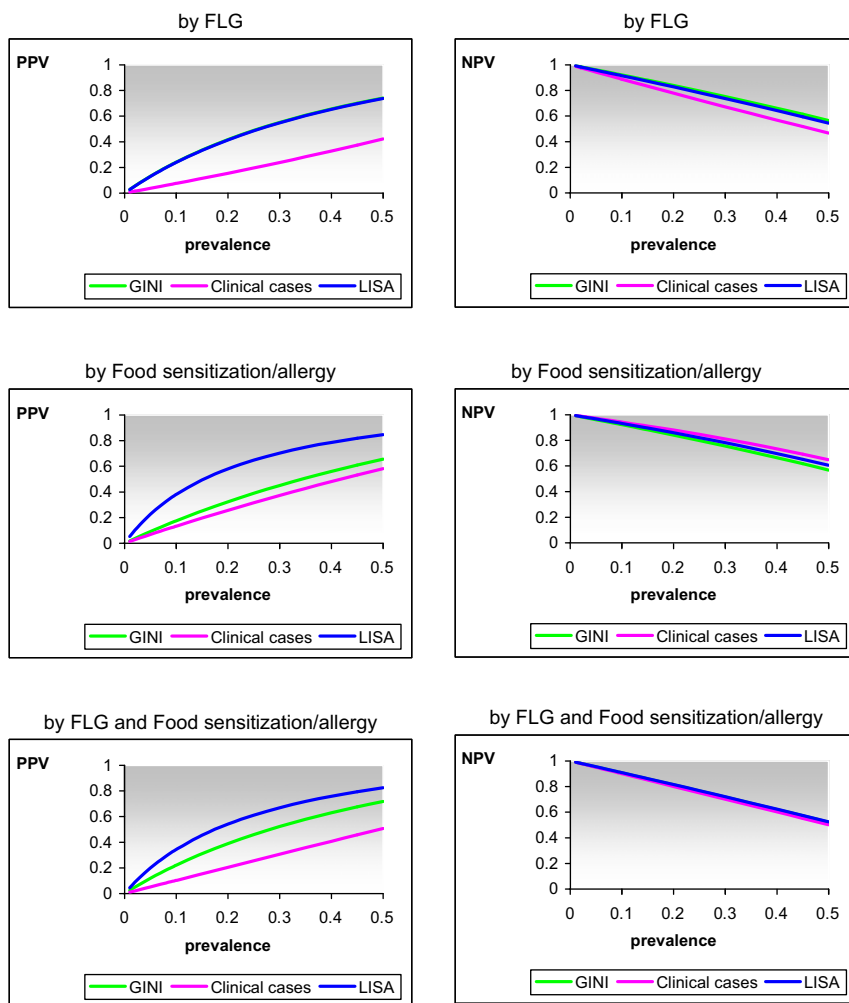


FIG E1. PPVs and NPVs (probability from 0 to 1) for the prediction of asthma at 7 to 10 years in children with early eczema for the prevalence of asthma (probability between 0 and 0.5) by different predictors.

TABLE E1. Influence of interested predictors on asthma by logistic models in the GINI and LISA cohorts

Model	GINI cohort (n = 111)		LISA cohort (n = 139)	
	Food sensitization (at 3 y)	FLG mutation	Food sensitization (at 2 y)	FLG mutation
M0 (single-factor models)	2.1 (0.7-6.8)	4.3 (1.2-15.1)	7.1 (2.2-22.3)	3.1 (0.9-11.2)
M1 (both factors)*	1.7 (0.5-5.6)	3.8 (1.1-13.9)	6.2 (1.9-20.2)	2.0 (0.5-8.2)
M2 (both, adjusted for sex, parental education, and parental atopy) [†]	1.7 (0.5-5.7)	3.8 (1.0-14.2)	6.3 (1.8-22.5)	1.8 (0.4-7.1)
M3 (both, adjusted for parental asthma) [‡]	3.1 (0.7-12.7)	2.4 (0.5-10.5)	5.4 (1.5-19.7)	2.3 (0.5-10.9)

Values are presented as odds ratios with 95% CIs.

*Interaction was not significant.

[†]Not necessary as a confounder and no influence on asthma.

[‡]Parental asthma has significant influence on asthma in the child.

TABLE E2. The prediction of wheezing in the 10th year of life in children with early eczema from the GINI and LISA cohorts

	No. not missing predictors, N	Frequency of positive predictors (%)	TP, n	FP, n	TN, n	FN, n	Sensitivity (% [95% CI])	Specificity (% [95% CI])	PLR	PPV for prevalence of 30%	NPV for prevalence of 30%
Predictor: food sensitization											
GINI cohort	192	22.9	11	33	128	20	35.5 (19.2-54.6)	79.5 (72.4-85.5)	1.7 (0.99-3.0)	42.6 (29.7-56.6)	74.2 (68.6-79.1)
LISA cohort	233	12.0	10	18	182	23	30.3 (15.6-48.7)	91.0 (86.2-94.6)	3.6 (1.9-6.9)	59.1 (42.2-74.0)	75.3 (70.8-79.3)
Predictor: <i>FLG</i> mutation											
GINI cohort	140	13.6	4	15	102	19	17.4 (5.0-38.8)	87.2 (79.7-92.6)	1.4 (0.5-3.7)	36.8 (17.5-61.5)	71.1 (66.9-75.1)
LISA cohort	148	10.1	4	11	122	11	26.7 (7.8-55.1)	91.7 (85.7-95.8)	3.2 (1.2-8.9)	58.0 (33.4-79.1)	74.5 (68.2-79.9)
Predictor: <i>FLG</i> mutation and food sensitization											
GINI cohort	114	6.1	2	5	91	16	11.1 (1.4-34.7)	94.8 (88.3-98.3)	2.1 (0.4-10.2)	47.8 (16.1-81.3)	71.3 (67.7-74.7)
LISA cohort	138	3.6	2	3	121	12	14.3 (1.8-42.8)	98.4 (94.3-99.8)	8.8 (1.3-57.6)	79.3 (36.8-96.2)	72.8 (68.4-76.9)

FN, False negative; FP, false positive; TN, true negative; TP, true positive.

TABLE E3. Description of children with eczema and food allergy

No. of children with milk allergy at recruitment	24 (63.2%)
No. of children with egg allergy at recruitment	29 (76.3%)
No. of children with peanut allergy at recruitment	7 (18.4%)
No. of children with other food allergy at recruitment	5 (13.2%)
No. of children with gastrointestinal symptoms at recruitment —	
in the context of milk allergy	4
in the context of egg allergy	10
in the context of peanut allergy	0
No. of children with skin symptoms at recruitment —	
in the context of milk allergy	24
in the context of egg allergy	23
in the context of peanut allergy	5
No. of children with lung symptoms —	
in the context of milk allergy	2
in the context of egg allergy	3
in the context of peanut allergy	3
No. of children with anaphylactic symptoms at recruitment —	
in the context of milk allergy	0
in the context of egg allergy	1
in the context of peanut allergy	1
No. of children with other symptoms at recruitment —	
in the context of milk allergy	0
in the context of egg allergy	1
in the context of peanut allergy	0
No. of children with persistent food allergy at follow-up	25 (65.79%)