The Burden of Allergens in Surimi-Based Products Diminishes With Industrial Processing

Pedrosa M[1,2], Loli-Ausejo D[1], Garcia-Lozano JR[3], Fiandor A[1,2], Lluch-Bernal M[1,2], Hurtado JL[4], Dominguez-Ortega J[1,2], Quirce S[1,2], Gasset M[5], Rodriguez-Perez R[2]

[1]Department of Allergy, La Paz University Hospital, Madrid, Spain

[2]Allergy Research Group, La Paz Hospital Institute for Health Research (IdiPAZ), Madrid, Spain

[3]Department of Immunology, Virgen del Rocio Hospital, Seville, Spain

[4]Angulas Aguinaga Research Center, Irura, Spain

[5]Institute of Physical-Chemistry Rocasolano (IQFR), Spanish National Research Council (CSIC), Madrid, Spain

J Investig Allergol Clin Immunol 2021; Vol. 31(5): 443-445
doi: 10.18176/jiaci.0674

Key words: Food allergens. Egg allergy. Milk allergy. Fish allergy. Surimi.

Palabras clave: Alérgenos alimentarios. Alergia al huevo. Alergia a la leche. Alergia al pescado. Surimi.

Surimi products are a mixture of the flesh of small fish of many species minced, thoroughly washed, and gelated [1]. To improve the properties of the gel and to give surimi products different textures and flavors, various proteins are added to the mixture, mainly soy protein isolate, whey protein from milk, and egg ovalbumin [2,3].

Allergic reactions to surimi-based products have been reported in fish-allergic patients [4,5]. However, there are no reports of IgE recognition of egg or milk allergens in surimi-derived products or of variations in allergen content during industrial processing.

We investigated the allergenicity of baby eel surimi (BES), which includes milk in its formulation, and crabstick surimi (CS), which includes egg. Prick-by-prick testing (PPT) and the basophil activation test (BAT) were performed in patients allergic to fish, egg, and milk. We included 11 patients with fish allergy, 5 patients with milk allergy, 7 patients with egg allergy, and 3 patients allergic to both fish and egg; all the patients had been referred to Hospital La Paz, Madrid, Spain. We investigated the effects of industrial processing of surimi on IgE recognition during 2 critical steps of manufacturing (before and after heat-induced gelation) and in the final product using ELISA, which was run using sera from a group of 56 patients (18 fish, 14 egg, 14 milk, 7 fish and egg, and 3 egg and milk). The Ethics Committee of Hospital La Paz approved the study (PI-3065), and written informed consent was obtained. Data on age, total IgE and fish-, egg,- and milk-specific IgE are presented in Supplementary Table 1.

For the ELISA experiments, we prepared 0.1 M phosphate-buffered saline (PBS)–based extracts of BES, CS, and 2 intermediate products in each (step 1, before gelation; and step 2, after gelation). PPT was performed according to European guidelines using BES and CS directly without manipulation. The PPT result was considered positive when the wheal diameter was >3 mm. BAT was performed with PBS-based extracts, using BD FastImmune CD63/CD123/HLA-DR mixture (BD Biosciences) according to the manufacturer's instructions. Samples were analyzed in a FACScan flow cytometer (BD Biosciences) [6]. The IgE reactivity of all the extracts was determined using ELISA as described elsewhere [7]. The ELISA results were evaluated using the Wilcoxon signed rank test. A 2-tailed contrast analysis was performed. Statistical significance was set at $P<.05$. The results are presented as median (IQR).

The PPT results with BES were positive in 13 fish-allergic patients (13/14; 92.8%) (6.0 [4.5-7.8]), as were those for CS (6.0 mm [4.9-9.0]), indicating the presence of fish allergens in both products. The PPT result was positive with CS in egg-allergic patients (10/10, 100%) (5.8 [4.9-9.2]), although the reaction was less pronounced with BES (2.8 [2.5-5.0]). The results are concordant with the presence of egg in the CS

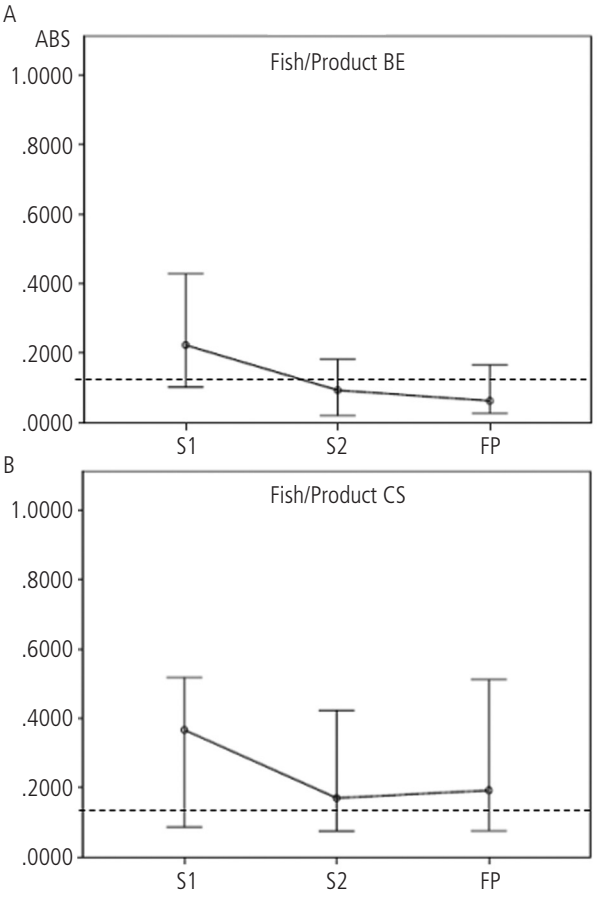

Figure. Variations in fish-specific IgE recognition during processing tested by ELISA. The Y axis represents absorbance units for fish-allergic patients tested with the different extracts. Median and error bars (95%CI) are shown. A, Sera from fish-allergic patients tested with BES. B, Sera from fish-allergic patients tested with CS. Dotted line: cut-off ≥0.14 absorbance units. ABS indicates absorbance; FP, final product; BES indicates baby eel surimi; CS, crabstick surimi; S1, step 1, before gelation; S2, step 2, after gelation.

formula but not in BES. Finally, the PPT result with BES was positive in 75% (3/4) of milk-allergic patients (12.0 [3.8-20.5]) and negative with CS (presence of milk in BES, but not in CS) (Supplementary Figure 1A).

In the BAT with the BES extract, the median %CD63$^+$ basophil value was 27.8% (8.4%-63.6%) for the 12 fish-allergic patients included. Percentages were lower for egg-allergic patients (n=9; 5.9% [0.8%-27.8%]), whereas results were positive for BES in milk-allergic patients (n=5; 22.1% [14.8%-46.7%]). The BAT results with CS were similar to those with BES for fish-allergic patients (28.7% [15.7%-71.5%]) but not for egg- and milk-allergic patients, thus reflecting the difference in their composition. The %CD63$^+$ for CS in egg-allergic patients was positive in 8/9 (88.8%; 26.9% [3.9%-58.7%]). Conversely, activation was less pronounced in milk-allergic patients with CS than with BES (n=5; 16.6% [3.0%-27.4%]) (Supplementary Figure 1B).

We used ELISA to study the variation in allergenicity during the processing of these products. Allergens were extracted from step 1 (before gelation) and step 2 (after gelation) and the final products (BES and CS). We analyzed sera from patients allergic to fish, egg, and milk. The manufacturing process led to decreased IgE recognition, although CS seems to retain more fish allergens than BES in the 3 steps studied. The reduction in IgE recognition reached statistical significance between steps 1 and 2 in BES (P<.001), and a reduction was observed in both products and in the final 2 steps (Figure, A and B). Regarding the egg allergen content, BES yielded negative results in all the steps, as this product does not include egg in its formulation (Supplementary Figure 2A). Conversely, high absorbance was observed for CS, with a significant decrease from step 1 to 2 (P=.024, Supplementary Figure 2C). We did not detect milk allergens in CS, as expected (Supplementary Figure 2D). However, the final BES product and its previous steps were strongly recognized by milk-allergic sera (P=.023; Supplementary Figure 2B).

β-Parvalbumin is a major fish panallergen that is present in most fish and recognized by 80% of allergic patients [8]. We confirmed the presence of β-parvalbumin using Western blot inhibition with recombinant mackerel β-parvalbumin (Sco j 1) before the gelation process, although its ability to recognize IgE disappeared after the gelation process (Supplementary Figure 3).

Allergenicity can be mitigated in surimi production by removal of the allergenic proteins, either during the leaching step or through structural modifications by heating during the gelation process [9-11]; hence the more pronounced fall in IgE recognition by ELISA before and after gelation and the presence of allergens in the final products.

We conclude that when used as additives, milk and egg do not lose IgE-binding ability during processing; therefore, they constitute a real danger for allergic patients. Regarding fish allergens, we observed that β-parvalbumin is undetectable in the final products, although sufficient IgE reactivity is retained. Carvalho et al [12] recently reported that a decrease in sIgE to the parvalbumin rGad c 1 and the mean wheal diameter in skin prick testing for hake and salmon can be used as markers of prognosis in the acquisition of tolerance by fish-allergic patients [12]. In line with this study, our results suggests that some fish-allergic patients could tolerate surimi products, although previous supervised oral food challenge is necessary.

Acknowledgments

We are indebted to Itsaso Losantos from the Bioinformatics core facility of the Hospital La Paz Institute for Health Research (IdiPAZ) for technical assistance. We would like to acknowledge Natalia Canales for technical support.

Funding

This research was funded by Spanish Ministry of Science and Innovation (CDTI-CIEN Tolera grant). The funders had no role in any of the following: study design; data collection, analyses, or interpretation; writing of the manuscript; decision to publish the results.

Conflicts of Interest

JLH works at the Angulas Aguinaga Research Center. The remaining authors declare that they have no conflicts of interest.

References

1. Ching LH, Chopin C, Mastail M, Keromnes B. Process for producing deboned, washed and refined fish meat, usually called surimi, and obtained products. Patent application PCT W09I.03950; 1991.
2. Rawdkuen S, Benjakul S. Whey protein concentrate: Autolysis inhibition and effects on the gel properties of surimi prepared from tropical fish. Food Chemistry. 2008;106:1077-84.
3. Jafarpour A, Hajiduon HA, Aie M. A comparative study on effect of egg white, soy protein isolate and potato starch on functional properties of common carp (Cyprinus Carpio) Surimi Gel. Food Process Technol. 2012;3:190-6.
4. Hebling A, Lopez M, Lehrer SB. Fish allergy: is it a real problem with surimi-based products? Int Arch Allergy Immunol. 1992;99:452-5.
5. Mata E, Favier C, Moneret-Vautrin DA, Nicolas JP, Han CL, Gueant JL. Surimi and native codfish contain a common allergen identified as a 63-kDa protein. Allergy. 1994;49:442-7.
6. MacGlashan DW Jr. Basophil activation testing. J Allergy Clin Immunol. 2013;132:777-87.
7. Ventura MT, Rodriguez-Perez R, Caballero ML, Garcia-Alonso M, Antonicelli L, Asero R. IgE, IgG1, and IgG4 response to specific allergens in sensitized subjects showing different clinical reactivity to Anisakis. Eur Ann Allergy Clin Immunol. 2017;49:52-8.
8. Ruethers T, Taki AC, Johnston EB, Nugraha R, Le TTK, Kalic T, et al. Seafood allergy: A comprehensive review of fish and shellfish allergens. Mol Immunol. 2018;100:28-57.
9. Sonu S. Surimi. NOAA Technical Memorandum National Marine Fisheries Service (NMFS), Terminal Island, 1986.
10. Nishioka F. Leaching treatment. In: Shimizu H, editors. Science and Technology of Fish Paste Products. Tokyo: Koseisha-Koseikaku Publishing Co, 1984;62-73.

11. Kourani E, Corazza F, Michel O, Doyen V. What Do We Know About Fish Allergy at the End of the Decade? J Investig Allergol Clin Immunol. 2019;29:414-21.
12. Carvalho S, Marcelino J, Cabral Duarte MF, Costa C, Barbosa MA, Pereira Dos Santos MC. Role of Recombinant Parvalbumin Gad c 1 in the Diagnosis and Prognosis of Fish Allergy. J Investig Allergol Clin Immunol. 2020;30:340-5.

Manuscript received November 26, 2020; accepted for publication January 27, 2021.

María Pedrosa
La Paz Hospital Institute for Health Research, IdiPaz,
Despacho 1.1
Paseo de la Castellana, 261
28046 Madrid, Spain
E-mail: m.pedrosa.d@gmail.com

