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RESEARCH LETTER

WILEY

Oral food challenge with raw egg: Sixty-minute- compared with 30-minute dosing interval

To the Editor,

Allergies to egg and milk are the most common food allergies in European children. The prevalence of IgE-mediated allergy to egg in 2-year-olds is about 1.6% and even higher in children with atopic dermatitis.

An oral food challenge (OFC) is the gold standard to diagnose IgE-mediated food allergy. The time from ingestion to an allergic reaction may differ between allergens, and the few available clinical studies on dosing intervals support that egg may elicit an allergic reaction later than milk.[1–3] During several years of daily clinical practice, we have experienced that children during OFC with raw egg, often react more than 30 min after ingesting the final dose. Consequently, on 1 February 2019, we changed our egg OFC protocol from 30- to 60-min dosing intervals. In order to keep the cumulative dose constant without increasing the total OFC time significantly, the number of doses was reduced and the last two doses equivalent increased. The aim was to investigate whether a longer time interval between fewer but larger doses would overall cause milder reactions in egg-allergic children.

All children aged 0–18 years evaluated for Type 1 allergy to hen's egg at the Paediatric Allergy Unit at Hans Christian Andersen Children's Hospital, Odense, Denmark (a secondary and tertiary referral allergy unit) and Viborg County Hospital, Viborg, Denmark (a secondary referral allergy unit) from September 2017 to December 2021 were consecutively included in the study. The children either presented with a case history of a likely clinical allergic reaction to ingestion of hen's egg or presented with moderate-to-severe atopic eczema and were sensitized to egg white.

A paediatric allergist took a thorough case history, and specific IgE to hen's egg white and ovomucoid (Gal d 1) were measured by ImmunoCAP® (Thermo Fisher, Uppsala, Sweden). An open-titrated OFC with raw pasteurized hen's egg was performed in all children to confirm or reject the clinical suspicion of egg allergy. The raw egg was served with either sugar or fruit puree on the top. Pasteurization of egg has proven not to alter the allergenicity of fresh egg.[4] The children were not challenged with baked egg.

From September 2017 to February 2019, a 6-step protocol with 30-min dosing intervals was used and from February 2019 to December 2021, it was changed to a 4-step protocol with 60-min intervals.

The 6-step protocol was as follows: Time 0: 0.05 g, Time 30 min: 0.25 g, Time 60 min: 1 g, Time 90 min: 5 g, Time 120 min: 20 g, Time 150 min: 25 g. The 4-step protocol was as follows: Time 0: 0.05 g, Time 60 min: 0.25 g, Time 120 min: 6 g and Time 180 min: 45 g.

Hence, the first two doses were identical, and the cumulative dose was equal, 51.30 g egg corresponding to approximately one hen's egg. The OFC was considered positive only in case of objective clinical reactions within 2 h after the last administered dose. Skilled allergy nurses and an experienced paediatric allergist performed all provocations.

Study data were prospectively collected and managed using REDCap electronic data capture tools hosted at Odense University Hospital. The database was approved by The Danish Data Protection Agency and registered in ClinicalTrials.gov (NCT04331522). Written consent to include data in the database for subsequent publication was obtained from all subjects or their parents in case of age < 15 years.

During the study period, 288 OFCs with egg were performed. In case, children had more than one OFC during the study period only data from the first OFC were included. Out of 91 OFCs using the 30-min protocol, 52 (57%) were positive and out of 197 OFCs using the 60-min protocol, 94 (48%) were positive. Consequently, 146 children aged 6 months to 15 years with a positive OFC with egg were included in the study.

There were no significant differences between baseline characteristics of the children in the 30-min and the 60-min protocol groups (Table 1).

One child (2%) in the 30-min group and two children (2%) in the 60-min group were treated with intramuscular adrenaline due to anaphylaxis defined as severe respiratory or cardiovascular symptoms.[5] No subjects developed hypotension.

Table 2 shows the outcomes of the OFC's. Significantly more subjects in the 30-min group compared with the 60-min group had symptoms from more than one organ system: 46% (95% CI: 32%; 61%) versus 30% (95% CI: 21%; 40%), $p = .048$, risk difference: 16% (95%CI: 0.4%; 33%).

In the 60-min group, significantly more subjects reacted to the first dose compared with the 30-min group: 26% (95% CI: 17%; 36%) versus 6% (95% CI: 1%; 16%), $p = .003$, risk difference: 20% (95% CI: 9%; 31%). In the 60-min protocol group, the allergic reaction

occurred more than 30 min after the last administered dose in 29 (31%) of the children: eight children (9%) after 31–45 min and 21 children after more than 45 min.

The number of children that reacted to one of the first two doses was 12 (26%) in the 30-min group and 37 (39%) in the 60-min group. Of these 49 children, 40 (82%) only had symptoms from a single organ system compared with 54 (56%) of the 97 children reacting to a cumulative dose of 1.3 g or more ($p = .004$).

The results suggest that in case of raw egg, a 60-min observation period before administering the next dose allows significantly more subjects to react to the first provided doses. Our data show that 6% versus 26% of egg-allergic children elicit objective allergic symptoms after ingesting 0.05 g raw egg if you wait 30 versus 60 min, respectively, before administering the second dose, corresponding to a risk ratio of 4.4. Similarly, a retrospective study comparing 30-, 40- and 60-min intervals for OFC with low-dose boiled egg white found that a 60-min interval resulted in significantly lower symptom scores and in addition, a lower total dose.[3]

Another recent study also indicated that the time from ingestion to the allergen elicits an allergic reaction is longer for egg than milk with a median time to onset of symptoms of 75 min for OFC with heated hen's egg powder[2] and 50 min for scrambled egg.[1]

Several studies imply a more important role of ovomucoid compared with other allergenic proteins in egg, and the presence of ovomucoid-specific IgE appears to be a good predictor of clinical allergy to egg.[6] Ovomucoid is a potent trypsin inhibitor and has a structural stability that makes the molecule maintain epitopes during the passage through the gastrointestinal tract.[7] Ovomucoid fragments still have IgE-binding activity after pepsin degradation, and the pepsin degradation products retain trypsin inhibitor activity.[8] These characteristics may imply that ovomucoid compared to other allergens can interact with the intestinal mucosa for a longer time, and hence may explain why time from ingestion to allergic reaction may be longer for egg than other allergens.[7]

Key Messages

- We compared outcomes for raw egg food challenges using 30- or 60-min dosing intervals.
- A 60-min dosing interval was associated with more first-dose reactions.
- A 60-min dosing interval was associated with less symptoms but similar adrenaline use.

The choice of dosing interval when performing OFCs is challenging. A too-short interval increases the risk that the patients do not react to the last administered dose. In addition, too short intervals and too small increments in doses may induce a state of temporary desensitization.[9]

Increasing the intervals has the disadvantage of a longer OFC time. We tried to overcome this burden by reducing the number of doses. Consequently, more children ingested the maximal dose of 51.3-g egg, but this was overall not associated with more severe reactions compared with the 30-min protocol.

The study has the strength of including children from both a regional paediatric hospital and a university hospital. We acknowledge several limitations in this study. This was not a randomized double-blinded and placebo-controlled study. However, the baseline characteristics were very similar in the two groups and as only objective immediate reactions were considered positive, assessment bias is limited. Young children with eczema (82%) dominated the study population. Young children may have a reduced ability to digest the proteins in egg white, and findings may be different in older children.[7]

TABLE 1 Patient characteristics

	30-min protocol	60-min protocol	*p*-value
N	52	94	
Site			.799
University Hospital	31 (60%)	54 (57%)	
Regional Paediatric Department	21 (40%)	40 (43%)	
Age (months)	22 [13;33]	17 [12;28]	.112
Gender, male	29 (56%)	66 (70%)	.080
Spec IgE egg white (kU/L)	2.70 [0.89; 5.77]	3.09 [1.20; 7.30]	.759
Spec IgE ovomucoid (kU/L)	0.96 [0.00; 3.94]	1.47 [0.00; 6.13]	.799
Reason for provocation			>.9
Immediate reaction	35 (67%)	64 (68%)	
Eczema and sensitization	17 (33%)	30 (32%)	
Comorbidities			
Asthma/recurrent wheeze	14 (27%)	20 (22%)	.460
Rhinoconjunctivitis	10 (19%)	9 (10%)	.108
Eczema	42 (81%)	78 (84%)	.635

Demographics and clinical characteristics of the children in the 30- and the 60-min-interval group. Data are expressed as *n* (%) or medians [interquartile range].

TABLE 2 Results

		30-min protocol $n = 52$	60-min protocol $n = 94$	p-Value
Skin symptoms		**47 (90%)**	**79 (84%)**	**.286**
Urticaria		39 (75%)	73 (78%)	.829
Localized urticaria		33 (63%)	54 (57%)	.478
Generalized urticaria		6 (12%)	19 (20%)	.183
Angioedema		7 (13%)	7 (7%)	.237
Flushing		11 (21%)	9 (10%)	.051
Acute flare of eczema		5 (10%)	11 (12%)	.699
Gastrointestinal symptoms		**15 (29%)**	**25 (27%)**	**.770**
Vomiting		15 (29%)	23 (24%)	.564
Diarrhoea		2 (4%)	3 (3%)	>.9
Respiratory symptoms		**19 (37%)**	**22 (23%)**	**.091**
Rhinoconjunctivitis		18 (35%)	17 (18%)	.025
Asthma/Wheeze		2 (4%)	5 (5%)	>.9
Severity				
Skin only		25 (48%)	55 (59%)	.225
Gastrointestinal only		2 (4%)	9 (10%)	.178
Respiratory only		1 (2%)	2 (2%)	>.9
Hypotension		0 (0%)	0 (0%)	>.9
>1 organ system		24 (46%)	28 (30%)	.048
3 organ systems		5 (10%)	4 (4%)	.281
Adrenaline use		1 (2%)	2 (2%)	>.9
Cumulative dose (median, IQR)		6.3 g [1.3;26.3]	6.3 g [0.05;26.3]	>.9
Number of children reacting to each of the cumulative doses	0.05 g:	3 (6%)	24 (26%)	.003
	0.30 g:	9 (17%)	13 (14%)	
	1.30 g:	13 (25%)		
	6.30 g:	12 (23%)	33 (35%)	
	26.30 g:	12 (23%)		
	51.30 g:	3 (6%)	24 (26%)	
Proportion of children having symptoms from only one organ system for each dose	0.05 g:	3/3 (100%)	21/24 (88%)	
	0.30 g:	6/9 (67%)	10/13 (77%)	
	1.30 g:	6/13 (46%)		
	6.30 g:	8/12 (67%)	17/33 (52%)	
	26.30 g:	4/12 (33%)		
	51.30 g:	1/3 (33%)	18/24 (75%)	

The symptoms and the eliciting doses for the OFC's with the 30-min interval protocol and the 60-min interval protocol, respectively. Data are expressed as *n* (%). *P*-values are calculated using the chi-squared-test or Fisher's exact test.

In conclusion, the present study suggests that increasing the dosing interval from 30 to 60 min when performing OFCs with raw pasteurized egg may result in more children reacting to a lower dose.

AUTHOR CONTRIBUTIONS

Petersen BT, Halken S and Gradman J all contributed to the design of the study and the collection and interpretation of data. Gradman J performed the statistical analysis and wrote the first draft of the manuscript. All authors critically reviewed the manuscript and have approved the manuscript before submission.

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KEYWORDS

allergen, dosing interval, egg white, food allergy, oral food challenge

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CONFLICT OF INTEREST

The authors declare no conflicts of interest in relation to this work.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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