

Sensitization to Cor a 9 and Cor a 14 is highly specific for a hazelnut allergy with objective symptoms in Dutch children and adults

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Background: Component-resolved diagnosis has been shown to improve the diagnosis of food allergy.

Objective: We sought to evaluate whether component-resolved diagnosis might help to identify patients at risk of objective allergic reactions to hazelnut.

Method: A total of 161 hazelnut-sensitized patients were included: 40 children and 15 adults with objective symptoms on double-blind, placebo-controlled food challenges (DBPCFCs) and 24 adults with a convincing objective history were compared with 41 children and 41 adults with no or subjective symptoms on DBPCFCs (grouped together). IgE levels to hazelnut extract and single components were analyzed with ImmunoCAP.

Results: IgE levels to hazelnut extract were significantly higher in children with objective than with no or subjective symptoms. In 13% of children and 49% of adults with hazelnut allergy with objective symptoms, only sensitization to rCor a 1.04 was observed and not to other water-soluble allergens. Sensitization to rCor a 8 was rare, which is in contrast to rCor a 1. Sensitization to nCor a 9, rCor a 14, or both was strongly associated with hazelnut allergy with objective symptoms. By using adapted cutoff levels, a diagnostic discrimination between severity groups was obtained. IgE levels to either nCor a 9 of 1 kU_A/L or greater or rCor a 14 of 5 kU_A/L or greater (children) and IgE levels to either nCor a 9 of 1 kU_A/L or greater or rCor a

14 of 1 kU_A/L or greater (adults) had a specificity of greater than 90% and accounted for 83% of children and 44% of adults with hazelnut allergy with objective symptoms.

Conclusion: Sensitization to Cor a 9 and Cor a 14 is highly specific for patients with objective symptoms in DBPCFCs as a marker for a more severe hazelnut allergic phenotype. (*J Allergy Clin Immunol* 2013;■■■:■■■-■■■.)

Key words: Component-resolved diagnosis, diagnostics, double-blind, placebo-controlled food challenge, hazelnut allergy, severity, food allergy

The clinical presentation of hazelnut allergy is highly variable and ranges from local and mild to systemic and severe allergic reactions.¹ The prevalence and severity of hazelnut allergy are different between children and adults. The reported prevalence of hazelnut allergy is approximately 0.2% among children² and up to 4.5% among adults from birch-endemic areas.^{3,4} In a birch-endemic area children with hazelnut allergy have often objective symptoms (67%)⁵⁻⁷ in contrast to mainly subjective oral symptoms in adults (93%).^{8,9} A differential sensitization profile might be responsible for this difference. Sensitization to the cross-reactive lipid transfer protein Cor a 8 has mainly been described in patients from the Mediterranean area,^{10,11} with a strong association with severe allergic symptoms. Evidence that sensitization to Cor a 9, an 11S globulin, might be associated with severe hazelnut allergy in children has been reported both in the United States and Europe.^{5,12} In birch pollen-related hazelnut allergy,^{10,13} sensitization to Cor a 1, the Bet v 1 homolog belonging to the pathogenesis-related protein 10 family, has been implicated as a cause of typically mild and local symptoms, mainly in adults.^{5,9,10} The clinical relevance of sensitization to profilin in hazelnut (Cor a 2, another pollen-related allergen) seems limited,¹⁴ and sensitization to cross-reactive carbohydrate determinants (CCDs) is now well established as being clinically inert.^{15,16} More recently identified allergens in hazelnut are the oleosins Cor a 12 and Cor a 13¹⁷ and Cor a 14, belonging to the 2S albumins.¹⁸ The clinical relevance for sensitization to the oleosins and Cor a 14 from hazelnut still needs to be evaluated.

Sensitization to hazelnut extract can occur regardless of whether patients react to hazelnut with severe or mild symptoms or even at all and is therefore not discriminative.^{5,7} Previous studies have suggested that component-resolved diagnosis might increase the diagnostic specificity and might predict the severity of an allergic reaction to hazelnut.^{5,10-12} The number of patients with hazelnut allergy with objective symptoms was limited in

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Abbreviations used

AUC: Area under the curve

CCD: Cross-reactive carbohydrate determinant

DBPCFC: Double-blind, placebo-controlled food challenge

previous studies, especially in adults from birch-endemic areas. The aim of this study was to analyze the sensitization to the currently available components rCor a 1, rCor a 8, nCor a 9, and rCor a 14 in a population of hazelnut-sensitized children and adults from a birch-endemic area. A large group of children, as well as adults, responding with objective symptoms on a double-blind, placebo-controlled food challenge (DBPCFC) or having a convincing history of hazelnut allergy with objective symptoms was enrolled to evaluate whether component-resolved diagnosis can help identify patients with objective allergic reactions to hazelnut. This group was compared with a group of hazelnut-sensitized children and adults with a negative DBPCFC result or with only subjective symptoms.

METHODS**Patient selection**

Patients sensitized to hazelnut (≥ 0.35 kU_A/L) were retrospectively recruited. The aim was to enroll equally powered groups of approximately 40 children and adults with a more severe phenotype, as judged based on objective symptoms in DBPCFCs (or, if the DBPCFC was declined, based on a convincing history of hazelnut allergy with objective symptoms) and similar numbers of sensitized children and adults without allergy or a mild phenotype as judged based on no or subjective symptoms in DBPCFCs, respectively. Forty patients per group were necessary to build a prediction model with IgE to 4 hazelnut components as variables. A total of 164 hazelnut-sensitized patients were enrolled. Three patients were excluded from further analyses because they lost their previously determined hazelnut sensitization. The final study population therefore consisted of 161 sensitized patients: 81 children and 80 adults. On the basis of the challenge outcome (performed in all 81 children and 56 adults) or a convincing objective history (24 adults), the population was divided into 79 patients (40 children and 39 adults) with an objective phenotype and 82 patients (41 children and 41 adults) without an objective phenotype (negative DBPCFC results or subjective symptoms on DBPCFCs, Fig 1). For those reporting adverse reactions to hazelnut in daily life, a detailed clinical history was recorded. In addition, data were collected on asthma, birch pollinosis, and atopic dermatitis.

The study was performed at the University Medical Center Utrecht between 2010 and 2012. The study was approved by the local ethics committee, and all patients provided written informed consent before entering the study.

DBPCFCs

DBPCFCs with hazelnut were performed during diagnostic workup, as previously described,⁶ and discontinued if objective symptoms occurred. Suggestive oral symptoms appearing at the last open dose resulted in a positive assessment of the challenge in 5 patients. In 8 patients the DBPCFC was prematurely discontinued after subjective symptoms on 3 consecutive dosages. In patients with birch pollinosis, the DBPCFC was performed outside the pollen season. A total of 24 adults declined to undergo a DBPCFC. All had a convincing history of an objective phenotype, with generalized urticaria, dyspnea, or wheezing.

Classification of symptoms during DBPCFCs

Symptoms recorded during DBPCFCs were used to stratify patients into severe and nonsevere phenotypes. The approach chosen for this stratification was to grade subjective symptoms (oral allergy, nausea, abdominal

discomfort, and throat tightness) as nonsevere and objective symptoms (generalized urticaria or angioedema, emesis, diarrhea, rhinoconjunctivitis, hoarseness, stridor, and wheezing) as severe. Although it could be argued that, for example, throat tightness might be seen as severe and rhinoconjunctivitis as mild, we have chosen this approach because it is least influenced by patient and observer bias, and overall, the division of subjective versus objective has acceptable overlap with the division of mild versus severe. Sensitized patients with negative DBPCFC results were stratified together with the mild subjective group. This clinical stratification was finished before serology data were generated or any further analysis was done to ensure blinding.

IgE measurements

IgE to hazelnut extract, rCor a 1, rCor a 8, nCor a 9, rCor a 14, birch pollen extract, rBet v 1, rBet v 2, and CCDs was determined by using ImmunoCAP (Thermo Fisher Scientific, Uppsala, Sweden). Experimental tests were used for nCor a 9 and rCor a 14. nCor a 9 was purified from defatted hazelnut extract by means of ion exchange chromatography and reversed-phase chromatography. rCor a 14¹⁸ was expressed as a hexahistidine-tagged protein and purified by means of immobilized metal ion affinity chromatography and ion exchange chromatography. A hazelnut extract depleted of Cor a 1 was prepared by means of immunoadsorption with an affinity column carrying an mAb directed against Cor a 1. IgE test results of 0.35 kU_A/L or greater were considered positive. Values of less than 0.35 kU_A/L were analyzed statistically as 0.34 kU_A/L, and values of greater than 100 kU_A/L were analyzed statistically as 101 kU_A/L.

Data analysis

The prevalence of asthma, atopic dermatitis, and birch pollinosis was compared between children and adults and between patients with a hazelnut allergy with objective and no or subjective symptoms by using the χ^2 test. The Mann-Whitney *U* test (continuous IgE values) and χ^2 test (binary, ≥ 0.35 kU_A/L) were used to compare IgE levels between patients with hazelnut allergy with objective and no or subjective symptoms. The diagnostic value of IgE levels (continuous values) for discrimination between hazelnut allergy with objective and no or subjective symptoms was determined by calculating the area under the curve (AUC) of the receiver operating characteristic. Sensitivity and specificity in the discrimination between hazelnut allergy with objective and no or subjective symptoms were calculated for different cutoff values of IgE and different combinations of IgE. A *P* value of less than .05 was considered significant. All analyses were performed with SPSS software (version 20.0; SPSS, Chicago, Ill).

RESULTS**Patients' characteristics**

Clinical characteristics of children and adults are shown in Table I. There was a male predominance in children and a female predominance in adults. Asthma was more common among adults with hazelnut allergy with objective than those with no or subjective symptoms (*P* = .03) and more common among children with no or subjective symptoms than adults with no or subjective symptoms (*P* = .04). Atopic dermatitis was more common in children than in adults (*P* < .001), whereas birch pollinosis was less common in children than in adults (*P* = .01) and less common in children with objective than those with no or subjective symptoms (*P* = .01). Clinical characteristics were comparable within the group of children and adults with hazelnut allergy with no or subjective symptoms, whereas age was significantly higher in children and adults with subjective symptoms compared with those with no hazelnut allergy (data not shown). Eliciting doses during DBPCFCs were lower in children and adults with hazelnut allergy with objective symptoms than those with no or subjective symptoms (*P* = .02).

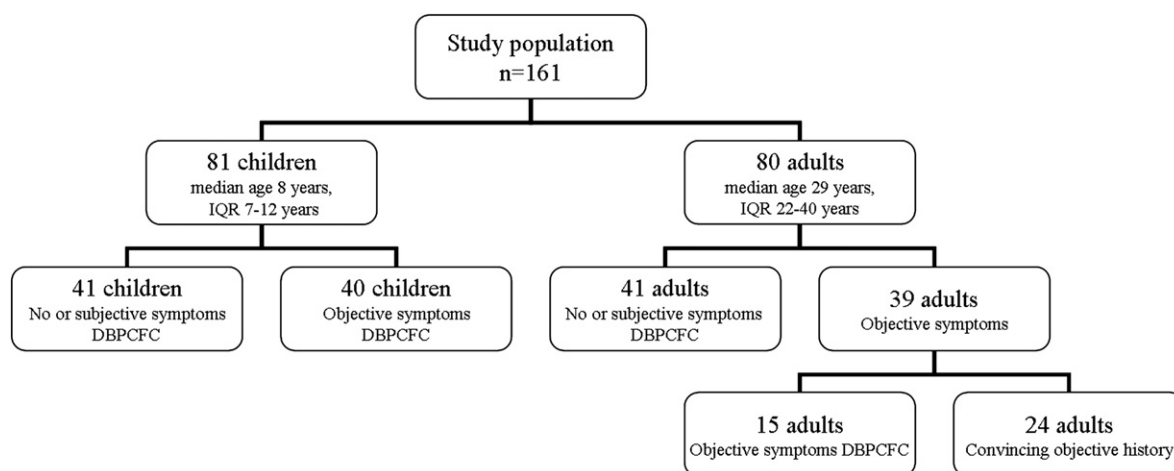


FIG 1. Composition of the study population and different groups with regard to age (children vs adults), severity (hazelnut allergy with no/subjective vs objective symptoms), and judgment of severity (DBPCFC result vs convincing history).

TABLE I. Patients' characteristics

	Children					Adults				
	No/subjective (n = 41)		Objective (n = 40)		P value	No/subjective (n = 41)		Objective (n = 39)		P value
	Median	IQR	Median	IQR		Median	IQR	Median	IQR	
Age (y)	9	8-12	7	5-11	.002	31	24-45	27	22-37	.19
	No.	Percent	No.	Percent		No.	Percent	No.	Percent	
Sex (male)	28	68	27	68	1.00	15	37	8	21	.14
Asthma	27	68	21	53	.11	18	45	29	73	.03
Atopic dermatitis	41	100	38	95	.24	30	73	29*	74*	.80
Pollinosis	33	81	24	60	.05	37	90	29	74	.08
Birch pollinosis	30	73	18	45	.01	35	85	28	72	.18
Grass pollinosis	22	54	15	38	.18	27	66	20	51	.26

IQR, Interquartile range.

*Missing data in 1 adult.

TABLE II. Number of children and adults sensitized (IgE ≥ 0.35 kU_A/L) to hazelnut extract, rCor a 1, rCor a 8, nCor a 9, rCor a 14, birch pollen extract, rBet v 1, rBet v 2, and CCDs determined by using ImmunoCAP

IgE ≥ 0.35 kU _A /L	Children					Adults				
	No/subjective (n = 41)		Objective (n = 40)		P value	No/subjective (n = 41)		Objective (n = 39)		P value
	No.	Percent	No.	Percent		No.	Percent	No.	Percent	
Hazelnut extract	41	100	40	100		41	100	39	100	
rCor a 1	37	90	28	70	.03	39	95	35	90	.43
rCor a 8	2	5	3	8	.68	1	2	2	5	.61
nCor a 9	8	20	33	83	<.001	2	5	14	36	.001
rCor a 14	10	24	28	70	<.001	2	5	15	39	<.001
Birch pollen extract	39	95	29	73	.007	40	98	35	90	.20
rBet v 1	37	90	29	73	.05	39	95	35	90	.43
rBet v 2	10	24	4	10	.14	5	12	4	10	1.00
CCD	2	5	2	5	1.00	2	5	6	15	.15

Sensitization to birch pollen

Birch pollen sensitization occurred in the majority of all children and adults. In line with the clinical history, birch pollen sensitization was less common among children with hazelnut allergy with objective than those with no or subjective symptoms

($P = .007$). All children (100%) and the majority of adults (97%) with subjective hazelnut allergy were sensitized to birch pollen. The median IgE level to birch pollen was lower in the sera of children with hazelnut allergy with objective than those with no or subjective symptoms (median, 8.78 kU_A/L vs 18.2 kU_A/L;

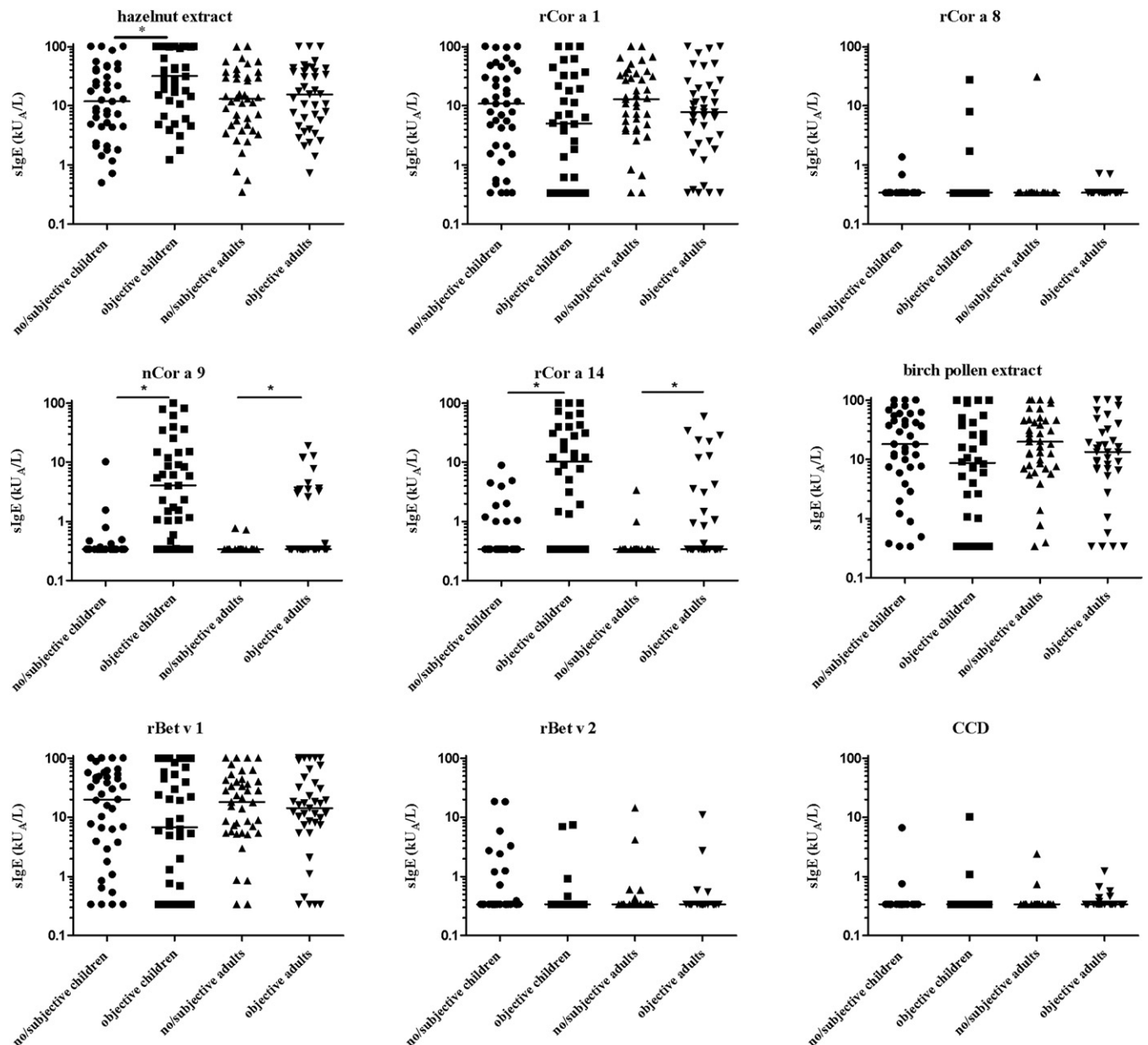


FIG 2. Levels of sensitization to hazelnut extract, rCor a 1, rCor a 8, nCor a 9, rCor a 14, birch pollen extract, rBet v 1, rBet v 2, and CCDs in children and adults with hazelnut allergy with objective and those with no or subjective symptoms. Horizontal bars indicate median levels, and IgE values of less than 0.35 kU_A/L were set to 0.34 kU_A/L.

$P = .10$). No such difference was observed among the adults. Similar to birch pollen, sensitization to rBet v 1 was frequently detected in children and adults but less common among children with hazelnut allergy with objective than those with no or subjective symptoms ($P = .05$). Again, no such difference was observed among the adults. The level of sensitization to rBet v 1 was comparable between patients with hazelnut allergy with objective and those with no or subjective symptoms both in children and adults. Sensitization to rBet v 2 and CCDs was present in a minority of children and adults and did not significantly differ between those with hazelnut allergy with objective and those with no or subjective symptoms (Table II and Fig 2).

Sensitization to hazelnut extract and components

The level of IgE to hazelnut extract was significantly higher in children with hazelnut allergy with objective than those with no or subjective symptoms (median, 31.8 kU_A/L vs 11.9 kU_A/L; $P = .006$), whereas a corresponding difference was not observed among these phenotypes in adults (Fig 2).

The majority of patients (70% to 95%) in all 4 groups were sensitized to rCor a 1, and the levels of IgE strongly correlated with those against rBet v 1 ($r = 0.96$, $P < .001$). Absence of sensitization to rCor a 1 was most frequently observed among children (12/16) and adults (4/6) with hazelnut allergy with objective symptoms. The level of IgE to rCor a 1 was comparable between patients with hazelnut allergy with objective and those

TABLE III. Sensitivity and specificity for different cutoff levels of IgE to nCor a 9 and rCor a 14 to identify children and adults with hazelnut allergy with objective symptoms among our selected population

	Children		Adults	
	Sensitivity	Specificity	Sensitivity	Specificity
IgE to nCor a 9 (kU _A /L) ≥				
0.35	83	80	36	95
1	75	95	33	100
5	48	98	13	100
IgE to rCor a 14 (kU _A /L) ≥				
0.35	70	76	38	95
1	70	76	31	98
5	60	98	18	100

with no or subjective symptoms. In 13% (5/40) of the children and 49% (19/39) of the adults with hazelnut allergy with objective symptoms, sensitization to no other allergen than rCor a 1 was observed. The level of IgE to rCor a 1 was not significantly different between these 24 patients and other patients with sensitizations to rCor a 1.

Sensitization to rCor a 8 was only detected in 8 patients distributed across all 4 study groups and mostly at moderate levels.

Sensitization to nCor a 9 and rCor a 14 was significantly more common in patients with hazelnut allergy with objective than those with no or subjective symptoms (Table II). Levels of IgE to nCor a 9 and rCor a 14 were significantly higher in children with hazelnut allergy with objective (median, 4.07 and 10.3 kU_A/L, respectively) than those with no or subjective symptoms (median, <0.35 kU_A/L for both allergens). Also among the adults, patients with hazelnut allergy with objective symptoms had significantly higher levels of IgE to nCor a 9 and rCor a 14 than those with no or subjective symptoms (Fig 2 and Table II). Both children and adults without birch pollinosis had significantly higher IgE levels to nCor a 9 than those with birch pollinosis. Levels of IgE to rCor a 14 were also higher in adults without than those with a history of allergic symptoms to birch pollinosis, and no such difference was observed for IgE to rCor a 14 among children (data not shown).

Identification of patients with hazelnut allergy with objective symptoms

In addition, the capacity of these components to identify patients with hazelnut allergy with objective symptoms was evaluated by using receiver operating characteristic curves and AUCs. Hazelnut extract, rCor a 1, and rCor a 8 did not display a useful level of discrimination between the severity groups in this study (Figs 1 and 2). In contrast, the prevalence and magnitude of sensitization to nCor a 9 and rCor a 14 differed markedly between these groups. The sensitivity and specificity with regard to discrimination between patients with objective symptoms and those with no or subjective symptoms, as a function of different cutoff levels, were calculated, as shown in Tables III and IV.

IgE to nCor a 9 had better capacity than IgE to rCor a 14 to identify children with objective symptoms in DBPCFCs based on AUC values of 0.87 and 0.80, respectively (Fig 3). This resulted in a sensitivity of 75% and a specificity of 95% for IgE levels to nCor

a 9 of 1 kU_A/L or greater (Tables III and IV). Sensitization to either nCor a 9 at 1 kU_A/L or greater or rCor a 14 at 5 kU_A/L or greater had a sensitivity of 83% and a specificity of 93%. In the group with no or subjective symptoms, only 3 (7%) of 41 children (2 with negative challenge results and 1 with subjective symptoms) would have received an incorrect diagnosis of potentially objective hazelnut allergy by using these cutoff values.

In adults IgE to either nCor a 9 or rCor a 14 had a similar capacity to identify adults with hazelnut allergy with objective symptoms based on AUC values of 0.66 and 0.67, respectively. The adults who had not undergone DBPCFCs did not display higher or more frequent sensitization to nCor a 9 or rCor a 14 than those who had been challenged and had objective symptoms. In the total adult population sensitization to nCor a 9 at 1 kU_A/L or greater was very specific (100%) for objective symptoms but accounted for only 33% of adults with hazelnut allergy with objective symptoms. Sensitization to rCor a 14 was specific (95%) for objective symptoms at a cutoff value of 0.35 kU_A/L or greater and accounted for 38% of adults with objective symptoms. The combination of an IgE level to either nCor a 9 of 1 kU_A/L or greater or rCor a 14 of 1 kU_A/L or greater in adults accounted for 44% of the adults with hazelnut allergy with objective symptoms, with a specificity of 98%. One of the 41 adults (2%, with a hazelnut allergy with subjective symptoms) would have received an incorrect diagnosis of potentially objective hazelnut allergy by using these cutoff values. For both children and adults, logistic regression analyses showed no significant additional value by combining IgE to the different components in the prediction of hazelnut allergy with objective symptoms, indicating that a prediction model was not necessary (data not shown).

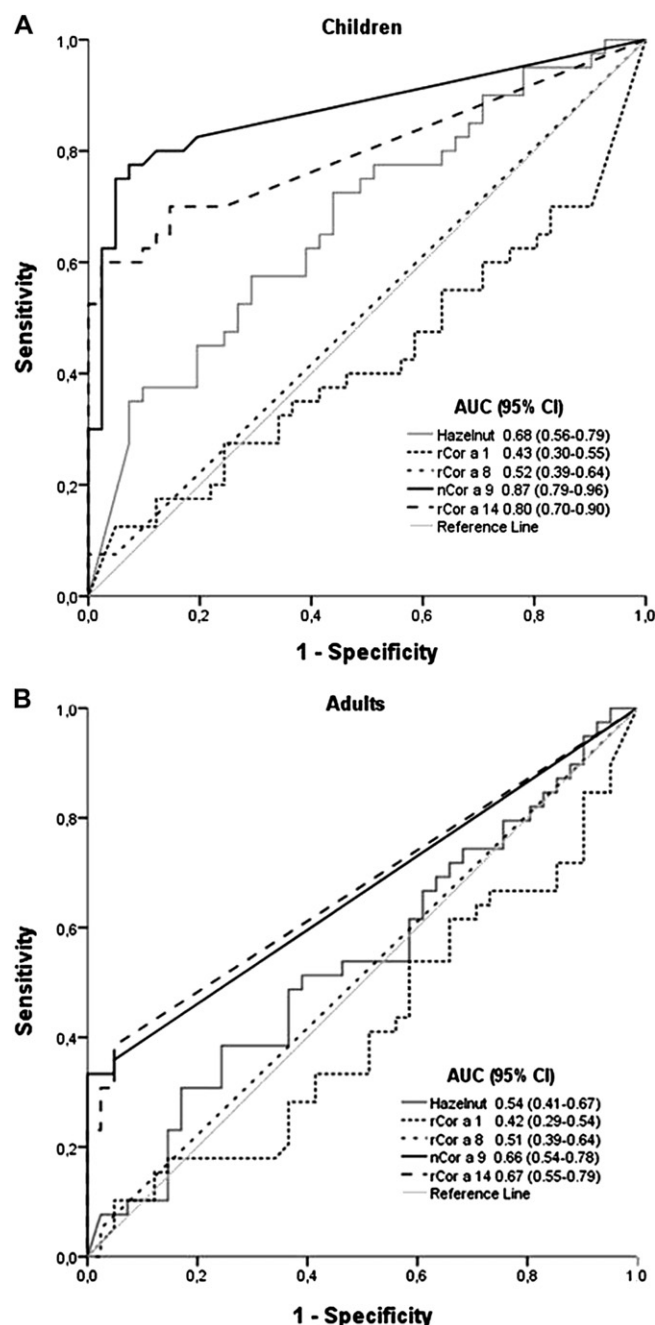
DISCUSSION

Routine diagnostic tools are not good predictors of severe hazelnut allergy.^{7,19} The present study has demonstrated that sensitization to nCor a 9, rCor a 14, or both is highly specific for hazelnut allergy with objective symptoms among Dutch hazelnut-sensitized children and adults. Sensitization to nCor a 9 or rCor a 14 was demonstrated in the majority of children and almost half of the adults with hazelnut allergy with objective symptoms. In children sensitization to either nCor a 9 at 1 kU_A/L or greater or rCor a 14 at 5 kU_A/L or greater had a specificity of 93% and accounted for 83% of children with hazelnut allergy with objective symptoms. In adults the combination of IgE levels to either nCor a 9 of 1 kU_A/L or greater or rCor a 14 of 1 kU_A/L or greater had a specificity of 98% and accounted for 44% of the adults with hazelnut allergy with objective symptoms. For patients with negative results for nCor a 9 or rCor a 14 or with IgE levels of less than these cutoff values, a DBPCFC would still be required to exclude or demonstrate hazelnut allergy with objective symptoms. The outcome of this study has relevance to real-life hazelnut exposure because there was a significant correlation between symptoms during DBPCFCs and reported real-life symptoms ($r = 0.412$ and $P < .001$ in children and $r = 0.533$ and $P < .001$ in adults).

Previous studies already showed recognition of Cor a 9 on Western blotting in the sera of 86% of patients with severe hazelnut allergy in the United States.¹² Also, in Belgium sensitization to Cor a 9 was detected in a high proportion of children with severe hazelnut allergy. Furthermore, the prevalence of sensitization to Cor a 9 was found to differ between age groups:

TABLE IV. Sensitivity and specificity for the combination of different cutoff levels of IgE to nCor a 9 and rCor a 14 to identify children and adults with hazelnut allergy with objective symptoms among our selected population

IgE nCor a 9 (kU _A /L) ≥	And/or	IgE rCor a 14 (kU _A /L) ≥	Children		Adults	
			Sensitivity	Specificity	Sensitivity	Specificity
0.35	And	0.35	65	98	23	100
0.35	Or	0.35	88	59	51	90
1	Or	1	88	71	44	98
1	Or	5	83	93	36	100

**FIG 3.** Receiver operating characteristic curves for IgE to hazelnut extract and individual hazelnut components in regard to discrimination between hazelnut allergy with objective and no or subjective symptoms in children (A) and adults (B) with sensitization to hazelnut.

65% of preschool children, 50% of schoolchildren, and 17% of adults with a severe reaction to hazelnut recognized nCor a 9.⁵ This age-related effect was also observed in our study: 83% of the children and 36% of the adults with hazelnut allergy with objective symptoms were sensitized to nCor a 9. Sensitization to rCor a 14 also showed an age-related prevalence: 70% of children and 39% of adults with hazelnut allergy with objective symptoms were rCor a 14 sensitized. An explanation might be that birch pollen-related hazelnut sensitization becomes a more frequent cause of hazelnut allergy through adolescence and early adulthood, resulting in a lower proportion of subjects with primary sensitization to hazelnut (hallmarked by sensitization to abundant storage proteins). Sensitization to rCor a 14 has previously been described in a pilot study of 11 Dutch patients, of whom 8 reported a history suggestive of hazelnut allergy, including 5 with reported severe symptoms.¹⁸

Cor a 9 and Cor a 14 are both abundant seed storage proteins in hazelnut. Seed storage proteins have been identified as major allergens in several other tree nuts and peanut. Sensitization to the peanut 2S albumin Ara h 2 has been shown to be a good predictor of peanut allergy.²⁰⁻²² The clinical relevance of sensitization to seed storage proteins in patients with tree nut allergy has not been extensively investigated. Our study convincingly shows that seed storage proteins are also relevant in patients with hazelnut allergy, especially in the objective phenotype. A good correlation between the sum of IgE levels to nCor a 9 and rCor a 14 compared with a Cor a 1-depleted hazelnut extract ($r = 0.96$ and $P < .001$ in children and $r = 0.78$ and $P < .001$ in adults, data not shown) suggests limited importance of other water-soluble hazelnut components, such as Cor a 11,^{23,24} which belongs to a family of abundant and water-soluble seed storage proteins that are well represented in natural allergen extracts. Still, validation of the data with respect to nCor a 9 and rCor a 14 in other populations is important, not least because sensitization profiles can vary between different geographic locations.¹⁰

The majority of children and adults without birch pollinosis had hazelnut allergy with objective symptoms and higher levels of IgE to nCor a 9 and rCor a 14 than those with birch pollinosis. However, 13% of children and 49% of adults with hazelnut allergy with objective symptoms were sensitized to rCor a 1 without sensitization to rCor a 8, nCor a 9, and rCor a 14 compared with 14% in a previous study (in which IgE levels to rCor a 14 were not determined).⁵ A close correlation between IgE levels to hazelnut extract and rCor a 1 in these patients ($r = 0.98$ and $P < .001$, data not shown) and lack of IgE binding to a Cor a 1-depleted hazelnut extract (data not shown) suggest a lack of sensitization to water-extractable hazelnut components other than rCor a 1. Thus the objective symptoms in these patients might be caused by either Cor a 1, analogous to the role of Gly m 4 in soy allergy,²⁵ or by 1 or several components that are not

present in aqueous hazelnut extract, such as oleosins.¹⁷ Further studies are required to resolve this issue. Hazelnut-tolerant patients could not be serologically distinguished from those with hazelnut allergy with subjective symptoms in this study because no significant difference was detected in the level of sensitization to hazelnut extract or components between these groups among either children or adults (data not shown).

Sensitization to rCor a 8 is important in the Mediterranean area because it is reported to occur in 50% to 71% of patients with severe hazelnut allergy from Spain.^{10,11} However, sensitization to rCor a 8 was hardly detected in our Dutch population and was not specific for hazelnut allergy with objective symptoms. This is in contrast to a previous study by our group with nCor a 8.²⁶ Such a discrepancy could be due to structural differences between natural and recombinant Cor a 8 or incomplete removal of other allergens from the natural protein preparation. Recent experiments, revealing that nCor a 8 preparations indeed contain trace amounts of Cor a 14 and possibly other seed storage proteins (data not shown), provide a plausible explanation for the observed discrepancy.

In summary, even though the proportion of patients with hazelnut allergy with no or subjective versus objective symptoms in our study is not fully representative of that of patients appearing at an allergy clinic, a majority of the children and almost half of the adults with hazelnut allergy with objective symptoms could be identified with these tools, and the majority of patients sensitized to Cor a 9, Cor a 14, or both reacted with objective symptoms to hazelnut. Although food challenges will still be necessary in many cases, we conclude that determination of IgE levels to nCor a 9 and rCor a 14 has the potential to strongly improve the diagnostic workup in cases of suspected severe hazelnut allergy.

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Key messages

- Sensitization to Cor a 9 and Cor a 14 is highly specific for patients with objective symptoms on DBPCFCs as a marker for a more severe hazelnut allergic phenotype.
- The majority of children and almost half of the adults with hazelnut allergy with objective symptoms were sensitized to Cor a 9 and Cor a 14.

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