

Clinical Communications

Species-specific evaluation of 2 patients with suspected crab allergy

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Clinical Implications

- Conserved sequence identity between shellfish allergens has been well described, but the clinical significance is not well established. Species differentiation should be considered when evaluating for shellfish allergy despite current limitations in diagnostic testing.

We present 2 patients with demonstrated reactions to specific crab species and symptoms from oral food challenge (OFC) but negative skin prick test (SPT) results to commercial extracts and serum specific IgE (sIgE) testing to “crab” (Table 1). Patient 1 is an 18-year-old woman with history of reactions to snow crab (*Chionoecetes opilio*). She regularly ingested Atlantic blue crabs, shrimp, clams, mussels, and oysters without any reaction. Patient 2 is a 44-year-old woman with a history of oral itching and pruritic rash after ingestion of blue swimmer crab, *Portunus armatus* (formerly *P. pelagicus*). She previously ingested Atlantic blue crab and blue swimmer crab without issues.

Both patients had positive SPT results to house dust mite (HDM) allergens and negative to shellfish. SPT wheal size with cooked crabmeat for patient 1 was 1 mm to snow crab and 3 mm to Dungeness and King crabs. Fresh meat SPT was not performed on patient 2. sIgE to crab (*C. opilio*) through LabCorp (Burlington, NC) showed IgE less than 0.10 kU/L for both patients, as well as for lobster, oyster, shrimp, and scallop. OFC for patient 1 performed with the primary sensitizer snow crab resulted in urticaria, nausea, and severe abdominal pain after the second dose (<3 g total crab protein) requiring epinephrine. OFC for patient 2 was also performed with the primary sensitizer, canned blue swimmer crab. She developed throat pruritus after the second dose, but no further symptoms were noted, ultimately completing the challenge with equivocal results.

Western blot analysis was conducted to examine IgE binding using patient sera on crude protein extracts isolated from claw meat of different crab species, HDM allergen, and SPT reagents (Figure 1). IgE binding was detected by incubation with secondary antibody (antihuman IgE) and tertiary antibody (anti-mouse Infrared dye [IR800]). Both patients demonstrated strong immunoreactivity to multiple protein bands between 15 and 25 kDa for HDM, *Dermatophagoides pteronyssinus*, and *Dermatophagoides farinae*. However, similar IgE immunoreactivity was not observed for either crab species or crab SPT extract. Although we observed weak immunoreactivity for both patients for a protein band approximately 17 kDa, we observed immunoreactivity at similar molecular weight in our control subjects. This led us to conclude that the approximately 17-kDa protein is possibly a false-positive result that has no diagnostic value and

likely represents a nonspecific interaction between the secondary and tertiary antibodies (independent of serum) and snow crab proteins.

Crab is a major cause of anaphylaxis in both children and adults.¹ Occupational IgE-mediated contact urticaria with crustaceans has also been reported in the seafood industry.² The diversity of seafood consumed, however, makes it difficult to identify the exact species and allergens involved in allergic reactions, and avoidance of all shellfish is often recommended.³

The identification of crab allergens at a molecular level has been well described, including the initial report of tropomyosin (*Cha f 1*) as the major allergen of the crucifix crab *Charybdis feriatus*.⁴ Subsequent reports have underscored the relevance of tropomyosin as a major allergen in numerous other crab species including King crab (*Paralithodes camtschaticus*) and snow crab (*C. opilio*).⁵ Tropomyosin is considered the major invertebrate pan-allergen found in all edible crustacean species.¹ Arginine kinase has been reported as a pan-allergen, notable in several crab species including snow crab.^{6,7} Both tropomyosin and arginine kinase share highly conserved amino acid sequences among crustaceans.

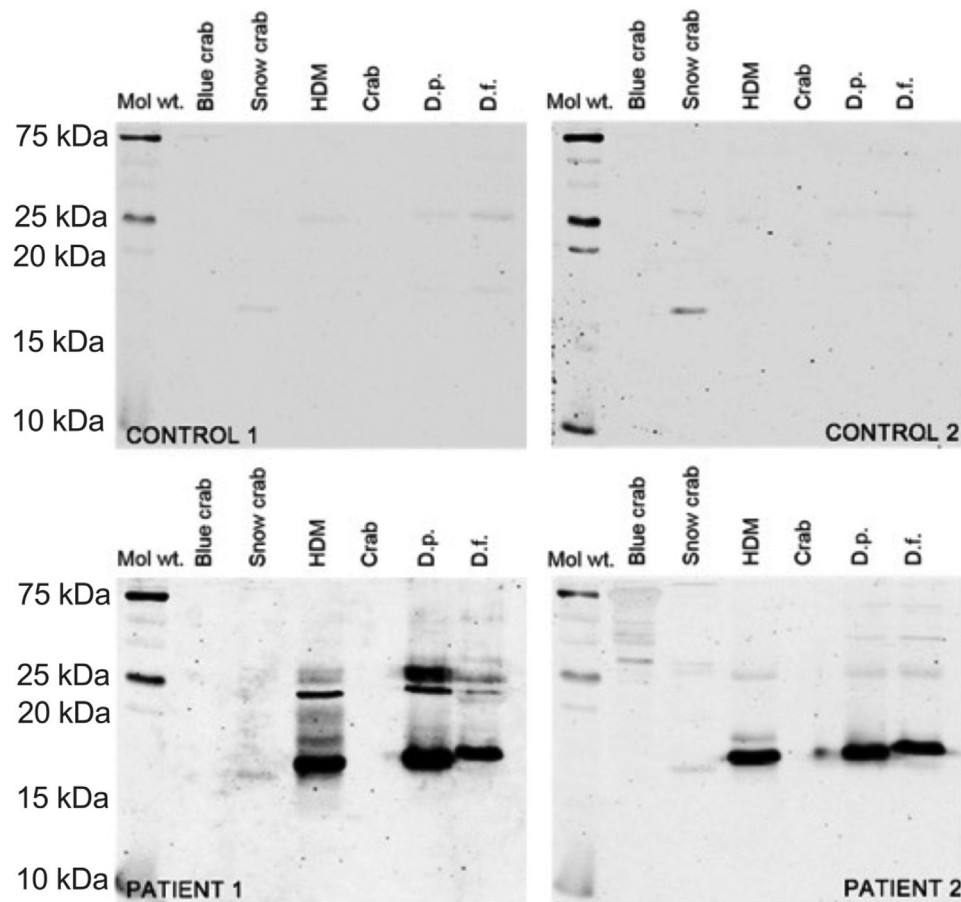
Current evaluation of shellfish allergy through SPT is still performed using commercial preparations derived from whole-protein extracts with incompletely defined protein composition. Commercial allergen extracts are commonly derived from a single crab species such as Atlantic blue crab *Callinectes sapidus* (Greer Laboratories, Lenoir, NC) and King crab *P. camtschaticus* (Center-AI brand, ALK laboratories, Horsholm, Denmark). Both patients had a negative SPT result using the Greer extract, supporting their history of previous tolerance of Atlantic blue crabs. It does not, however, aid in the evaluation of their clinical reactions to snow or blue swimmer crabs.

sIgE testing is also inconsistent, with variable crab species used in the development of ImmunoCAP testing between laboratories. For example, ImmunoCAP testing through Quest Diagnostics (Secaucus, NJ) and ARUP Laboratories (Salt Lake City, Utah) is derived from edible brown crab (*Cancer pagurus*) compared with snow crab (*C. opilio*) through LabCorp and Mayo Clinic Laboratories (Rochester, Minn). Our first patient had sIgE testing to the crab species in question, but our second patient reacted to a species for which testing is not available by either commercial extract or sIgE. Despite testing negative for sIgE to snow crab, patient 1 had anaphylaxis with ingestion of snow crab during OFC. This unexpected outcome raised further questions regarding the role of her clinical reaction in the context of HDM allergy and known cross-reactivity.

Component-resolved diagnostics is increasingly being applied in the assessment of food allergy, with the goal to distinguish the true allergens from nonspecific reactions in the context of peanut, tree nuts, milk, and egg. However, component-resolved diagnostics has not yet become widely available for use in the clinical evaluation of seafood allergy.^{8,9} Western blot analysis is a powerful tool to capture IgE binding to specific crab species. It can be particularly useful in the identification of specific allergens when the technique is interfaced with proteomics approach. Both patients had positive IgE to HDM on SPT and western blot analysis but negative IgE to crab. This suggests that allergic

TABLE I. Summary of clinical and laboratory evaluation

Allergy test		Patient 1 results	Patient 2 results
SPT	HDM	Positive	Positive
	Commercial crab extract (Greer, <i>Callinectes</i> spp)	Negative	Negative
	Fresh cooked crab	3-mm wheals Dungeness & King crabs, 1-mm wheal snow crab	Not applicable
sIgE	Crab sIgE (LabCorp, snow crab, <i>Chionoecetes</i> spp)	<0.1 kU/L	<0.1 kU/L
	Total IgE	19 kU/L	33 kU/L
OFC	Crab species	Snow crab (<i>C opilio</i>)	Blue swimmer crab (<i>P armatus</i>)
	Results	Urticaria, nausea, severe abdominal pain (<3 g protein)	Equivocal with mild throat pruritus (>8 g protein)
Western blot (IgE)	HDM	Positive	Positive
	Crab	Negative (similar to control)	Negative (similar to control)

**FIGURE 1.** Serum IgE western blot analysis. Serum (diluted 1:5 (vol/vol)) was tested against crude protein extracts from blue and snow crab (20 µg/lane), HDM (20 µg/lane), and SPT reagents (crab extract, 1:20 wt/vol, undiluted; *D.p.* and *D.f.* 10,000 au/mL, diluted 1:1 vol/vol). *D.f.*, *Dermatophagoides farinae*; *D.p.*, *Dermatophagoides pteronyssinus*; Mol wt., molecular weight marker.

sensitization to crab species and HDM is not likely linked by shared molecular determinants. It is also possible that the offending allergens relevant in our patients have conformational epitopes not detected using the western blotting techniques discussed here, because it was performed under reducing

conditions to investigate linear epitopes. Western blot analysis using native (or nondenaturing) gels may help clarify this preliminary observation.

Despite high gene sequence identity between species, it is not well known how this translates to significant clinical

reactions, as seen in our patients, and further research in this area could impact the way we evaluate, treat, and manage shellfish allergy.

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