Diagnostic accuracy of patch test in children with food allergy

WILEY
Online Proofing System Instructions

The Wiley Online Proofing System allows authors and proof reviewers to review PDF proofs, mark corrections, respond to queries, upload replacement figures, and submit these changes directly from the PDF proof from the locally saved file or while viewing it in your web browser.

1. For the best experience reviewing your proof in the Wiley Online Proofing System please ensure you are connected to the internet. This will allow the PDF proof to connect to the central Wiley Online Proofing System server. If you are connected to the Wiley Online Proofing System server you should see the icon with a green check mark above in the yellow banner.


Connected


Disconnected

2. Please review the article proof on the following pages and mark any corrections, changes, and query responses using the Annotation Tools outlined on the next 2 pages.

3. To save your proof corrections, click the "Publish Comments" button appearing above in the yellow banner. Publishing your comments saves your corrections to the Wiley Online Proofing System server. Corrections don't have to be marked in one sitting, you can publish corrections and log back in at a later time to add more before you click the "Complete Proof Review" button below.

4. If you need to supply additional or replacement files bigger than 5 Megabytes (MB) do not attach them directly to the PDF Proof, please click the "Upload Files" button to upload files:

Click Here

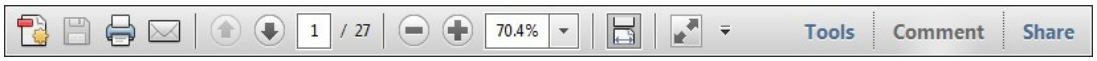
5. When your proof review is complete and you are ready to submit corrections to the publisher, please click the "Complete Proof Review" button below:

Click Here

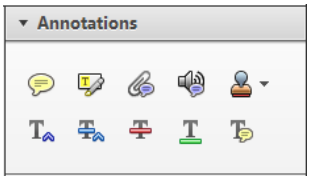
IMPORTANT: Do not click the "Complete Proof Review" button without replying to all author queries found on the last page of your proof. Incomplete proof reviews will cause a delay in publication.

IMPORTANT: Once you click "Complete Proof Review" you will not be able to publish further corrections.

Once you have Acrobat Reader open on your computer, click on the Comment tab at the right of the toolbar:



This will open up a panel down the right side of the document. The majority of tools you will use for annotating your proof will be in the Annotations section, pictured opposite. We've picked out some of these tools below:

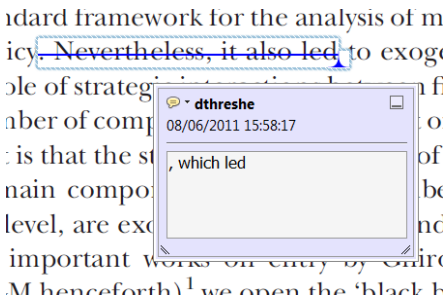


1. Replace (Ins) Tool – for replacing text.

Strikes a line through text and opens up a text box where replacement text can be entered.

How to use it

- Highlight a word or sentence.
- Click on the Replace (Ins) icon in the Annotations section.
- Type the replacement text into the blue box that appears.

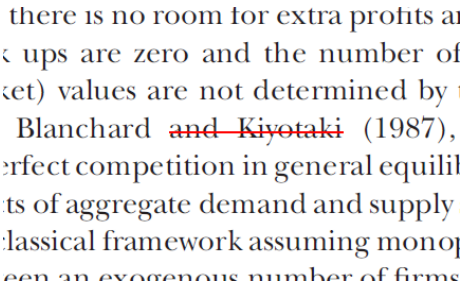


2. Strikethrough (Del) Tool – for deleting text.

Strikes a red line through text that is to be deleted.

How to use it

- Highlight a word or sentence.
- Click on the Strikethrough (Del) icon in the Annotations section.

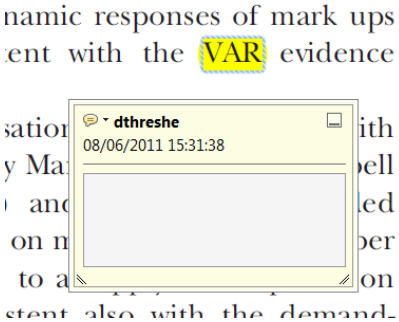


3. Add note to text Tool – for highlighting a section to be changed to bold or italic.

Highlights text in yellow and opens up a text box where comments can be entered.

How to use it

- Highlight the relevant section of text.
- Click on the Add note to text icon in the Annotations section.
- Type instruction on what should be changed regarding the text into the yellow box that appears.

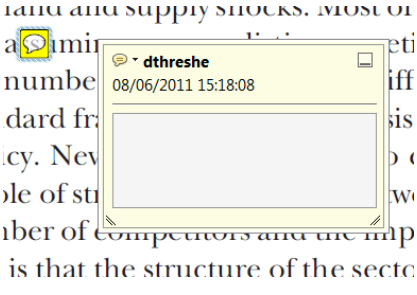


4. Add sticky note Tool – for making notes at specific points in the text.

Marks a point in the proof where a comment needs to be highlighted.

How to use it

- Click on the Add sticky note icon in the Annotations section.
- Click at the point in the proof where the comment should be inserted.
- Type the comment into the yellow box that appears.

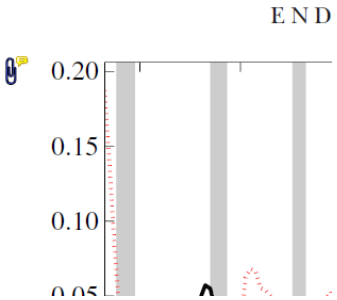


5. Attach File Tool – for inserting large amounts of text or replacement figures.

Inserts an icon linking to the attached file in the appropriate place in the text.

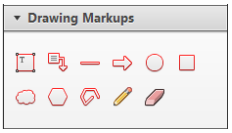
How to use it

- Click on the Attach File icon in the Annotations section.
- Click on the proof to where you'd like the attached file to be linked.
- Select the file to be attached from your computer or network.
- Select the colour and type of icon that will appear in the proof. Click OK.



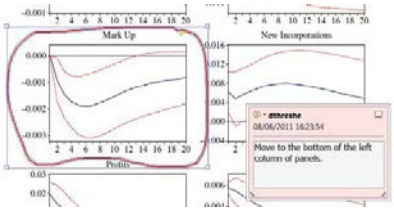
6. Drawing Markups Tools – for drawing shapes, lines and freeform annotations on proofs and commenting on these marks.

Allows shapes, lines and freeform annotations to be drawn on proofs and for comment to be made on these marks.



How to use it

- Click on one of the shapes in the Drawing Markups section.
- Click on the proof at the relevant point and draw the selected shape with the cursor.
- To add a comment to the drawn shape, move the cursor over the shape until an arrowhead appears.
- Double click on the shape and type any text in the red box that appears.



ORIGINAL ARTICLE

Diagnostic accuracy of patch test in children with food allergy

Sule Caglayan Sozmen[1], Carlotta Povesi Dascola[2], Edoardo Gioia[2], Carla Mastrorilli[2], Laura Rizzuti[2] & Carlo Caffarelli[2]

[1]Pediatric Department, Division of Allergy and Clinical Immunology, Faculty of Medicine, Dokuz Eylul University, Izmir, Turkey; [2]Pediatric Department, Department of Clinical and Experimental Medicine, Unit of Allergy and Immunology in Evolutive Age, University of Parma, Parma, Italy

To cite this article: ~~Sozmen SC, Dascola CP,~~ Gioia E, Mastrorilli C, Rizzuti L, Caffarelli C. Diagnostic accuracy of patch test in children with food allergy. *Pediatr Allergy Immunol* 2015: **00**.

Keywords
atopy patch test; skin prick test; food allergy; cow's milk allergy; egg allergy; child; oral food challenge; atopic eczema; anaphylaxis; gastrointestinal symptoms

Correspondence
Carlo Caffarelli, Clinica Pediatrica, Ospedale Maggiore, Università di Parma, Via Gramsci 14, 43126 Parma, Italy
Tel.: +39 0521 702207
Fax: +39 0521 702358
E-mail: carlo.caffarelli@unipr.it

Accepted for publication xxxx

DOI:10.1111/pai.12377

Abstract

Background: The gold standard test for confirming whether a child has clinical hypersensitivity reactions to foods is the oral food challenge. Therefore, there is increasing interest in simpler diagnostic markers of food allergy, especially in children, to avoid oral food challenge. The goal of this study was to assess the diagnostic accuracy of atopy patch test in comparison with oral food challenge.
Methods: We investigated 243 children (mean age, 51 months) referred for evaluation of suspected egg or cow's milk allergy. Skin prick test and atopy patch test were carried out, and after a 2 weeks elimination diet, oral food challenge was performed.
Results: Two hundred and forty-three children underwent OFC to the suspected food. We found clinically relevant food allergies in 40 (65%) children to egg and in 22 (35%) to cow's milk. The sensitivity of skin prick test for both milk and egg was 92%, specificity 91%, positive predictive value 35%, and negative predictive value of 93%. Sensitivity, specificity, positive predictive value, and negative predictive value of atopy patch test for both milk and egg were 21%, 73%, 20%, and 74%, respectively.
Conclusion: Our study suggests that there is insufficient evidence for the routine use of atopy patch test for the evaluation of egg and cow's milk allergy. OFC remains gold standard for the diagnosis of egg and milk allergy even in the presence of high costs in terms of both time and risks during application.

Food allergy has emerged as an increasing medical problem with main burden occurring in childhood and refers to an abnormal immunologic response directed toward food (1). Cow's milk and egg allergy are the most common food allergies in younger age group (1).

Careful diagnosis is important because overdiagnosis by parents and even medical professionals results in a restrictive and inadequate diet which can impair both growth and limit participation in social activities (2, 3). Diagnosis of food allergies in children starts with a careful dietary history to define the potential food triggers (4). Skin prick test (SPT) results and measurement of serum-specific IgE antibodies (sIgE) can be helpful in identifying offending foods (5). The oral food challenge (OFC) test is the definitive mean to ascertain clinical reactions to food (6). There is increasing interest in easier diagnostic markers of food allergy, especially in children, to avoid OFC. Atopy patch test (APT) is a useful tool for the diagnosis of contact dermatitis (7), and it has been proposed as a non-invasive test for diagnosing food allergy in children. However, in populations with delayed onset symptoms such as atopic eczema and gastrointestinal symptoms, previous studies have provided contrasting results on APT usefulness in identifying children with food allergies (8–12). Furthermore, APT has not yet been standardized for routine use in the diagnosis of food allergy (13).

The objective of the current study was to examine the diagnostic reliability of APT compared to OFC to identify individuals with allergy to cow's milk and hen's egg.

Materials and method

Children referred with a suspicion of cow's milk or egg allergies to a hospital-based outpatient center were consecutively enrolled in the study. Patients were subjected to an allergological

PAI Journal Code 12377 Manuscript No. | WILEY | Dispatch: 30.3.15 No. of pages: 7 | CE: Selin Aarthi PE: Iniya Selvi

work-up consisting of a detailed history, focusing on clinical signs of food allergy, combined with the SPT, APT, and OFC. All children followed diet without the suspected food for a period of at least 2 weeks before OFC.

Skin prick test

All children underwent SPTs with commercial extracts of egg yolk, egg white (ALK-Abello), cow's milk, alpha-lactalbumin, beta-lactoglobulin, and casein (Lofarma). Furthermore, patients were skin prick tested with 1 mg/ml histamine and control solution. The wheal reactions were read after 15 min. Tests were considered positive when the wheal size was ≥3 mm in diameter in comparison with the negative control. The parents were asked to withhold antihistamine medications at least 72 hrs before, and 2–3 weeks before the skin test and topical steroid (14).

Atopy patch test

The ATP was carried out using lyophilized food in white vaseline as excipient (Lofarma, Milano, Italy). The concentration of egg was 5%, casein 5%, and cow's milk 10%. The allergen concentration has been changed to 10% for egg and casein and to 20% for milk from January 2007. The extract was put on an 8-mm aluminum cup on adhesive tape (Finn Chambers; Epitest Ltd. Oy) that was applied to the intact skin [4] the back and covered with a hypoallergenic tape. Application sites were checked after 20 min for immediate reactions. APT was evaluated 48 hrs after application, and results were read 20 min after removal of the hypoallergenic tape with cup. The second reading was made 24 hrs after the first reading (72 hrs) in case of an unclear response (15). APT was graded as follows (13): − negative; only erythema, questionable; + erythema, infiltration; ++ erythema, few papules; +++ erythema, many or spreading papules; ++++ erythema, vesicles.

Oral food challenge

An investigation about possible intake of drugs within the days before OFC was carried out. Children had to stop antihistamines 10 days before, steroids 3 days before, theophylline, beta2-agonists, and leukotriene modifiers 24 hrs before challenge. Before OFC, each child was visited, height and weight were measured, and an intravenous access was obtained. The test was performed only when the child was asymptomatic and had completely normal physical examination. Emergency medications and equipment were immediately available for the control of adverse reactions. Challenge to egg consisted of increasing increments every 20 min of 25 mg lyophilized white egg protein, 400 mg, 800 mg, 1.6 g, 3.2 g, and 6.4 g of egg protein given as boiled egg. OFC with pasteurized cow's milk (or formula milk in infants) was performed by administering increasing doses (0.25, 0.5, 1, 3 ml, and then doubled until 100 ml) at 20-min intervals. Patients were observed for at least 2 hrs. During the OFC, children were subjected to blood pressure monitoring. Challenge was discontinued when the patients developed clinical symptoms (16, 17). Adverse reactions were considered immediate when they appeared within 2 hrs and late if after 2 hrs. When the challenge result was negative, children received the food at home for 4 days. At the end of the challenge period, children were checked for the onset of adverse reactions. When OFC resulted in subjective or unclear symptoms, a double-blind placebo-controlled OFC was performed using the same titration steps as the open ones (18). The local Ethical Committee approved the study. Parents gave their written informed consent.

Statistical analysis

Student's *t*-test and analysis of variance were used to analyze differences between groups in continuous variables. Comparison of categorical variables was made by means of chi-square test. All analyses were two-tailed. Sensitivity, specificity, and negative and positive predictive accuracy for APT and SPT results were calculated. Spearman's correlation analysis was used to evaluate correlation between OFCs and ages of patients. The statistical analysis was performed using SPSS [5] 15.0 version.

Results

Two hundred and eighty-seven children were eligible for the study. We experienced 44 dropouts. Parents did not agree to perform APTs due to the fact that it took such a long time.

Table 1 Comparison of the dropout group against participants to the study

	Dropout Group	Study Group	p value
No.	44	243	0.09
Age, mean (months)	42.8	51.6	
Male, no. (%)	23 (52)	131 (54)	0.32
Cow's milk allergy, no. (%)	23 (52)	105 (43)	0.84
Egg allergy	21 (48)	138 (57)	0.32
Skin symptoms after egg ingestion, no. (%)	33 (77)	194 (80)	0.54
Non-cutaneous symptoms after egg ingestion, no. (%)	11 (23)	49 (20)	0.54
SPT, positive, no. (%)	32 (73)	175 (72)	1.00
OFC, positive, no. (%)	17 (39) (41% to egg and 59% to milk)	62 (25) (65% to egg and 35% to milk)	0.09

Table 2 Comparison of children with positive atopy patch test (APT) against those with negative APT to cow's milk and egg

	Suspicion of egg or cow's milk allergy			Suspicion of egg allergy			Suspicion of cow's milk allergy		
	Positive APT	Negative APT	p value	Positive APT	Negative APT	p value	Positive APT	Negative APT	p value
No.	56	187		37	101		19	86	
Age, mean (months)	52.7	48.1	0.59	58.2	52.5	0.97	42.2	42.9	0.54
Male, no. (%)	32 (57)	99 (52)	0.58	18 (48)	55 (54)	0.54	14 (73)	44 (51)	0.07
SPT, positive, no. (%)	38 (67)	131 (70)	0.75	28 (75)	77 (76)	0.94	10 (52)	54 (62)	0.41
OFC, positive, no. (%)	10 (17)	52 (27)	0.13	9 (24)	31 (30)	0.46	1 (5)	21 (24)	0.07
Skin symptoms after OFC, no. (%)	7 (12)	41 (21)	0.12	6 (16)	27 (26)	0.20	1 (5)	14 (16)	0.29
Non-cutaneous symptoms after OFC, no. (%)	3 (5)	1 (5)	0.88	34 (91)	97 (96)	0.32	0 (0)	7 (8)	0.34
Gastrointestinal symptoms after OFC, no. (%)	0 (0)	4 (2)	0.57	0 (0)	2 (2)	0.38	0 (0)	2 (2)	1.00

Table 3 Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of skin prick test (SPT) and atopy patch test (APT) to cow's milk and egg, compared with OFC in 243 children with suspected cow's milk or egg allergy, in 138 patients with suspected egg allergy and in 105 patients with suspected milk allergy

	Sensitivity (CI 95%)	Specificity (CI 95%)	PPV (CI 95%)	NPV (CI 95%)
Patients with suspected cow's milk or egg allergy (n = 243)				
At least a positive APT to egg* or milk†	21 (13–64)	73 (66–79)	20 (12–33)	74 (67–80)
At least a positive SPT to egg‡ or milk§	92 (83–96)	39 (33–46)	35 (29–43)	93 (85–97)
SPT plus APT	89 (79–95)	29 (23–37)	30 (24–38)	89 (78–95)
Patients with suspected egg allergy (n = 138)				
Positive APT to egg*	24 (13–40)	72 (62–80)	24 (13–39)	72 (63–80)
Positive SPT to egg yolk or egg white	95 (85–99)	31 (24–41)	33 (26–43)	95 (84–99)
Positive SPT to egg white	91 (79–96)	32 (24–41)	33 (25–42)	91 (78–96)
Positive SPT to egg yolk	67 (53–80)	50 (41–58)	32 (23–42)	81 (71–88)
SPT‡ plus APT*	94 (85–99)	19 (7–27)	29 (20–37)	91 (85–96)
Patients with suspected cow's milk allergy (n = 105)				
At least a positive APT to milk†	15 (6–38)	75 (64–84)	15 (5–36)	76 (66–85)
At least a positive SPT to milk extracts§	86 (69–95)	51 (40–62)	39 (28–51)	91 (79–96)
Positive SPT to whole milk	75 (57–87)	69 (58–78)	47 (33–61)	88 (78–94)
Positive SPT to casein	33 (19–52)	89 (80–94)	50 (29–71)	80 (71–87)
Positive SPT to alpha-lactalbumin	55 (37–72)	74 (63–82)	43 (28–59)	82 (72–90)
Positive SPT to beta-lactoglobulin	62 (44–77)	79 (66–87)	53 (37–69)	85 (74–91)
SPT§ plus APT†	81 (74–88)	46 (37–55)	33 (24–40)	88 (80–96)

*Extracts: egg. †Extracts: cow's milk, casein. ‡Extracts: egg yolk, egg white. §Extracts: whole cow's milk, alpha-lactalbumin, beta-lactoglobulin, casein.

This is the reason why there were some dropouts. There was no significant difference between the dropout group and the study participants in clinical characteristics (Table 1). The remaining 243 children participated in the study, 151 (62%) male and 92 (38%) female, aged between 12 months and 16 years, mean age 51.6 months. One hundred and thirty-eight of them were with a clinical suspicion of egg allergy and 105 of them with a clinical suspicion of milk allergy. Patients referred the following symptoms occurred when they ingested egg or cow's milk: skin symptoms (50% eczema, 46% urticaria or angioedema, %4 rash) in 194 cases, gastrointestinal symptoms (63% vomiting, 23% proctocolitis, 9% enterocolitis, 5% diarrhea) in 22 cases, respiratory symptoms (67% bronchospasm, 33% dyspnea) in 9 cases; anaphylaxis in 10 (4%) cases, and oral allergy syndrome in 8 (3%) cases. The outcome of OFCs was positive in 62 (25%) of 243 instances; 40 of 138 to egg, and 22 of 105 to milk. There was no need to perform double-blind placebo-controlled OFC. Cutaneous reactions (eczema, urticaria, rash) occurred in 48 (78%) of positive challenges, gastrointestinal symptoms (vomiting, diarrhea, proctocolitis) in 4 (6%), oral allergy syndrome in 4 (6%), respiratory symptoms in 3 (5%), and anaphylaxis in 3

Table 4 Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of atopy patch test (APT) at different concentrations of egg, cow's milk, and casein compared with OFC

	Sensitivity (CI 95%)	Specificity (CI 95%)	PPV (CI 95%)	NPV (CI 95%)
APT†	24 (12–43)	80 (66–90)	43 (21–67)*	0.63 (50–75)*
APT‡	19 (0.9–36)	71 (63–78)	14 (0.6–27)*	0.79 (71–85)*
APT (cow's milk plus casein)†	15 (0.4–42)	80 (58–92)	33 (10–70)	0.59 (41–75)
APT (cow's milk plus casein)‡	17 (0.3–56)	74 (60–84)	0.07 (0.1.31)	0.88 (75–95)
APT (egg)†	33 (14–81)	81 (60–92)	0.50 (22–78)	0.68 (48–83)
APT (egg)‡	20 (0.7–39)	70 (59–78)	0.17 (0.7–34)	0.74 (63–83)

Concentration of egg was 5%† or 10%‡, of casein was 5%† or 10‡, of cow's milk was 10%† or 20%‡
*$p \leq 0.05$.

(5%). There was no correlation between patient's age and OFC results. We performed 243 APTs and SPTs. APT-positive results to at least one of the cow's milk allergen or egg allergens were observed in 56 (23%) children and SPT in 169 (69%) children. There was no significant difference between children with positive APT results and those with negative APT results in age, gender, positive SPT results, positive OFC outcome, and symptoms occurring during OFC (Table 2). A positive APT result to at least one cow's milk allergen (whole cow's milk, casein) or egg had lower sensitivity and PPV than specificity and NPV (Table 3). SPT revealed a higher sensitivity, PPV and NPV than APT (Table 3). The combination of SPT and APT results did not improve sensitivity, specificity, and predictive values of SPT (Table 3).

One hundred and thirty-eight patients, age range 14 months to 14 years, mean age 55 months, had a history of allergic reactions to egg, The majority of patients experienced cutaneous symptoms: 123 (49% eczema, 46% urticaria or angioedema, 5% rash). Other clinical manifestations were gastrointestinal symptoms in 7 cases (86% vomiting, 14% enterocolitis), respiratory symptoms in 4 cases (50% dyspnea, 50% bronchospasm), oral allergy syndrome in 2 cases, and anaphylaxis in 2 cases. Forty OFCs to egg had positive outcome. Skin symptoms occurred in 33 (82%) children, oral allergy syndrome in 2 (5%), gastrointestinal symptoms in 2 (5%), respiratory symptoms in 2 (5%), and anaphylaxis in 1 (3%). Thirty-seven (27%) of APT results were positive to egg. Sensitivity, specificity, and predictive values of APT results were low (Table 3). Seventy-seven (56%), 25 (18%), and 3 (2%) SPT reactions were positive for both egg yolk and egg white, only egg white, and only egg yolk, respectively. The sensitivity and the NPV of SPT to combined egg yolk and egg white were higher than those of single allergens (Table 3). SPT results showed a higher sensitivity and predictive values than APT results (Table 3). The combination of SPT and APT results did not increase sensitivity, specificity, and predictive values of SPT alone (Table 3).

One hundred and five patients, age range 12 months and 16 years, mean 47 months, had a history of allergic reaction to milk. Skin symptoms occurred in 69 (25% urticaria, 25% eczema, 6% rash) cases, gastrointestinal in 15 cases (53% vomiting, 33% proctocolitis, 7% enterocolitis, 7% diarrhea), respiratory in 7 cases (57% bronchospasm, 43% dyspnea), oral allergy syndrome in 6 cases, and anaphylaxis in 8 cases. Twenty-two OFCs to milk had a positive outcome. Skin symptoms were elicited in 15 (68%) cases, oral allergy syndrome in 2 (9%) cases, gastrointestinal symptoms in 2 (9%) cases, anaphylaxis in 2 (9%) cases, and respiratory symptoms in 1 (3%) cases. APT reactions were positive to at least one extract in 19 children. APT results were positive to whole milk in 18 (69%) children and in 8 (31%) to casein. APTs were often false positive or false negative, which resulted in low positive predictive values (Table 3). Forty-one patients had negative SPT results. There were 109 positive SPT reactions to four cow's milk allergens; 39 (36%) cow's milk, 31 (28%) beta-lactoglobulin, 27 (25%) alpha-lactalbumin, and 12 (11%) casein. SPT results revealed a higher sensitivity and NPV than APT results (Table 3). Sensitivity and NPV were higher when we used four allergens for SPT than a single allergen (Table 3). ~~The accuracy of the four allergens,~~ SPTs together with APTs, did not improve sensitivity, specificity, and predictive values of SPT alone (Table 3).

As mentioned previously, from January 2007, the allergen concentrations in APT commercial extract were increased from 5% to 10%. Before 2007, 73 APTs (37 for milk and 36 for egg) were performed and 12 (16%), 7 for egg and 5 for milk, were positive. After 2007, 170 (68 for milk and 102 for egg) APTs were performed and 44 (26%), 14 for milk and 30 for egg, were positive ($p > 0.05$).

When we compared data of patch test results before and after 2007, there was a statistically significant difference between low and high concentrations used for APT only in NPV and PPV ($p < 0.05$) (Table 4).

Discussion

We found that APT to egg and cow's milk was not useful for diagnosing clinical hypersensitivity in children. Our results show low specificity, sensitivity, PPV, and NPV both to cow's milk and egg. Moreover, we did not demonstrate APTs results improve diagnostic accuracy of SPT to egg and cow's milk allergy.

There were conflicting results in the previous literature regarding the use of APT for the diagnosis of food allergies (Table 5).

Table 5 Studies on diagnostic accuracy of APT to cow's milk and/or egg compared with OFC results in studies with at least 50 patients in the last 10 years

Study (year)	Study population	~~Food Chamber~~	~~APT~~ ~~Egg~~		~~Cow's milk~~	
Breuer et al. (2004)	41 out of 64 children with AE, aged 1–10 years	- Fresh cow's milk, hen's egg powder - 12 mm	PPV:30 (Results of late eczematous reactions (n = 22))		PPV:32 (Results of late eczematous reactions (n = 22))	
Osterballe et al. (2004)	22 children with suspected cow's milk and egg allergy of a cohort of 396 children, aged 3 years.	- Fresh food - 8 mm	SE:40% SP:99% PPV:57% NPV:89%		SE:0% SP:99% PPV:0% NPV:99%	
Mehl et al. (2006)	437 children (90% with AE), aged 3 months to 14 years	- Fresh food - 12 mm	SE:41% SP:95% PPV:86% NPV:85%		SE:41% SP:87% PPV:86% NPV:43%	
Canani et al. (2007)	60 children with G-I symptoms, aged 3–48 months	- Fresh food and commercial extracts - 12 mm	Fresh food SE:84% SP:100% PPV:100% NPV:75%	Commercial extract SE:5% SP:100% PPV:100% NPV:33%	Fresh food SE:64% SP:95% PPV:95% NPV:67%	Commercial extract SE:6% SP:95% PPV:66% NPV:43%
Devillers et al. (2009)	135 children with AE, aged 0–3 years	- Fresh cow's milk and boiled egg - 12 mm	Odd's ratio:9.61 (0.99–99.52)		Odd's ratio:1.05 (0.02–11.60)	
Chung et al. (2010)	101 children with AE, under the age of 6 years	- Fresh cow's milk and boiled egg - 8 mm	SE:50% SP:91% PPV:57% NPV:89%		SE:67% SP:92% PPV:50% NPV:86%	
Canani et al. (2011)	119 children with G-I symptoms, aged 3–48 months	- Fresh food -12 mm			SE:66% SP:84% PPV:78% NPV:74%	
Costa et al. (2011)	192 children with G-I symptoms, aged 1–5 years	- Fresh food - Filter paper of 1 cm^2 in area			SE:25% SP:82% PPV:46% NPV:64%	
Nocerino et al. (2013)	172 children with non IgE mediated G-I symptoms, aged 2–12 months	-Fresh food -12 mm			SE:67% SP:88% PPV:82% NPV:76%	
Mowszet et al. (2014)	61 children with G-I symptoms, aged 3–6 months	-Fresh food -8 mm			SE21% SP.:91% PPV:80% NPV:39%	
Yang et al. (2014)	150 children wih atopic eczema, aged <2 years	-Cow's milk powder and fresh egg -12-mm	Result of (+) APT SE:53% SP:91% PPV:89%		Result of (+) APT SE:35% SP:88% PPV:77%	

AE, atopic eczema; G-I, gastrointestinal; SP, specificity; SE, sensitivity, PPV, positive predictive value; NPV, negative predictive value.

In agreement with previous studies (8–12, 19–23), we found that sensitivity to egg and cow's milk was low. On the other hand, our results showed remarkably lower specificity (73%) for APT in comparison with the previous studies (8, 9, 11, 19, 20, 23). We found that PPV of patch test to egg and cow's milk was low. This is compatible with many studies (9–11, 19, 20, 22, 23) with the exception of Canani's study (8). Canani et al. (8) found that PPV of patch test to fresh egg and cow's milk was over 90%. Differently from us, they selected only patients with gastrointestinal symptoms and used 12-mm aluminum

cups with fresh food. The study of Niggemann et al. (24) showed that 12-mm cups should be used for APT and demonstrated lower PPV to cow's milk with 6-mm aluminum cups. These can be an explanation for the different PPV results for APT. In contrast with their results, Canani et al. (21) in a second study found lower PPV to fresh cow's milk when they included 119 patients rather than 60 in the same age group. In accordance with previous studies (8–11, 20–22), we showed that NPV to egg and cow's milk was low. However, Osterballe (19) showed that NPV for fresh cow's milk was 99% in 22 children with possible food allergies.

Several further explanations may be offered for differences in our results and those of previous studies. Diagnostic accuracy of a given test depends on frequency of the disease in the studied population. So, the reliability of APT may vary in different populations. Moreover, our population consisted of children with suspected food allergies. Previous studies were conducted in children with atopic eczema (9, 12, 20, 23, 25) or gastrointestinal symptoms (8, 10, 17, 21, 22). Another explanation for our different findings may be that we use commercial extracts rather than fresh food. Finally, the age of children included in former studies differs from the age of our population. However, our results showed there was no association between age of the patient and OFC outcome, and we did not demonstrate any difference between patients with and without positive APT results in age and clinical findings (Table 2).

After epicutaneous application of allergens, APTs elicit a T-cell-mediated responses. Immunologic examination of biopsies from APT lesions showed initially a Th2 cytokine pattern, and after 48 hrs, it showed a Th1 pattern like in chronic lesions of AD (26). Circulant Th1 cells are involved in the IgE-mediated reaction to allergens. However, the cutaneous Th1 cells differ from those in other organs (27, 28). This may explain why we found that APT to egg and cow's milk had a low diagnostic accuracy.

This study has several strengths. Trained staff following standardized procedures performed APTs and OFCs. Another strength is that the range of data allowed investigation for the effects of SPTs on the reliability of APT results. Our study may have several limitations. The trial was conducted in children referred to a tertiary allergy clinic because of possible allergy. To avoid selection bias, we should have examined each child in a large unselected population. This is unfeasible in practice. The diagnosis was not based on double-blind placebo-controlled OFC that is the 'gold standard' for diagnosing food allergy. We choose to perform open OFC as it is commonly used in daily practice in childhood. The procedure of APT to food allergens lacks of standardization in food preparation. It is usually proposed the use of fresh foods (15). We used commercial extracts to better standardize the material, enhance stability, and reduce the influence of Staphylococcus aureus-derived enterotoxins on patch test results (29). We believe that the use of a commercial extract may have enhanced the reproducibility of the outcome of the test.

We found that SPT to cow's milk had low sensitivity, specificity, and PPV. The NPV for SPT to whole cow's milk and cow's milk proteins was high. This concurs with the findings of Calvani et al. (27). We found an high sensitivity and NPV (95%) for hen's egg, and this was in line with previous investigations (30). Although SPT has high sensitivity and high NPV, its low specificity and poor PPV may result in mislabeling some patients as having food allergies. In agreement with previous studies (8, 9), our results showed that the combination of SPT and APT results did not improve the overall diagnostic accuracy.

In conclusion, performing the OFC to confirm the diagnosis of egg and milk allergies is necessary. The APT is not practical and accessible in daily clinical practice. The results of our study suggest that APT does not have an additional value in predicting outcomes of OFC.

Acknowledgments

This work was completed as part of 2014 European Academy of Allergy and Clinical Immunology (EAACI) Clinical Fellowship Award.

References

1. Nwaru BI, Hickstein L, Panesar SS, et al. Prevalence of common food allergies in Europe: a systematic review and meta-analysis. *Allergy* 2014: **69**: 992–1007.
2. Noimark L, Cox HE. Nutritional problems related to food allergy in childhood. *Pediatr Allergy Immunol* 2008: **19**: 188–95.
3. Giovannini M, D'Auria E, Caffarelli C, et al. Nutritional management and follow up of infants and children with food allergy: Italian Society of Pediatric Nutrition/Italian Society of Pediatric Allergy and Immunology Task Force Position Statement. *Ital J Pediatr* 2014: **40**: 1.
4. NIAHD-Sponsored Expert Panel, Boyce JA, Assa'ad A, et al. Guidelines for the diagnosis and management of food allergy in the United States: report of the NIAID-sponsored expert panel. *J Allergy Clin Immunol* 2010; **126**: S1–58.
5. Caffarelli C, Dondi A, Povesi Dascola C, et al. Skin prick test to foods in childhood atopic eczema: pros and cons. *Ital J Pediatr* 2013; **39**: 48.
6. Burks AW, Tang M, Sicherer S, et al. ICON: food allergy. *J Allergy Clin Immunol* 2012: **129**: 906–20.
7. Darsow U, Wollenberg A, Simon D, et al. ETFAD/EADV eczema task force 2009 position paper on diagnosis and treatment of atopic dermatitis. *J Eur Acad Dermatol Venereol* 2010: **24**: 317–28.
8. Canani RB, Ruotolo S, Auricchio L, et al. Diagnostic accuracy of the atopy patch test in children with food allergy-related gastrointestinal symptoms. *Allergy* 2007: **62**: 738–43.
9. Chung BY, Kim HO, Park CW, Lee CH. Diagnostic usefulness of the serum-specific IgE, the skin prick test and the atopy patch test compared with that of the oral food challenge test. *Ann Dermatol* 2010: **22**: 404–11.
10. Costa AJ, Sarinho ES, Motta ME, Gomes PN, de Oliveira de Melo SM, da Silva GA. Allergy to cow's milk proteins: what contribution does hypersensitivity in skin tests have to this diagnosis? *Pediatr Allergy Immunol* 2011; **22**: e133–8.
11. Mowszet K, Matusiewicz K, Iwanczak B. Value of the atopy patch test in the

diagnosis of food allergy in children with gastrointestinal symptoms. *Adv Clin Exp Med* 2014: **23**: 403–9.
12. Breuer K, Heratizadeh A, Wulf A, et al. Late eczematous reactions to food in children with atopic dermatitis. *Clin Exp Allergy* 2004: **34**: 817–24.
13. Turjanmaa K, Darsow U, Niggemann B, Rance F, Vanto T, Werfel T. EAACI/GA2LEN position paper: present status of the atopy patch test. *Allergy* 2006: **61**: 1377–84.
14. Muraro A, Werfel T, Hoffmann-Sommergruber K, et al. EAACI food allergy and anaphylaxis guidelines: diagnosis and management of food allergy. *Allergy* 2014: **69**: 1008–25.
15. Niggemann B, Reibel S, Wahn U. The atopy patch test (APT)– a useful tool for the diagnosis of food allergy in children with atopic dermatitis. *Allergy* 2000: **55**: 281–5.
16. Caffarelli C, Rico S, Rinaldi L, Povesi Dascola C, Terzi C, Bernasconi S. Blood pressure monitoring in children undergoing food challenge: association with anaphylaxis. *Ann Allergy Asthma Immunol* 2012; **108**: 285–6.
17. Perry TT, Matsui EC, Conover-Walker MK, Wood RA. Risk of oral food challenges. *J Allergy Clin Immunol* 2004: **114**: 1164–8.
18. Nowak-Wegrzyn A, Assa'ad AH, Bahna SL, et al. Work Group report: oral food challenge testing. *J Allergy Clin Immunol* 2009; **123**: S365–83.
19. Osterballe M, Andersen KE, Bindslev-Jensen C. The diagnostic accuracy of the atopy patch test in diagnosing hypersensitivity to cow's milk and hen's egg in unselected children with and without atopic dermatitis. *J Am Acad Dermatol* 2004: **51**: 556–62.
20. Mehl A, Rolinck-Werninghaus C, Staden U, et al. The atopy patch test in the diagnostic workup of suspected food-related symptoms in children. *J Allergy Clin Immunol* 2006: **118**: 923–9.
21. Canani RB, Buongiovanni A, Nocerino R, Cosenza L, Troncone R. Toward a standardized reading of the atopy patch test in children with suspected cow's milk allergy-related gastrointestinal symptoms. *Allergy* 2011: **66**: 1499–500.
22. Nocerino R, Granata V, Di Costanzo M, et al. Atopy patch tests are useful to predict oral tolerance in children with gastrointestinal symptoms related to non-IgE-mediated cow's milk allergy. *Allergy* 2013: **68**: 246–8.
23. Yang H, Xiao YZ, Luo XY, Tan Q, Wang H. Diagnostic accuracy of atopy patch tests for food allergy in children with atopic dermatitis aged less than two years. *Allergol Immunopathol (Madr)* 2014: **42**: 22–8.
24. Niggemann B, Ziegert M, Reibel S. Importance of chamber size for the outcome of atopy patch testing in children with atopic dermatitis and food allergy. *J Allergy Clin Immunol* 2002: **110**: 515–6.
25. Devillers AC, de Waard-van der Spek FB, Mulder PG, Oranje AP. Delayed- and immediate-type reactions in the atopy patch test with food allergens in young children with atopic dermatitis. *Pediatr Allergy Immunol* 2009; **20**: 53–8.
26. Kerschenlohr K, Decard S, Przybilla B, Wollenberg A. Atopy patch test reactions show a rapid influx of inflammatory dendritic epidermal cells in patients with extrinsic atopic dermatitis and patients with intrinsic atopic dermatitis. *J Allergy Clin Immunol* 2003: **111**: 869–74.
27. Picker LJ, Treer JR, Ferguson-Darnell B, Collins PA, Bergstresser PR, Terstappen LW. Control of lymphocyte recirculation in man. II. Differential regulation of the cutaneous lymphocyte-associated antigen, a tissue-selective homing receptor for skin-homing T cells. *J Immunol* 1993: **150**: 1122–36.
28. Kabelitz D, Marischen L, Oberg HH, Holtmeier W, Wesch D. Epithelial defence by gamma delta T cells. *Int Arch Allergy Immunol* 2005: **137**: 73–81.
29. Langer K, Breuer K, Kapp A, Werfel T. Staphylococcus aureus-derived enterotoxins enhance house dust mite-induced patch test reactions in atopic dermatitis. *Exp Dermatol* 2007: **16**: 124–9.
30. Verstege A, Mehl A, Rolinck-Werninghaus C, et al. The predictive value of the skin prick test weal size for the outcome of oral food challenges. *Clin Exp Allergy* 2005: **35**: 1220–6.

Author Query Form

Journal: PAI
Article: 12377

Dear Author,
During the copy-editing of your paper, the following queries arose. Please respond to these by marking up your proofs with the necessary changes/additions. Please write your answers on the query sheet if there is insufficient space on the page proofs. Please write clearly and follow the conventions shown on the attached corrections sheet. If returning the proof by fax do not write too close to the paper's edge. Please remember that illegible mark-ups may delay publication.
Many thanks for your assistance.

Query reference	Query	Remarks
1	**WILEY: Please supply date of acceptance.**	
2	**AUTHOR: Please confirm that given names (red) and surnames/family names (green) have been identified correctly.**	
3	**AUTHOR: Please check that authors and their affiliations are correct.**	
4	**AUTHOR: Please give address information for Epitest Ltd. Oy: town, state (if applicable), and country.**	
5	**AUTHOR: Please give address information for SPSS: town, state (if applicable), and country.**	
6	**AUTHOR: Please check whether the footnotes "*, †, ‡, §" are correctly matching with their citations in table 3.**	
7	**AUTHOR: Kindly check and approve the edits made in the sentence "The accuracy. . .of SPT alone."**	
8	**AUTHOR: Please check whether the footnotes "*, †, ‡" are correctly matching with their citations in table 4.**	
9	**AUTHOR: Please check the layout of Table 5.**	
10	**AUTHOR: Kindly check and approve the edits made in the sentence "Immunologic examination. . .lesions of AD."**	