# **Selective Hypersensitivity to Boiled Razor Shell**

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### Abstract

Many types of seafood require cooking before ingestion and it has been demonstrated that this cooking process may affect the antigenicity and allergenicity of the food. We describe a case of anaphylaxis caused by selective sensitization to razor shell, a mollusc. In vivo and in vitro studies confirmed sensitization to boiled razor shell. Analysis of the nature of the allergen yielded results that were consistent with the findings of other authors and suggested that allergens involved in seafood allergy are commonly high molecular weight proteins that, in most cases, are heat stable.

Key words: Shellfish. Razor shell. Boiled razor shell. Food allergy. Ensis macha.

## Resumen

La mayoría de las diferentes clases de marisco necesitan ser cocinados antes de su ingesta y se ha demostrado que este proceso de cocción puede afectar la antigenicidad y alergenicidad de los mismos. Describimos un caso de anafilaxia causada por la sensibilización selectiva frente a navaja cocida, un molusco. Los ensayos *in vivo* e *in vitro* confirmaron la sensibilización frente la navaja cocida. Los análisis del alérgeno nativo son consistentes con los hallazgos de otros autores y sugieren que los alérgenos involucrados en la alergia a mariscos son normalmente proteínas de alto peso molecular, que en la mayoría de los casos son termoestables.

Palabras clave: Marisco. Navaja. Navaja cocida. Alergia a alimentos. Ensis macha.

# Introduction

Although seafood is an important element of the human diet, it is known to be a common cause of severe allergic reactions and is a major cause of food hypersensitivity. Many types of seafood must be cooked before consumption. However, studies have suggested that the cooking process might modify the allergenicity of the food [1] due to the formation of new allergens [2]. Hoffman et al [3] demonstrated the presence of both thermostable and thermolabile antigens in shrimp. The thermostable antigen was identified and had a molecular weight of 38 kilodaltons (kd). Subsequently, the shrimp allergen was identified as tropomyosin, a protein from muscle [4].

# **Case Description**

A 34-year-old woman suffered immediate facial edema, widespread wheals with pruritus, sneezes, rhinorrhea, nasal itching, cough, chest tightness, and dyspnea 10 minutes after the ingestion of boiled razor shell (*Ensis macha*), a mollusc widely consumed in Spain. The patient attended the emergency department and the symptoms disappeared after treatment with corticosteroids and antihistamines. The patient was able to consume other boiled molluscs (mussel, octopus, and clam) and shellfish (shrimp and king prawn) without clinical symptoms.

Skin prick tests (SPT) were negative with a battery of commercially available shellfish allergen extracts, including

Figure 1. Sodium dodecyl sulfate polyacrylamide gel electrophoresis of extract from raw razor shell (lane 1) and boiled razor shell (lane 2). MW indicates low molecular weight markers; kd, kilodalton.

1

2

MW

crustaceans (shrimp and king prawn), bivalves (clam, mussel, and oyster), and cephalopods (squid and octopus) (Laboratories LETI, Tres Cantos, Madrid, Spain). In addition, extracts were prepared from raw and boiled razor shells by Laboratorios LETI S.L. (Madrid, Spain). To obtain the boiled extract, razor shells were boiled for 15 minutes in phosphate buffered saline (PBS). Afterwards extracts from raw and boiled material were prepared according to the same protocol. Briefly, the material was homogenized and extracted for 4 hours at 4°C with continuous stirring. After centrifugation, the supernatants were collected, dialyzed overnight against double-distilled water, frozen, and freeze dried. Using these extracts, SPT with raw razor-shell extract was negative, while SPT with boiled razor-shell extract was positive. Prick-to-prick testing with boiled razor-shell extract was also positive.

The protein profile of the razor-shell extracts was assessed by sodium dodecyl sulfate polyacrylamide gel electrophoresis. Extracts were loaded onto the gel (15% T, 2.67% C) under reducing conditions. Following electrophoresis, the gel was stained with Coomassie blue. Analysis of the protein profile revealed important differences between raw and boiled razorshell extracts. Several proteins with a molecular weight range of 10 to 98 kd were identified. While the most prominent band in the raw razor shell extract had a molecular weight of approximately 42 kd, the most prominent band in boiled extract had a molecular weight of 34 kd (Figure 1).

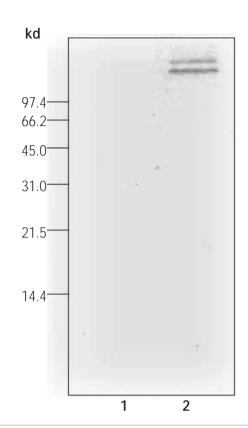


Figure 2. Immunoblot showing binding of immunoglobulin E in the patient's serum to high molecular weight bands in extract from boiled razor shell (lane 2) but not raw razor shell (lane 1). kd indicates kilodalton.

The allergenic profile of the extracts was determined by immunoblot assay. Briefly, after electrophoresis, proteins were transferred to an Immobilon membrane (Millipore, Bedford, USA). The membrane was incubated overnight with the patient's serum diluted 1:5 in PBS containing 0.1% Tween. After washing, the membrane was incubated with peroxidase-conjugated anti-human immunoglobulin (Ig) E (Ingenasa, Madrid, Spain). Antibody binding was revealed by chemiluminescence. Two high molecular weight bands were recognized in the boiled razor shell extract by the patient serum. No bands were recognized in the raw extract (Figure 2).

## Discussion

Hypersensitivity reactions after the ingestion of shellfish represent one of the most frequent food allergies in adults. Most cases involve allergy to multiple shellfish, such as lobster, crabs, and shrimp or prawns. The affected individuals may develop urticaria, angioedema, asthma, and anaphylaxis. Allergy to crustaceans is more common than allergy to molluscs, including clam, mussel, etc [4]. We describe a case of anaphylaxis caused by a selective sensitization to razor shell. The patient tolerated all crustaceans and other molluscs. Diagnosis of shellfish hypersensitivity should always be

kd

97.4

66.2-

45.0-

31.0

21.5

14.4-

established using extracts of both raw and cooked material, as a difference has been demonstrated between them in terms of recognition [5]. SPT suggested a specific IgE sensitization to the boiled extract. Due to the results of skin tests and the severity of the reaction caused by ingestion of the boiled razor shell, oral challenge with boiled razor shell was ruled out by the patient and the attending physician.

Our data revealed modification of the antigenic profile of razor-shell extract after boiling. Several bands were detected in a molecular weight range of 9 to 90 kd; however, antigenic characterization of the extracts revealed different profiles. The most prominent band in extract of boiled material corresponded to a protein of about 34 kd that could be similar to tropomyosin, as described previously in shrimp [4]. Consistent with this finding, Leung et al [6] also identified a 34-kd protein, Cha f 1, as the major allergen in the crab Charybdis feriatus and demonstrated its similarity to shrimp tropomyosin. Immunoblot of the boiled extract demonstrated that serum from our patient did not recognize the 34-kd protein, while 2 high molecular weight proteins were recognized. In contrast, no bands were detected in the raw extract. Those allergens were the only bands recognized by the patient serum in boiled extract, demonstrating that they were generated during thermal processing of razor shell. These allergens may play an important role in the symptoms induced by the ingestion of boiled razor shell.

To our knowledge, only 1 other case of immediate hypersensitivity to razor shell has been documented to date [7]. In the previous report, prick-to-prick testing with the implicated razor shell was positive. However the authors did not evaluate sensitization in response to raw versus boiled razor shell. In our case, it was noteworthy that the sensitization was due to boiled razor shell.

In summary, we present a case of specific IgE-mediated hypersensitivity after ingestion of razor shell. In vivo and in vitro studies suggested sensitization to boiled razor shell. Our study and those of other authors confirm that proteins involved in seafood allergy seem to be high molecular weight allergens and in most cases, heat stable [8].

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