

# Trehalose phosphorylase as a novel potential allergen in a case of allergic reaction due to oyster mushroom (*Pleurotus ostreatus*) ingestion



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To our knowledge, we present the first case report of allergic reaction from oyster mushroom ingestion, which was confirmed by an oral food challenge test. Trehalose phosphorylase was identified as a novel potential allergen by IgE immunoblotting and mass spectrometry. (J Allergy Clin Immunol Global 2023;2:100095.)

**Key words:** Mushroom allergy, oyster mushroom, trehalose phosphorylase, anaphylaxis, allergen

Mushrooms are a common food that is consumed around the world. Inhalation-related mushroom spore respiratory reactions<sup>1,2</sup> and a case of anaphylactic reaction following inhalation of oyster mushroom (*Pleurotus ostreatus*)<sup>3</sup> have been reported. However, allergic reactions following ingestion of mushrooms are rare, and only a few mushroom allergens have been identified.<sup>4-6</sup> Herein, we present the case of a boy who had an allergic reaction after ingestion of oyster mushrooms. Trehalose phosphorylase was identified as a potential allergen by IgE immunoblotting and mass spectrometry. Informed consent to publish the case report was obtained from the patient's parents. This study was approved by the Mae Fah Luang University Ethics Committee on Human Research.

A 12-year-old Thai boy presented to the emergency department with generalized hives, abdominal pain, and vomiting an hour after having eaten homemade oyster mushroom soup. At the emergency department, he had normal vital signs and oxygen

## Abbreviations used

OFC: Oral food challenge  
PTP: Prick-to-prick  
SPT: Skin prick testing

saturation. A physical examination revealed generalized urticaria and eyelid and lip angioedema. The boy's abdomen had

**TABLE I.** Results of skin prick test and prick-to-prick test

Type of mushroom	Wheal diameter (mm)	
	Raw form	Cooked form
Standard mushroom solution (1:10 wt/vol)		3
Oyster mushroom ( <i>Pleurotus ostreatus</i> )	8.5	8.5
King oyster mushroom ( <i>Pleurotus eryngii</i> )	2	2.5
Straw mushroom ( <i>Volvariella volvacea</i> )	0	0
Shiitake mushroom ( <i>Lentinula edodes</i> )	3	3
Enokitake ( <i>Flammulina velutipes</i> )	3	4
Jelly ear mushroom ( <i>Auricularia cornea</i> )	2.5	3

Histamine (10 mg/mL) and glycerinate phenol-saline were used as positive and negative controls, respectively. The largest wheal of each allergen was measured. A positive response was considered wheal size at least 3 mm larger than the negative control.

generalized tenderness. Severe allergic reaction was diagnosed. He was immediately treated with intramuscular epinephrine. The patient fully recovered after treatment.

The patient was previously healthy. He never had any symptoms of chronic urticaria, asthma, or allergic rhinitis. His mother prepared his meals. The ingredients, consisting of oyster mushrooms, chicken, and mixed herbs, were freshly bought from a local market. The family frequently consumed foods with these types of spices without any reactions. The boy experienced an anaphylaxis episode that required hospital admission 2 years previously after he had consumed deep-fried tempura made from a mixture of different kinds of mushroom at school. Several times previously, he had mild itchy rash after ingesting mushrooms. Hence, the patient visited our hospital for further investigation of mushroom allergy.

The patient's mother provided various kinds of typically consumed mushrooms that had been purchased from the local market. All of the mushroom samples were sent for species identification at The Center of Excellence in Fungal Research, Mae Fah Luang University, Chiang Rai, Thailand, by DNA sequencing. The mushrooms were identified as oyster mushroom (*P. ostreatus*), king oyster mushroom (*Pleurotus eryngii*), straw mushroom (*Volvariella volvacea*), shiitake mushroom (*Lentinula*

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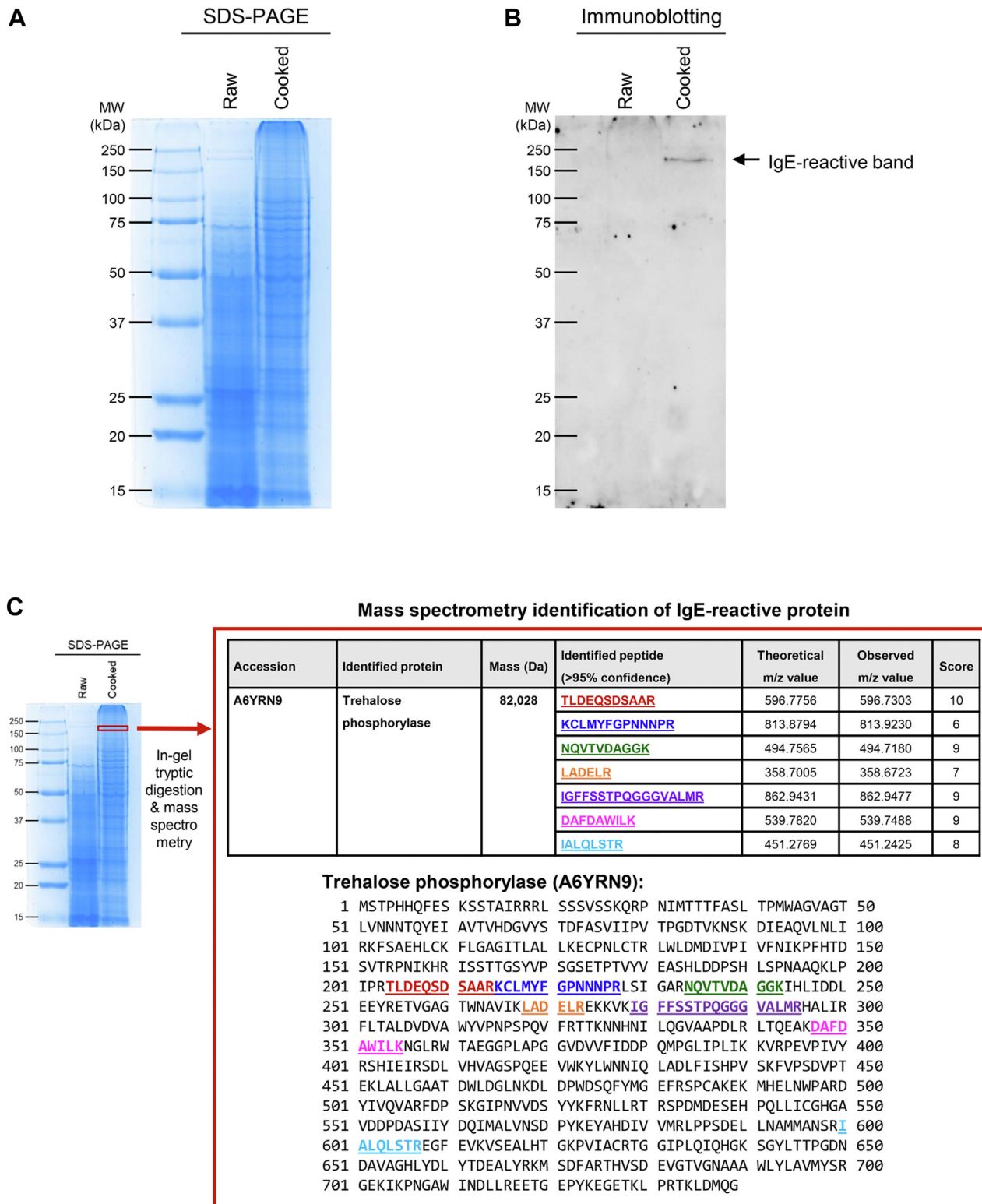
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**FIG 1.** Identification of potential allergenic proteins in oyster mushrooms. *P. ostreatus* proteins from raw and cooked oyster mushrooms were subjected to SDS-PAGE (A) and immunoblotting (B) with the patient's serum. A major IgE-reactive protein band with a molecular weight between 150 and 250 kDa was detected exclusively in the cooked mushroom (B). The corresponding IgE-reactive protein band on Coomassie blue-stained gel was excised and subjected to in-gel tryptic digestion and mass spectrometry. A search against the UniProt database identified the protein as trehalose phosphorylase. Details of the mass spectrometry result table and location of the identified peptides (*bold underlined residues*) within the amino acid sequence of trehalose phosphorylase are shown (C).

edodes), enokitake mushroom (*Flammulina velutipes*), and jelly ear mushroom (*Auricularia cornea*). The cooked samples of each mushroom were prepared separately by boiling for 15 minutes. A skin prick test (SPT) was performed using commercial mushroom allergen extracts (ALK Abelló, Port Washington, NY). The prick-to-prick (PTP) skin testing method using raw and cooked mushrooms has been previously described.<sup>4</sup> We also performed common aeroallergen SPT using commercial extracts. The patient had a positive SPT result for commercial mushroom extracts. The results of PTP testing were positive for 3 types of mushrooms (oyster, shiitake, and enokitake) in both cooked and raw forms, whereas only the cooked form of jelly ear mushroom was reported to have elicited a positive result (Table I). For the common aeroallergens, the results of SPT were positive for house dust mites, cockroaches, and cat pelt but negative for *Cladosporium*, mixed mold, and pollens. The result of testing for serum-specific IgE (using ImmunoCAP, Thermo Fisher, Waltham, Mass) was negative for *Aspergillus fumigatus* (0.05 kUA/L).

The open-label oral food challenge (OFC) with 6 different kinds of cooked mushrooms in separate sessions included oyster, king oyster, straw, shiitake, enokitake, and jelly ear mushrooms. A positive result was obtained only with oyster mushrooms. The patient had generalized urticarial rash and oral pruritus 10 minutes after ingesting 10 g of cooked oyster mushroom. The testing procedure was stopped, and oral antihistamine was given. No severe systemic reaction occurred during the OFC, which may imply that sensitization only by skin test might not be enough to demonstrate a true mushroom allergy.

To identify potential allergenic proteins in oyster mushrooms, proteins were extracted from raw and cooked oyster mushrooms and subjected to SDS-PAGE and immunoblotting with the patient's serum (see the Supplementary Methods in the Online Repository at [www.jaci-global.org](http://www.jaci-global.org)). An IgE-reactive protein band with a molecular weight between 150 and 250 kDa was detected exclusively in the cooked mushroom (Fig 1, A and B). Mass spectrometric analysis based on a search against the UniProt database identified this protein as trehalose phosphorylase (UniProt identifier A6YRN9) (Fig 1, C). Amino acid sequence alignment with known allergens in the COMprehensive Protein Allergen REsource (COMPARE) database<sup>7</sup> showed that trehalose phosphorylase had no significant hit with any allergen in the database, suggesting that this protein could be a novel allergen from the mushroom (see the Supplementary Methods).

Trehalose phosphorylase is an enzyme involved in the biosynthesis of trehalose in fungi. The monomer of trehalose phosphorylase from *P. ostreatus* has a theoretical molecular

weight of 82 kDa. An active form of this enzyme is presented as a homodimer.<sup>8</sup> The major immunoreactive band with a molecular weight around 160 to 170 kDa that was observed in this study suggests that this enzyme was presented mainly as a heat-stable homodimer in the cooked mushroom.

In this study, the PTP skin test result was positive for both raw and cooked oyster mushroom, whereas the IgE-reactive protein band was detected only in the cooked sample by immunoblotting assay. These findings suggest that the allergenicity of the mushroom might be influenced by heating or modification by cellular activity, but its underlying mechanism requires further clarification.

To our knowledge, this is the first case report of allergic reaction from ingestion of oyster mushroom. The results PTP skin testing showed were positive for various kinds of mushrooms, suggesting a high degree of cross-reactivity.<sup>4,6</sup> Nevertheless, OFC confirmed that only oyster mushroom was the culprit. Before it can be concluded that trehalose phosphorylase is a novel allergen and allergenic protein in oyster mushrooms, experiments using recombinant or purified protein must be conducted for further validation. In-depth study of mushroom allergen may be beneficial for a better understanding of allergic diseases related to the fungi kingdom.

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