Author’s response to reviews

Title: An epidemiological investigation of food allergy among children aged 3 to 6 in an urban area of Wenzhou, China

Authors:
Huan Dai (6221696@qq.com)
Fangmin Wang (wfm1992@163.com)
Like Wang (licoko@163.com)
Jinyi Wan (wzwanjy@163.com)
Qiangwei Xiang (7088269@qq.com)
Hui Zhang (zhh855@126.com)
Wei Zhao (zhaow@vcu.edu)
Weixi Zhang (zhangweixi112@163.com)

Version: 1 Date: 17 Mar 2020

Author’s response to reviews:

Response to Reviewers’ Comments

We really appreciate the critical reading of our manuscript entitled "An epidemiological investigation of food allergy among children aged 3 to 6 in an urban area of Wenzhou, China " and we thank the two reviewers for their valuable suggestions. We have carefully considered the comments and have revised the manuscript accordingly. The responses to the comments are listed one by one as follows (please see below). We are looking forward to hearing your final decision.

Reviewer 1
Question 1: Introduction
1) In the first sentence, the definition of food allergy is incorrect. Please refer to the 2006 practice parameters for food allergy: https://www.aaaai.org/Aaaai/media/ MediaLibrary/PDF %20Documents/Practice%20and%20Parameters/food-allergy-2006.pdf. Food allergy is defined as an IgE mediated reaction to food. Adverse reactions to food are NOT food allergy. Also given the reports of reactions to gelatin, FA doesn't have to be always due to reaction to a food protein.
Response: Following reviewer’s valuable suggestion, we carefully read the reference, in 2006 practice parameters for food allergy, it focused on the IgE-mediated food allergy. Food allergy is an adverse immune reaction to food proteins, has a wide spectrum of clinical presentations, ranging from mild skin problems to severe systematic reactions [Le TTK, et al. Pediatr Allergy Immunol. 2019;30:348-55.]. Sicherer SH, et al. [Sicherer SH, Sampson HA. J Allergy Clin Immunol. 2018;141:41-58.] indicated that food allergy, in general, there are IgE-mediated, non–IgE-mediated(cell-mediated), or mixed (IgE and cell-mediated) pathophysiolgies.
2) The introduction should also include information on how food allergy is diagnosed.
Response: Thank you very much for your suggestion. As suggested, we modified the introduction.
Question 2: Methods
1) Include reference for EuroPrevall FA questionnaire
Response: Thank you very much for your suggestion. We have added reference in methods.
2) Include cutoff ranges for SPTs and sIgE measurements that you were using to diagnose concern for IgE mediated food sensitivity
Response: We appreciate reviewer’s suggestion. Result of SPT is considered positive if there is a mean wheal diameter of 3 mm or greater, after subtraction of the saline control. When the result of sIgE measurements is above 0.35 kIU/L, we consider it positive.
3) Please define what you mean by diagnostic criteria for FA (Is this simply the cutoff values for SPT and sIgE?). What pieces of the history did you use to determine that patient had FA?
Response: Thank you for your suggestion. The methods to diagnose FA include history (including diet records), physical examination, SPT, sIgE measurement, trial elimination diets and food challenges. Double-blinded placebo-controlled food challenge (DBPCFC) is the gold standard for FA diagnosis. In clinical work, an OFC is enough to diagnose FA in children [Gupta M, et al. Immunol Allergy Clin North Am 2018, 38(1):39-52.]. When the results of SPTs and sIgE measurements reach the cutoff value (Table 2), we can diagnose FA without food challenge.
Children’s historical aspects of food reactions can certainly aid in identification of suspected allergens and help determine whether other factors play a role in the presentation of symptoms. The clinician should consider about foods that consistently elicit symptoms of food allergy. Foods that have been eaten on multiple occasions and historically tolerated are less likely to be causal foods; however, the ingestion of subthreshold doses or certain preparations (eg, extensively baked or fermented) might result in ingestion of a food allergen without reaction [Sampson HA, et al. J Allergy Clin Immunol 2014;134(5):1016-25.e43].
4) Statistical methods: I only found 1 mention of a P value in your paper. This is incorrect use of Chi-Squared test. Chi-squared test looks at whether there is a significant relationship between two nominal (categorical) variables and typically involves a contingency table. If you want to compare proportions, you should be using the Z test.
Response: We appreciate your suggestion. We used Chi-squared test between two categorical variables, maybe we were wrong in expression, we have revised.

Question 3: Results
1) Where were 352 children excluded?
Response: We appreciate reviewer’s concern. In our study, 534 children’s parents indicated that their children had diseases or problems caused by certain foods or certain types of food. And we telephoned these parents, checked the situation that they describe in the questionnaire which include the timing of symptoms in relation to food ingestion, reproducibility of symptoms with subsequent food ingestion, form, and amount of food ingested. After that 352 of them were excluded. In one of the 352 children, the symptoms were not caused by food, and another gradually became food tolerant.
2) Your methods should detail what you do for the 36 patients who dropped out. Other studies often look at best and worst case scenarios. You could provide a range for prevalence of FA if all 36 had FA or all 36 did not.
Response: Thank you for your comment. As suggested, we assumed the opportunity of confirmed FA was equal among the participants and dropouts, the adjusted estimated prevalence of FA in children was 1.36%.
3) What is mixed food group? (mentioned under SPT and sIgE measurement). Please define what you mean by positive. What did you do with patients who were negative? Did you tell the patients to eat the food and say they did not have FA?
Response: Thank you for your question. Mixed food group includes egg white, milk, wheat, fish, peanut and soybean.
Result of SPT is considered positive if there is a mean wheal diameter of 3 mm or greater, after
subtraction of the saline control. When the result of sIgE measurement is above 0.35 kIU/L, we consider it positive.

In children whose results of SPT and sIgE measurement were all negative, some of them had strongly supported FA histories, we suggest them to undergo OFC. Others we may suggest them try food in small dose when their body is not in allergic statement.

4) Why did you assume that all 10 children who refused OFC did NOT have FA?
Response: We appreciate reviewer’s concern. In our study, 9 children’s parents or guardians of refused the OFC. One of the conclusions: the prevalence of FA was at least 0.84%. We use the phrase “at least” just because these 9 children who were very likely to have FA but dropped out part way, we cannot know whether they have FA or not finally. Therefore, in our study we only provide the minimum rate of the FA prevalence.

Question 4: Discussion
1) The first part of the discussion is a long list of prevalence rates from the literature and is difficult to read. I have difficulty figuring out what the authors are trying to conclude. The entire discussion needs to be revised so that each paragraph drives home the point of the paper:
- Prevalence of food allergy among preschoolers in Asia could be 1 point - provide examples of this age group in this section
- Overestimate of FA - this is well-known and highlights the importance of OFC and allergist referrals. You can provide papers evaluating why this is the case
- Discussion of why the main prevalence is egg. You mentioned dietary differences. Expand on this topic.
Response: Thank you very much for your suggestion. Based on your helpful comments, we have revised the discussion.

2) I would only believe a child truly has a FA if he/she reacts to food on OFC. I disagree that you can diagnose with SPT or sIgE alone. Refer to the practice parameters for further details.
Response: We agree with reviewer’s opinion. Berni Canani R, et al. found food-specific serum IgE concentrations in which approximately 95% of the patients are predicted to have clinical reaction. Above these levels in Table 2, food challenge is not recommended because of the high probability (95%) of clinical reactions [Berni Canani R, et al. Curr Opin Pediatr. 2008;20:584–9.]. Combining with patients’ histories, when SPTs and sIgE measurements reach a 95% PPV, FA can be diagnosed without an OFC.

3) For limitations - you should definitely talk about selection bias, which is unavoidable in population based studies. I don't think lack of basophil activation testing or component resolved diagnosis are that important.
Response: We agree with reviewer’s opinion. Selection bias is one of our limitations in our study. A portion of children’s parents refused to be recruited at the beginning, it can influence the final rate. As suggested, we delete the content about basophil activation testing and component resolved diagnosis.

Reviewer 2
Question 1: Numerous grammatical errors throughout:
Line 45: incomplete sentence: "FA can be induced by IgE or non-IgE"
Line 47: Awkward phrasing: "FA has recently emerged as dominant, following asthma and allergic rhinitis, in the second wave of the allergy epidemic"
Line 197: Recommend alternative word for "intemperance"
Response: We apologize for grammatical errors in our manuscript. As suggested, we have revised and modified the sentence (lines 48-52, lines 235). We also involved native English speakers for language corrections again.

Question 2: Line 105: For the OFCs - what was the top dose? What was the cumulative dose? Were food challenges performed for all foods identified via SPT and immunocap testing?
Response: We appreciate reviewer’s concern. In an OFC, the total amount administered during a
gradually escalating OFC equals 8 to 10 g of the dry food, 16 to 20 g meat or fish, and 100 mL wet food. And we usually started with 0.1% to 1% total challenge food. The challenge food is mixed with the vehicle, and total amount/weight or volume is recorded, considered 100%, and administered in gradually increasing increments every 15 to 30 minutes. Dosing increments may be different, such as doubling of the dose every 15 to 30 minutes until the maximum dose has been reached or the patient reacts, or an increment using logarithmic mean, such as 1, 3, 10, 30, 100, and so forth. [Nowak-Wegrzyn A, et al. J Allergy Clin Immunol. 2009;123:S365-83.]

Not everyone need to undergo OFC, above levels in Table 2, food challenge is not recommended. At present our food challenge performed limited foods, like milk, egg, shrimp, crab and fish.

Question 3: Line 201: It would be interesting to know what is more sensitive (SPT vs. sIgE) for this particular population.

Response: Thank you for your comment. The general sensitivity and specificity of SPT or sIgE measurement for the diagnosis of food allergy are estimated to be greater than 90% and approximately 50%, respectively [ Sampson HA, et al. J Allergy Clin Immunol 2014;134(5):1016-25.e43]. There’s no exact sensitivity of SPT or sIgE measurement in preschool children at present. It worth our continued research.

Question 4: What was the rate of multi-food allergic patients?

Response: Thank you for your question. In our study, only 2 children were allergic to multi-food. The rate of multi-food allergic patients was 0.05%. It may result from the limited allergen test spectrum and food tolerance.

Question 5: Although the adverse events associated with OFC are described in Table 1 – Severity of reactions was not noted.

Response: Thank you very much for your suggestion. We have modified in line 126.

Question 6: Report any differences in eliciting dose among the different allergens? Response: As suggested, we further read some references. A study showed that the cumulative eliciting doses (Eds) in the distribution models ranged from 3.1 to 4.1 mg for the ED05, from 10.6 to 14.6 mg walnut protein for the ED10, and from 590 to 625 mg of walnut protein for the ED50 [Mark AB, et al. J Allergy Clin Immunol Pract 2017;5(2):376-80]. Allen KJ, et al. [ Allen KJ, et al. J Allergy Clin Immunol. Jan 2014;133(1):156-64.] compared cooked whole egg, raw whole egg and raw egg white. They found raw egg white had a much lower ED05 (0.2 mg; [95% CI; 0.05-0.6]) when compared to both cooked and raw whole egg (ED05 of 4.9 mg; [95% CI; 2.1-11.5] and 3.4 mg; [95% CI; 0.6-20.1] respectively). Natasha P, et al. [Natasha P, et al. Front Immunol 2018;9:2057] found the median ED was ≤35 mg of food protein for almond, milk, sesame, cashew, walnut, egg, pecan, wheat, hazelnut and pistachio, peanut had the highest median ED at 75 mg, and pistachio, having the lowest at 5 mg.

Question 7: A little more detail about the food challenges will be helpful? How was the dosing decided? Was it the same protein increments across all patients?

Response: We appreciate reviewer’s question. In an OFC to diagnose immediate, IgE-mediated food hypersensitivity, the total amount administered during a gradually escalating OFC equals 8 to 10 g of the dry food, 16 to 20 g meat or fish, and 100 mL wet food (ie, apple sauce). In non–IgE-mediated food protein–induced enterocolitis syndrome in which there is a low risk for immediate reactions, the total challenge dose is calculated as 0.15 to 0.3 g protein/kg body weight, not to exceed 3 g protein or 10 g whole food.

Different patients may have different protein increments, it depends on patients’ histories and patient reacts.

Question 8: Were any co-occurrence of food allergens noted?

Response: We appreciate reviewer’s concern. In study, we found 2 patients were allergic to egg and milk. Due to some limitations, the actual co-occurrence of food allergens may be more.

Question 9: Was there any exclusion criteria?

Response: Thank you for your question. We exclude the children whose date of birth was not within the
range of 2011.9.1-2015.8.31 or parents cannot cooperate in the beginning. We also considered that some children taken certain drugs such as glucocorticoid before the SPT or OFC would affect the results, so we reasonably arrange the test time to avoid this situation.

Question 10: Were there any associations between baseline characteristics and participants who were more likely to have challenge proven food allergy?

Response: We appreciate reviewer’s question. In FA questionnaires, we gathered children’s past histories, family histories. The rate of children who have rhinitis, rash or wheeze among the 35 FA children was higher than the one of all the 4151. And there was a similar trend in the rate of children whose parents also have atopic disease($P < 0.05$).