

Component-resolved diagnostics for the evaluation of peanut allergy in a low-prevalence area

Narissara Suratannon¹, Jarungchit Ngamphaiboon¹, Jongkonnee Wongpiyabovorn², Panitchaya Puripokai² & Pantipa Chatchatee¹

¹Department of Pediatrics, Chulalongkorn University, Bangkok, Thailand; ²Department of Microbiology, Chulalongkorn University, Bangkok, Thailand

To cite this article: Suratannon N, Ngamphaiboon J, Wongpiyabovorn J, Puripokai P, Chatchatee P. Component-resolved diagnostics for the evaluation of peanut allergy in a low-prevalence area. *Pediatr Allergy Immunol* 2013; **24**: 665–670.

Keywords

Asia; component-resolved diagnostic; peanut allergy

Correspondence

Pantipa Chatchatee, MD, Department of Pediatrics, Chulalongkorn University, Rama IV Road, Prathumwan, Bangkok 10330, Thailand

Tel.: +66 22564933

Fax: +66 22564933

E-mail: pantipa1111@yahoo.com

Accepted for publication 31 July 2013

DOI:10.1111/pai.12125

Abstract

Background: Major allergenic components of peanut from distinct geographical regions are widely dispersed. Most of the diagnostic studies are from countries with a high prevalence. There have been only few reports of allergen component sensitizations from countries with a low prevalence of peanut allergy. We aimed to investigate roles of component-resolved diagnostic (CRD) to differentiate peanut allergy and peanut tolerance in the Asian population from a country with low prevalence of peanut allergy.

Methods: Participants with peanut sensitization were enrolled. Clinical reactions were determined. Skin prick test (SPT) and specific IgE (sIgE) to peanut and related allergen components were performed.

Results: Forty subjects with peanut sensitization were included. The mean wheal sizes of SPT and peanut sIgE were not good predictors for differentiating peanut reactions. SIgE to rAra h 2 was more often found in patients with peanut allergy and anaphylaxis. sIgE to rAra h 9 was also more frequent in the peanut-allergic group but not related to severe reactions. In the peanut-tolerant group, despite positive SPT and/or sIgE to peanut, 90% had negative sIgE to rAra h 2 and rAra h 9. Combining rAra h 2 and rAra h 9 resulted in high performance of the test with sensitivity, specificity, positive predictive value, and negative predictive value of 84%, 90%, 0.89, and 0.86, respectively. The ratio between rAra h 2 sIgE to peanut sIgE of 0.6 can be helpful in predicting patients who will develop severe reaction. SIgE to cross-reactive carbohydrate determinants (CCD) was exclusively found in the peanut-tolerant group (33.3% vs. 0%, $p = 0.012$).

Conclusions: Our study identifies three allergen components: rAra h 2, rAra h 9, and CCD as important components in the diagnosis of peanut allergy in an Asian country with low prevalence. The ratio between rAra h 2 sIgE to peanut sIgE can be used for predicting patients who will develop anaphylaxis.

Prevalence of peanut allergy varies in different geographical regions (1–3). A recent report from a UK cohort study estimated the prevalence of peanut allergy to be at 1.8% (4). In contrast, the prevalence is perceived to be low in Asian countries as shown in a large population-based survey from

Singapore and the Philippines (5). The percentages of peanut-allergic patients in Singapore aged between 4 and 6 and 14 and 16 years were 0.64% and 0.47%, accordingly. For 14 to 16-year-old Filipinos, the prevalence was 0.43%. In Thailand, there is very low prevalence of peanut allergy. Peanut allergy was not reported in Thai children from 6 months to 7 years of age in the prevalence surveys of food allergy (6, 7). The underlying factors contributing to the differences in prevalence between western and eastern countries are not well established.

Over recent years, many reports have demonstrated the role of component-resolved diagnostic (CRD) as an additional tool

Abbreviations

CRD, component-resolved diagnostic; SPT, skin prick test; sIgE, specific IgE; CCD, cross-reactive carbohydrate determinants; LTP, lipid transfer protein; PR-10, pathogen-related protein 10; DBPCFC, double-blind placebo-controlled food challenge.

for discriminating truly allergic from asymptomatic sensitized patients (8–10).

For diagnosis of peanut allergy, the results of SPT against peanut extract or measurement of peanut sIgE have a low positive predictive value because many sensitized individuals are tolerant to peanut (8). In an unselected population-based cohort in Manchester, 12% of the 8-year-old children were found to be sensitized to peanut but only one-fifth revealed to have clinical peanut allergy determined by oral food challenge (10). This is because the current tests, using crude natural extracts which contain both allergenic and cross-reactive molecules, can produce false positive results due to many reasons; cross sensitization from plants or fruits containing pathogen-related protein 10 (PR-10) (11), prolifins (12), cross-reactive carbohydrate determinants (CCD) (13), or lipid transfer protein (LTP) (14). Some hypotheses proposed that cross reactivity can be generated between seed storage proteins (9). For this reason, CRD which is a technique using pure selective allergens, opens a new era of diagnostic testing by providing the ability to discriminate primary sensitization from cross-reactive allergens (8, 15). From previous studies in the United States and some European countries, Ara h 1, Ara h 2, and Ara h 3 are considered major peanut allergens (15). rAra h 2 has been highlighted as a marker of genuine sensitization, persistent allergy, and also the marker of severe reactions (10, 16–18). However, major allergenic components from distinct geographical regions are widely dispersed. A report from Vereda et al. (19) clearly emphasized this issue. Patients who lived in the United States were more likely to have sIgE against rAra h 2 (90%), whereas serum from Spanish and Swedish patients tended to react with rAra h 9 (60%) and rAra h 8 (65.7%), respectively.

There is little information regarding CRD testing and clinical characteristics of peanut allergy in Asian populations. A study from Singapore performing in-house ELISAs in peanut-sensitized populations revealed the prevalence of Ara h 1/Ara h 2 and Ara h 3 sensitization of 87.1% and 54.8% accordingly (20). No clinical correlation relating to the components patients react to was discussed. Another study from Japan revealed that positive Ara h 2 sIgE can help differentiate peanut-allergic patients from the peanut-tolerant children with a sensitivity of 88% and a specificity of 84%. The combination of Ara h 1, Ara h 2, and Ara h 3 sIgE increased the specificity of the test to 94% (18). The purpose of our study was to investigate sensitization patterns against peanut allergen components and the role of *in vivo* and *in vitro* diagnostic tests for peanut allergy especially sIgE to allergen components to differentiate true peanut allergy and peanut tolerance in a country with low prevalence of peanut allergy such as Thailand.

Patients and methods

Study design and participants

This study was a cross-sectional study. Forty participants with peanut sensitization had undergone evaluation at King Chulalongkorn Memorial Hospital, one of the country's

referral centers, between January 2008 and December 2010. Clinical information was collected either through questionnaire or chart review. Written informed consent was obtained. The patients were classified into two categories: peanut allergy and peanut tolerance. Peanut allergy was documented when a patient reported a convincing history of acute reaction after peanut ingestion and detectable peanut sIgE or had a positive food challenge result. Peanut tolerance was defined as a subject who had detectable peanut sIgE but tolerated peanut on at least one occasion or had a negative food challenge. Peanut anaphylaxis was diagnosed when two or more of the following symptoms occurred rapidly after exposure to peanut: involvement of the skin or mucosal tissue, respiratory compromise, cardiovascular compromise, persistent gastrointestinal symptoms *or* when the subject with known peanut allergy had hypotension after peanut exposure (21). The study was approved by the Hospital Institutional Review Board.

Sensitization measurements

Skin prick test (SPT) was performed on the volar side of the forearm or upper back with a lancet according to the guideline (22). A positive SPT was defined as a mean wheal diameter ≥ 3 mm. SIgE against peanut and allergen components including rAra h 1, rAra h 2, rAra h 3, rAra h 9, nGly m 5, nGly m 6, rBet v 1, rBet v 2, rPhl p 12, and CCD were performed by the ImmunoCAP System FEIA (ThermoFisher scientific, Uppsala, Sweden) according to the manufacturer's instructions. A lower and upper detection limit was 0.35 and 100 kUA/l accordingly.

Oral food challenges

The challenge was conducted in the hospital under supervision of an allergist. Informed consent was obtained. IV lock was placed. The challenge was performed in seven steps. Starting dosage was given at 0.1 g and increased every 15 min until a total dose of 10 g was reached or if the patient reported symptoms. If a certain diagnosis could not be made or the patient primarily presented with subjective symptoms, the double-blind placebo-controlled food challenge (DBPCFC) was performed. If no reaction was reported, eating peanut at home was encouraged to confirm tolerance.

Statistical analysis

Variables were displayed in medians and interquartile ranges (IQR). Mann–Whitney *U* test was used for comparing the median of SPT wheal sizes, sIgE to peanut, and allergen components. Sensitization rates against allergen components were compared using the chi-squared test and Fisher exact tests. Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were reported with 95% confidence interval. Best cutoff for the ratio between rAra h 2/peanut sIgE and area under the curve at the cut point of SPT (8 mm), peanut sIgE (>14 kUA/l), and sIgE to rAra h 2 (>0.35 kUA/l) were calculated using ROC curves. Analysis was performed using Stata 11.0 (StataCorp, Texas, USA).

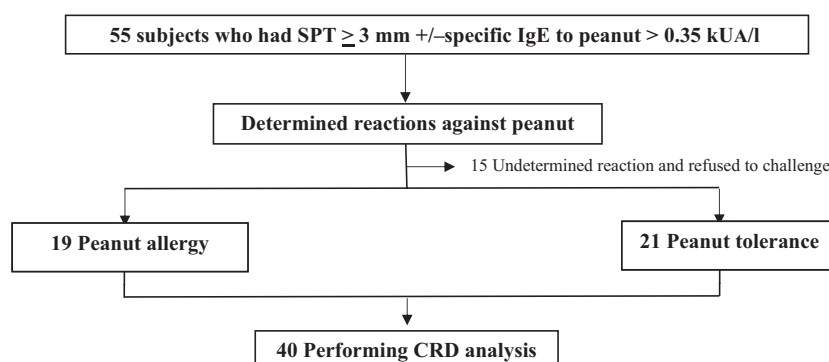


Figure 1 Flow of participants.

Table 1 Demographic characteristics

Characteristic	Peanut allergy (n = 19)	Peanut tolerance (n = 21)
Current age		
Median month (IQR)	107.0 (59.8, 213.0)	100 (52.8, 166.5)
Age of first peanut reaction		
Median month (IQR)	24.0 (12.0, 74.5)	—
Male gender, n (%)	14 (73.7)	11 (52.4)
Born or lived in	5 (26.3)	2 (9.5)
United States, n (%)		
Reported anaphylactic reaction against peanut, n (%)	5 (26.3)	—
Parent-reported soy allergy, n (%)	4 (21.1)	0 (0)
Ever having food allergies, n (%)	12 (63.2)	15 (78.9)
Respiratory allergies, n (%)	11 (57.9)	12 (60)
Grass sensitization, n (%)	9 (64.3)	11 (68.8)

IQR, interquartile range.

Table 2 Comparison of laboratory values between peanut-allergic groups and peanut-tolerant groups

Laboratory values	Peanut allergy	Peanut tolerance	p-Value
Peanut SPT wheal size (mm)			
Median (IQR)	7.0 (6.3, 11.5)	4.8 (3.0, 6.5)	0.003*
Peanut-specific IgE (kUA/l)			
Median (IQR)	6.3 (1.5, 12.5)	1.1 (0, 5.4)	0.040*
rAha h2-specific IgE (kUA/l)			
Median (IQR)	2.1 (0.0, 12.4)	0 (0.0, 0.0)	<0.001*

*Mann–Whitney *U* Test, *p* < 0.05.

SPT, skin prick test; IQR, interquartile range.

Results

Participants' characteristics

Flow of participants is described in Fig. 1. Nineteen peanut-allergic subjects and 21 peanut-tolerant subjects were identified. Baseline characteristics of the patients were shown in

Table 3 Univariate analysis of variables evaluating possible predictors for the diagnosis of peanut allergy

Predictors	OR	95% CI	p-Value
Peanut SPT MWD, ≥8 vs. <8 mm	3.00	0.63, 14.23	0.167
Peanut-specific IgE, ≥14 vs. <14 kUA/l	1.13	0.24, 5.34	0.874
Specific IgE to rAra h2, ≥0.35 vs. <0.35 kUA/l	43.33	4.66, 402.67	<0.001*

*Mann–Whitney *U* Test, *p* < 0.05.

SPT, skin prick test; MWD, mean wheal diameter.

Table 1. Median age of onset of peanut allergy was 2 years. Anaphylaxis was reported in 25% of the patients. Respiratory allergies and grass sensitization did not differ between groups. About one-fourth of peanut-allergic patients were born in the US or lived there in their early life. Interestingly, 20% of peanut-allergic patients had parent-reported soy allergies, while there was none in peanut-tolerant patients.

SPT and sIgE to peanut

The median wheal size of SPT and sIgE levels to peanut was significantly greater in the peanut-allergic group (*p* = 0.003 and *p* = 0.04 accordingly). However, when we looked into the individual's reactions and immunological profiles, some peanut-allergic patients had smaller SPT wheal sizes and lower sIgE than asymptomatic sensitized subjects. When we performed the univariate analysis, our results clearly demonstrated that the previous proposed cutoff values of SPT wheal size (≥8 mm) (23) and peanut sIgE levels (>14 kUA/l) (24) were not good predictors in the diagnosis of peanut allergy in our populations (Table 3). The sensitivity and specificity of such values in the diagnosis of peanut allergy were as follows: SPT wheal size ≥8 mm (46.2%, 77.8%) and peanut sIgE levels > 14 kUA/l (21.1%, 81%). Combining SPT wheal size and peanut sIgE did not increase the performance of the test (Table 4).

sIgE to allergen components

The sensitization rates against allergen components are shown in the bar chart in Fig. 2. The highest prevalence in the

Table 4 Performance characteristics of different methods for diagnosis of peanut allergy

Marker	% positive in peanut allergic group	AUC	Sensitivity (95%CI)	Specificity (95%CI)	PPV	NPV
SPT > 8 mm	46.2	0.62	46.2 (19.2, 74.9)	77.8 (52.4, 93.6)	0.60	0.67
Peanut sIgE > 14 kUA/l	21.1	0.51	21.1 (6.1, 45.6)	81.0 (58.1, 94.6)	0.50	0.53
sIgE to rAra h2 $\geq$ 0.35 kUA/l	68.4	0.82	68.4 (43.4, 87.4)	95.2 (76.2, 99.9)	0.92	0.77
sIgE to rAra h1 or rAra h2 $\geq$ 0.35 kUA/l	68.4	–	68.4 (43.4, 87.4)	90.5 (69.6, 98.8)	0.87	0.76
sIgE to rAra h1 or rAra h2 or rAra h3 > 0.35 kUA/l	68.4	–	68.4 (43.4, 87.4)	85.7 (63.7, 97)	0.81	0.75
sIgE to rAra h2 or rAra h9 > 0.35 kUA/l	84.2	–	84.2 (60.4, 96.6)	90.5 (69.6, 98.8)	0.89	0.86
SPT > 8 mm or peanut sIgE > 14 kUA/l	47.4	–	47.4 (27.3, 68.3)	71.4 (50.1, 86.2)	0.60	0.60
Peanut sIgE > 14 kUA/l or rAra h2 > 0.35 kUA/l	68.4	–	68.4 (43.4, 87.4)	76.2 (52.8, 91.8)	0.72	0.73

AUC, area under the curve; PPV, positive predictive value; NPV, negative predictive value.

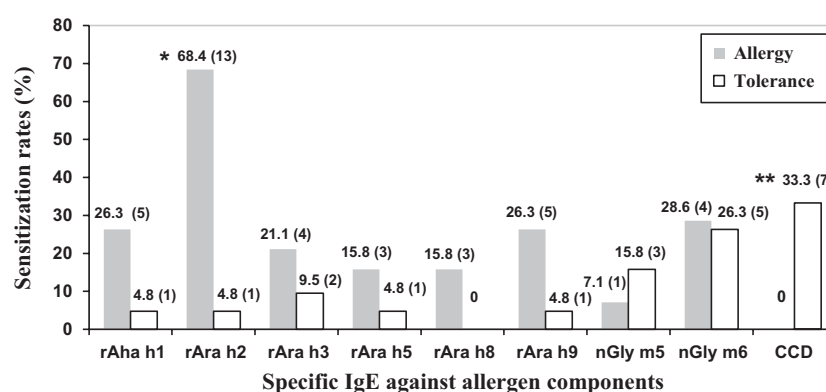


Figure 2 Bar chart demonstrated percentages of positive IgE results against allergen components in peanut-allergic group and peanut-tolerant group. * $p < 0.001$; ** $p = 0.012$, Fisher's exact test was used in comparing positive results of specific IgE to allergen components between peanut-allergic and peanut-tolerant group. Numbers above bar charts and in brackets indicate percent and absolute numbers of subjects with positive results, respectively.

peanut-allergic group was sIgE against rAra h 2 (68.4%), followed by rAra h 1 and rAra h 9 at the same percentages (26.3%). rAra h 5 sIgE was found in 16% and 5% of peanut-allergic patients and peanut-tolerant subjects accordingly. Sixteen percent of peanut-allergic patients also had rAra h 8 sensitization. The percentages and the median levels of rAra h 2 sIgE were significantly higher in peanut-allergic patients than in the peanut-tolerant group (68.4% vs. 4.8%, $p < 0.001$ and 2.12 (0, 12.4) kUA/l vs. 0 (0) kUA/l, $p < 0.001$ accordingly). Univariate analysis showed rAra h 2 sensitized patients to have a high risk of having peanut allergy (OR = 43.33, $p < 0.001$) (Table 3). The sensitivity and specificity of sIgE to rAra h 2 were higher than SPT and sIgE to peanut (sensitivity 68.4% vs. 46.2% vs. 21.1%, specificity 95.2% vs. 77.8% vs. 81%, respectively) at the cutoff value 0.35 with area under the ROC curve 0.82. The positive predictive value (PPV) was 0.92.

All patients with anaphylaxis had IgE binding to rAra h 2 compared with 57% of non-anaphylaxis. Interestingly, while SPT wheal size and levels of peanut sIgE and rAra h 2 sIgE were not significantly different between the two groups, the ratio of rAra h 2 sIgE to peanut sIgE was significantly higher in

patients with anaphylaxis with the median of 1.0 (0.67, 1.68) vs. 0 (0, 0.78), $p = 0.02$. The cutoff value of 0.6 can be used to differentiate between the patients with anaphylaxis and non-anaphylaxis with sensitivity 100% and specificity 71% with area under the ROC curve 0.86.

Focusing on rAra h 1, rAra h 3, and rAra h 9 in peanut-allergic group, all rAra h 1 and rAra h 3 sensitized patients had positive rAra h 2 specific IgE. Patients who were sensitized to only rAra h 1 or rAra h 3 but not rAra h 2 did not have clinical reactions. Adding rAra h 1 and rAra h 3 into the panels did not increase the sensitivity and specificity of CRD tests. In contrast, 15% of peanut allergy who had positive IgE to rAra h 9 did not have sIgE to rAra h 2. This specific group elicited only mild reactions against peanut. Combination of rAra h 2 and rAra h 9 increased sensitivity of the test from 68.4% to 84.2%. The negative predictive value (NPV) also increased from 0.77 to 0.86. Combining rAra h 2 sIgE and sIgE to peanut did not increase performance of the test (Table 4).

Specific IgE to CCD was exclusively found in the peanut-tolerant group. No peanut-allergic patients had positive titers to this component (33.3% vs. 0%, $p = 0.012$). About fifteen

and twenty-five percent of peanut-tolerant subjects were sensitized to nGly m 5 and nGly m 6 even though they did not report any allergic symptoms to soybean. One asymptomatic peanut-sensitized patient had high levels of rAra h9 sIgE (>100 kUA/l). Ninety percent of peanut-tolerant patients had negative results of sIgE to rAra h 2 and rAra h 9.

Discussion

Our study confirmed that rAra h 2 is the most important peanut allergen since it was found with the highest prevalence in peanut-allergic group and related with anaphylaxis. The results are in line with the findings from the United States, Europe and some Asian countries (18, 19, 25-27). Additionally, twenty percent of our peanut-tolerant subjects had peanut sIgE > 14 kUA/l. This group was likely to be diagnosed as peanut-allergic based on previously published serologic criteria (23). When CRD assays were performed, all patients in these groups had negative rAra h 2 sIgE demonstrating the impact of CRD testing in differentiating peanut-tolerant from peanut-allergic subjects. SIgE to rAra h 2, therefore, should be selected as the first marker to be tested. When sIgE to rAra h 2 is positive, peanut allergy is highly suggestive and testing for other allergens is not necessary.

However, the sensitivity of the rAra h 2 sIgE varies between countries. In our population, rAra h 2 sIgE has a sensitivity of 68.4%. This may not be enough for making the diagnosis of peanut allergy. Additional tests for other peanut allergens should be considered when rAra h 2 sIgE was negative.

The non-specific lipid transfer protein (nsLTPs) Ara h 9 has been mentioned in the study of peanut-allergic populations from Mediterranean studies (27-29). In the subjects who react to Ara h 9, the cross-reactivity with Pru p 3 from peach which is also prevalence in the area should be taken into consideration (27, 29). Although it is not considered as a major allergen in general populations (27), this allergen can elicit a variety of symptoms ranging from asymptomatic to systemic reactions (28). The reason is that LTP is a partial stable allergen and not degraded by heat or digestive enzyme (30). LTP sensitization in Asian populations was reported in a study from Japan demonstrated that 15% of the peanut-allergic children were sensitized to Ara h 9 with or without having Ara h 2 sensitization. No correlation with clinical reactivity was mentioned in the study (18). In our study, 15% of the peanut-allergic patients were exclusively sensitized to rAra h 9 without having IgE binding to rAra h 2. Seventy-five percent of LTP sensitized patients developed reactions when ingesting peanut. One subject in peanut-tolerant group did not develop allergic reactions to peanut even with a high level of rAra h 9 sIgE (> 100 kUA/l). Our result showed that the severity of reactions does not correlate with the level of rAra h 9 sIgE. Testing for rAra h 9 sIgE in our populations should be beneficial in terms of increasing sensitivity and NPV of the test when rAra h 2 sIgE was negative. Further research is needed to identify the primary sensitizer of LTP in our populations.

CCDs are carbohydrate moieties of glycoproteins that induce the production of highly cross-reactive IgE (30) and

are usually not associated with symptoms. Among 19 peanut-tolerant subjects with negative rAra h 2 sIgE and rAra h 9 sIgE, sIgE to CCD was found in 37% while none of the peanut-allergic patients was positive. Therefore, in our population, sIgE to CCDs was a good indicator of peanut tolerance and if positive, could reduce the need for food challenge.

Despite being a seed storage protein, sIgE to rAra h 1 does not seem to add to the diagnostic accuracy as all patients in our study who reacted to rAra h 1 also had positive results to rAra h 2. This issue was demonstrated when we explored the details of individuals' reactions. Patients who were sensitized to only rAra h 1 or rAra h 3 but not rAra h 2 did not have clinical reactions. This is different from previous studies showing that the combination of rAra h 1, rAra h 2 and rAra h 3 sensitizations increased the performance of CRD testing (18, 25) and correlated with more severe reaction (8, 31). Most of them had positive IgE against nGly m 5 and nGly m 6. We hypothesized that positive titers to rAra h 1 and rAra h 3 are probably a cross-reaction from soybean or other allergens having similar molecular structures. This concept has been supported by an *in vitro* study carried out by Chruszcz, et al. (32) showing that beta-conglycinin Gly m 5 is the closest homolog of Ara h 1 and has a 51% amino acid sequence identity and also a similar 3D structure. Ara h 3 and soybean glycinin Gly m 6 share a 3D structure similar to that of Ara h 1 despite less sequence homology (32). Another study from Ballabio et al. demonstrated that beta-conglutinin and gamma-conglutinin play roles in the cross-reactivity between lupin and peanut proteins (33). All of these data support our hypothesis that sensitizations to the seed storage proteins may not be genuine.

We also explored the association of soy allergy and the role of soy allergen components sensitizations. Twenty percent of peanut-allergic patients had the history of parent-reported soy allergy. There was no soy allergy in the peanut-tolerant group. SIgE to nGly m 5 and nGly m 6 did not correlate with clinical allergy. All subjects with positive sIgE to nGly m 5 or nGly m 6 could tolerate soy. Our findings demonstrated that nGly m 5 and nGly m 6 may not be good indicators for soy allergy in areas where soy is widely consumed. In addition, these components may cause false-positive reactions in skin prick tests to peanut or peanut sIgE due to their sequence homology (32).

There are some limitations of this study. Due to the small sample sizes, we may not have enough power to make a significant comparison for subgroup analyses.

In conclusion, we have shown the roles of component-resolved diagnostic as a useful diagnostic tool for peanut allergy in the Asian population from a country with low prevalence. rAra h 2 is confirmed to be the most important allergen and a severity marker. rAra h 9 should be included in peanut panel testing when rAra h 2 sIgE was negative. Measurement of IgE to CCD in subjects with positive sIgE to peanut but having negative results to rAra h 2 and rAra h 9 sIgE further helps to discriminate between peanut-tolerant and peanut-allergic subjects. nGly m 5 and nGly m 6 are not the severity markers of soy allergy in our population.

Acknowledgments

We are thankful to Nirun Intarut from Data Management Center, Chulalongkorn University for performing

statistical analysis, and Professor Pakit Vichyanond from Siriraj Hospital for patient referral. We also thank Thermo Fisher Scientific for providing reagents for *in vitro* testing.

References

1. Sicherer SH, Sampson HA. Peanut allergy: emerging concepts and approaches for an apparent epidemic. *J Allergy Clin Immunol* 2007; **120**: 491–503; quiz 4–5.
2. Grundy J, Matthews S, Bateman B, Dean T, Arshad SH. Rising prevalence of allergy to peanut in 31/07/2013 children: Data from 2 sequential cohorts. *J Allergy Clin Immunol* 2002; **110**: 784–9.
3. Sicherer SH, Munoz-Furlong A, Sampson HA. Prevalence of peanut and tree nut allergy in the United States determined by means of a random digit dial telephone survey: a 5-year follow-up study. *J Allergy Clin Immunol* 2003; **112**: 1203–7.
4. Le TM, Lindner TM, Pasmans SG, et al. Reported food allergy to peanut, tree nuts and fruit: comparison of clinical manifestations, prescription of medication and impact on daily life. *Allergy* 2008; **63**: 910–6.
5. Shek LP, Cabrera-Morales EA, Soh SE, et al. A population-based questionnaire survey on the prevalence of peanut, tree nut, and shellfish allergy in 2 Asian populations. *J Allergy Clin Immunol* 2010; **126**: 324–31, 31 e1–7.
6. Lao-araya M, Trakultivakorn M. Prevalence of food allergy among preschool children in northern Thailand. *Pediatr Int* 2012; **54**: 238–43.
7. Santadusit S, Aththapaisalsarudee S, Vichyanond P. Prevalence of adverse food reactions and food allergy among Thai children. *J Med Assoc Thai* 2005; **88** (Suppl. 8): S27–32.
8. Borres MP, Ebisawa M, Eigenmann PA. Use of allergen components begins a new era in pediatric allergology. *Pediatr Allergy Immunol* 2011; **22**: 454–61.
9. Masilamani M, Commins S, Shreffler W. Determinants of food allergy. *Immunol Allergy Clin North Am* 2012; **32**: 11–33.
10. Nicolaou N, Poorafshar M, Murray C, et al. Allergy or tolerance in children sensitized to peanut: prevalence and differentiation using component-resolved diagnostics. *J Allergy Clin Immunol* 2010; **125**: 191–7 e1–13.
11. Mittag D, Akkerdaas J, Ballmer-Weber BK, et al. Ara h 8, a Bet v 1-homologous allergen from peanut, is a major allergen in patients with combined birch pollen and peanut allergy. *J Allergy Clin Immunol* 2004; **114**: 1410–7.
12. Cabanos C, Tandang-Silvas MR, Odijk V, et al. Expression, purification, cross-reactivity and homology modeling of peanut profilin. *Protein Expr Purif* 2010; **73**: 36–45.
13. Jin C, Hantusch B, Hemmer W, Stadlmann J, Altmann F. Affinity of IgE and IgG against cross-reactive carbohydrate determinants on plant and insect glycoproteins. *J Allergy Clin Immunol* 2008; **121**: 185–190 e2.
14. Shreffler WG. Microarrayed recombinant allergens for diagnostic testing. *J Allergy Clin Immunol* 2011; **127**: 843–9; quiz 50–1.
15. Lidholm J, Ballmer-Weber BK, Mari A, Vieths S. Component-resolved diagnostics in food allergy. *Curr Opin Allergy Clin Immunol* 2006; **6**: 234–40.
16. Flinterman AE, van Hoffen E, den Hartog Jager CF, et al. Children with peanut allergy recognize predominantly Ara h2 and Ara h6, which remains stable over time. *Clin Exp Allergy* 2007; **37**: 1221–8.
17. Nicolaou N, Moverare R, Sato S, Poorafshar M, Simpson A, Custovic A. Quantification of specific IgE to whole peanut extract and peanut components in prediction of peanut allergy. *J Allergy Clin Immunol* 2011; **127**: 684–5.
18. Ebisawa M, Moverare R, Sato S, Maruyama N, Borres MP, Komata T. Measurement of Ara h 1-, 2-, and 3-specific IgE antibodies is useful in diagnosis of peanut allergy in Japanese children. *Pediatr Allergy Immunol* 2012; **23**: 573–81.
19. Vereda A, van Hage M, Ahlstedt S, et al. Peanut allergy: clinical and immunologic differences among patients from 3 different geographic regions. *J Allergy Clin Immunol* 2011; **127**: 603–7.
20. Chiang WC, Pons L, Kidon MI, Liew WK, Goh A, Wesley Burks A. Serological and clinical characteristics of children with peanut sensitization in an Asian community. *Pediatr Allergy Immunol* 2010; **2**: e429–38.
21. Cianferoni A, Muraro A. Food-induced anaphylaxis. *Immunol Allergy Clin North Am* 2012; **32**: 165–95.
22. Dreborg S. The skin prick test in the diagnosis of atopic allergy. *J Am Acad Dermatol* 1989; **21** (4 Pt 2): 820–1.
23. Sporik R, Hill DJ, Hosking CS. Specificity of allergen skin testing in predicting positive open food challenges to milk, egg and peanut in children. *Clin Exp Allergy* 2000; **30**: 1540–6.
24. Sampson HA. Utility of food-specific IgE concentrations in predicting symptomatic food allergy. *J Allergy Clin Immunol* 2001; **107**: 891–6.
25. Eller E, Bindslev-Jensen C. Clinical value of component-resolved diagnostics in peanut-allergic patients. *Allergy* 2013; **68**: 190–4.
26. Klemans RJ, Otte D, Knol M, et al. The diagnostic value of specific IgE to Ara h 2 to predict peanut allergy in children is comparable to a validated and updated diagnostic prediction model. *J Allergy Clin Immunol* 2013; **131**: 157–63.
27. Pedrosa M, Boyano-Martinez T, Garcia-Ara MC, Caballero T, Quirce S. Peanut seed storage proteins are responsible for clinical reactivity in Spanish peanut-allergic children. *Pediatric allergy and immunology: official publication of the European Society of Pediatric Allergy and Immunology* 2012; **23**: 654–9.
28. Krause S, Reese G, Randow S, et al. Lipid transfer protein (Ara h 9) as a new peanut allergen relevant for a Mediterranean allergic population. *J Allergy Clin Immunol* 2009; **124**: 771–8 e5.
29. Lauer I, Dueringer N, Pokoj S, et al. The non-specific lipid transfer protein, Ara h 9, is an important allergen in peanut. *Clin Exp Allergy* 2009; **39**: 1427–37.
30. Aalberse RC, Koshte V, Clemens JG. Immunoglobulin E antibodies that crossreact with vegetable foods, pollen, and Hymenoptera venom. *J Allergy Clin Immunol* 1981; **68**: 356–64.
31. Astier C, Morisset M, Roitel O, et al. Predictive value of skin prick tests using recombinant allergens for diagnosis of peanut allergy. *J Allergy Clin Immunol* 2006; **118**: 250–6.
32. Chruszcz M, Maleki SJ, Majorek KA, et al. Structural and immunologic characterization of Ara h 1, a major peanut allergen. *J Biol Chem* 2011; **286**: 39318–27.
33. Ballabio C, Penas E, Uberti F, et al. Characterization of the sensitization profile to lupin in peanut-allergic children and assessment of cross-reactivity risk. *Pediatr Allergy Immunol* 2013; **24**: 270–5.