Summary

Buckwheat (BW) is a major food allergen and one of the leading causes of food-induced anaphylaxis in Japan. The standard method of diagnosing food allergy is the oral food challenge (OFC). The BW-specific IgE (BW-sIgE) value is used to assess BW allergy but its utility is limited.

The aim of the present study was to identify factors with predictive value for the diagnosis of BW allergy using the OFC.

We evaluated 37 patients who were classified into the positive or negative group according to their OFC results. Ten patients (27.0%) showed objective or persistent, moderate subjective symptoms during the OFC. The positive group had a significantly higher BW-sIgE/total IgE ratio than the negative group ($p<0.001$), but the total IgE ($p=0.139$) and BW-sIgE ($p=0.130$) did not differ significantly. Receiver operator characteristic (ROC) analysis showed that the BW-sIgE/total IgE ratio had a larger area under the curve (AUC, 0.885) than BW-sIgE (AUC, 0.667). The statistically optimal cut-off was 0.0058 for the BW-sIgE/total IgE ratio, which corresponded to a clinical sensitivity and specificity of 90.0% and 81.5%, respectively.

BW-s IgE/total IgE ratio may be more useful predictor of BW OFC results than BW-s IgE.

Keywords: buckwheat; food allergy ; oral food challenge; predictor; specific-total IgE ratio.

Introduction

Buckwheat (*Fagopyrum esculentum*; BW) is a member of family *Polygonaceae* and is commonly consumed worldwide. Examples of BW-based foods include soba (Japanese noodles), guksu (Korean noodles), memilmuk (Korean jelly), groat porridge (Asia, Eastern Europe), pizzoccheri (Italian pasta), polenta taragna (combined with maize), several forms of pancake-blinis in Russia, galettes in Brittany and pofferjes in Netherlands (1,2). Thanks to the recent trend of avoiding gluten, BW is becoming increasingly popular in the West among individuals with celiac disease (2).

Buckwheat (BW) is a major food allergen and one of the leading causes of food-induced anaphylaxis in Japan and Korea (3,4). However, several studies in Europe and Australia have also reported sensitization and allergy to BW (5,6). The standard method of diagnosing any food allergy is the oral food challenge (OFC). However, when BW allergy is suspected, food avoidance without the OFC is frequently prescribed to avoid the risk of anaphylaxis (7).

The BW-specific immunoglobulin E (BW-sIgE) value is used to assess for BW allergy, but its diagnostic accuracy is controversial.[7] We herein hypothesized that the BW-specific IgE/ total IgE ratio would be useful for diagnosing BW allergy as it is for allergies to other food items, including peanut and tree nut (8). The present study evaluated the diagnostic performance of the BW-sIgE/total IgE ratio in light of BW OFC results.

Materials and methods

Patient Selection

The records of patients who underwent an OFC between April 2017 and January 2020 at Tokyo Metropolitan Children's Medical Center were retrospectively reviewed. The patients were classified into a positive or negative group by their OFC results. Patients with a negative OFC result at a total intake < 80 g and those who did not undergo an OFC with a total intake ≥ 80g were excluded.

The present study was conducted in accordance with the principles of the Declaration of Helsinki and the ethical guidelines of Japan and was approved by the Ethics Committee at Tokyo Metropolitan Children's Medical Center [H2019b-181].

OFC

To assess for the presence of BW allergy, an OFC was performed for the patients with suspected BW allergy history and in those who were positive for BW-sIgE but had no history of BW ingestion or a history of consuming only small quantities of the item. The OFC was performed at admission in accordance with the Japanese Food Allergy guidelines.[9] The BW dosage in the form of boiled noodles was determined by each physician. The cumulative dosage was divided into a low-dose category (< 80g containing approximately 3000 mg of BW protein) and a non-low dose category (≥ 80 g). Eighty grams of boiled BW noodle is the highest total challenge dosage recommended by the Japanese Food Allergy guidelines as a total challenge dose (9). The OFC was performed by a physician. In accordance with the Japanese Food Allergy guidelines, an OFC was considered positive if objective symptoms or persistent, moderate, subjective symptoms were observed (9). The severity of the symptoms appearing during an OFC were classified in accordance with the criteria described in the afore-mentioned guidelines (9). The definition of anaphylaxis was based on the diagnostic criteria of the World Allergy Organization Anaphylaxis Guidelines (10). At symptom onset, the patients were treated in accordance with the methods recommended by the European Academy of Allergy and Clinical Immunology's (EAACI) food allergy and anaphylaxis guidelines (11). Written informed consent was obtained from the patients' guardians before each OFC.

Laboratory data

Blood samples before the OFC were analyzed. Total IgE and BW-specific IgE (BW-sIgE) levels were assessed using ImmunoCAP (Thermo Fisher Scientific/Phadia, Uppsala, Sweden), which can detect levels as low as 0.1kU$_A$/L. If the BW-sIgE value was less than 0.1 kU$_A$/L, the measurement was treated as equivalent to 0.05 kU$_A$/L as previously described (12).

Outcome measures

The patient characteristics, including sex, age at OFC, history of immediate-type reactions to BW, anaphylaxis due to BW, and allergic complications at the time of the OFC, were reviewed. The present study's primary aim was to evaluate the diagnostic performance of the BW-sIgE value and the BW-sIgE/total IgE ratio for BW allergy. The secondary objective was to evaluate the difference in baseline characteristics between the positive and negative groups.

Statistical Analysis

Patient age, sex, symptoms during OFC, treatments administered, and laboratory data, including total IgE, BW-sIgE, and the BW-sIgE/total IgE ratio, were employed as variables in the analysis. Univariate analysis of the groups was conducted using the Mann-Whitney U test or Fisher's exact test. $P<0.05$ was considered to indicate statistical significance. Receiver operating characteristic (ROC) curves were generated for the BW-sIgE value and the BW-sIgE / total IgE ratio. The diagnostic performance of the variables was evaluated using the area under the curve (AUC). All statistical analyses were performed using IBM SPSS statistics Version 26.0 (IBM Corp., Armonk, NY, USA).

Results

Patient characteristics

Figure 1 shows the OFC results. Thirty-seven patients were enrolled in the final analysis. Table I summarizes the patient characteristics. Twenty-five patients (67.6%) were male. The median age at diagnosis was 89 months (range: 46-173 months). Twenty-two patients (59.6%) had never eaten BW with fear of an allergic reaction caused by BW because they were BW s-IgE positive or had concomitant food allergies. Nine patients (24.3%) had a history of eating buckwheat without an allergic reaction, but they had only eaten a very small quantities. Six patients (16.2%) had a

suspected history of immediate-type reaction to BW, and two of them experienced anaphylaxis due to BW. Almost all the patients (97.3%) had a history of allergy to foods other than BW. Table II lists the causative foods in the concomitant cases of non-BW food allergies. The most common causative food was egg. Fourteen patients (37.8%) had atopic dermatitis, and seven patients (18.9%) had bronchial asthma.

OFC results, induced symptoms, and treatments

Ten of the 37 (27.0%) patients were classified as positive. Table Ⅲ shows the symptoms and treatments of the positive group during the OFC. In the positive group, eight, one, and one patient presented with mild, moderate, and severe symptoms, respectively. The most common symptom during the OFC was gastrointestinal symptoms, which were observed in seven patients. Antihistamine, the most frequently prescribed treatment, was administered to four patients. Two patients demonstrated anaphylaxis but none of the patients received intramuscular adrenaline because the skin and respiratory symptoms in those two patients did not appear simultaneously and were therefore treated at different times with antihistamine and β stimulant inhalation.

Comparison of OFC results between the positive and negative groups

As shown in Table I, patients in the positive group more frequently had a history of immediate-type reactions to BW (P=0.035). The serum BW-sIgE value did not differ significantly between the groups (P=0.116), but the BW-sIgE/total IgE ratio was significantly higher in the positive group (P<0.001).

Receiver operator characteristic (ROC) analysis (Fig 2) demonstrated that the area under the curve (AUC) for the BW-sIgE value and BW-sIgE/total IgE ratio in the positive group was 0.664 and 0.888, respectively (Table Ⅳ). The statistically optimal cut-off value for the BW-sIgE/total IgE ratio was 0.0068, corresponding to a clinical sensitivity and specificity of 90.9% and 81.5%, respectively.

Discussion

To the best of our knowledge, the present study is the first to demonstrate that the BW-sIgE/total IgE ratio may have a higher predictive value for BW allergy than BW-sIgE. The allergen-specific IgE/total IgE ratio is a biomarker that can be easily calculated. Its utility has been assessed for many food items, such as milk, egg, wheat, peanut, and tree nut (8,13). The utility of the specific IgE/total IgE ratio is thought to derive from the probability that the surface density of IgE antibody molecules on mast cells and basophils with specificity for the same allergen, which are capable of inducing mediator release after an allergen encounter, increases in proportion to the increase in the ratio of a particular IgE antibody specificity. The ratio may more accurately reflect the specific binding capacity on the surface of mast cells and basophils and therefore by extension the probability of allergen cross-linking and subsequent activation. This reduces the false-positive test rate that may be due to non-associated IgE and confounding immune markers not represented on testing (8).

In the largest retrospective study of BW OFC ever done, 44 of 419 (10.5%) pediatric patients with suspected BW allergy tested positive; of these 54.5% presented anaphylaxis, and 22.7% presented with a severe reaction (14). In our study an anaphylactic reaction occurred in two of ten patients in the positive group, of whom one presented with a severe reaction. On the other hand, 6-10% of patients who tested positive on an OFC with milk, eggs, wheat, and soy presented with severe symptoms (15). The incidence of anaphylactic reactions during BW OFC with a severity equal to or greater than that induced by other food items suggests the need for careful caution when performing the test. Knowing the risk factors of an allergic reaction prior to performing a BW OFC may therefore be desirable.

The present study demonstrated not only that the presence of a high BW-sIgE/total IgE ratio, but also a history of allergy to BW were associated with positivity on a BW OFC, in line with the findings of a previous study (14). These risk factors were also found in a retrospective study of 93 children who underwent a wheat OFC (16).

Although previous studies reported the skin prick test (SPT) and Fag e 3-sIgE, one of the components of BW, as useful predictors of OFC results (17,18), these were not assessed in the present study because SPT using BW is not part of routine clinical practice at the study institution, and Fag e 3-sIgE was unable to be clinically assessed. However, the allergen-specific IgE/total IgE ratio is easy to determine on the basis of blood test results alone and may therefore be extremely useful in daily clinical practice. Although diagnostic performance of the BW sIgE/total IgE ratio was not sufficient to obviate the need for an OFC, it was superior to BW-sIgE alone in predicting the risks and results of BW OFC when combined with an assessment of the patient's medical history.

The present study had a number of limitations. First, it was retrospective, and the OFC conditions (dosage, interval, and frequency of loading) were decided by individual physicians, thus possibly introducing variations affecting the OFC results. Second, we were unable to ascertain whether patients with negative results on a low dose (< 80g) BW OFC would be able to tolerate a dosage ≥80g partly because the patients' parents were unwilling to consent to the test out of fear of a severe allergic reaction, etc. These patients were excluded from the analysis despite the possibility of introducing a selection bias because OFC results were thought to be highly dose-dependent. Third, the number of patients analyzed was too small to allow any definitive conclusions to be drawn. However, this fact may conversely be a strength in that statistically significant results were able to be obtained for the diagnostic performance of the BW sIgE/total IgE ratio in spite of the small sample size. Forth, the OFC were not double-blind placebo-controlled food challenges (DBPCFC). Since subjective symptoms, such as stomachache, occurred in patients with a positive OFC result, DBPCFC would theoretically be appropriate for diagnosing the allergy. However, the OFC is time-consuming when performed multiple times, making DBPCFC for BW difficult to perform in practice.

Conclusions

The findings of the present study suggest that the BW-sIgE/total IgE ratio may be a more useful predictor of BW OFC results than the BW-sIgE value.

Acknowledgements

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Conflicts of interest

The authors declare that they have no conflict of interests.

Author contributions

NK wrote the manuscript. NK, KY, EK, and MN designed the study. NK, KY, KH, and SY conducted the OFC. KY, EK, and MN reviewed the manuscript for critical content. All the authors have read and approved the final version of the manuscript.

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Legends

Fig1. Results of BW OFC

BW, buckwheat; OFC, oral food challenge

Fig 2. Receiver operating characteristic curve for positive oral food challenge results

BW-sIgE, buck wheat-specific immunoglobulin E; Total IgE, total immunoglobulin E