Custard Apple Allergy with Glycosyltransferase as the Allergen Involved

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The Custard Apple, also known as Cherimoya, is a tropical fruit that belongs to the *Annonaceae* family. Although it originates from Central America, Spain is the leading producer in Europe. Its consumption is increasing in recent years due to the nutritional composition of this fruit, its content in specific phytochemical compounds and its impact on health benefits [1]. Nevertheless, hypersensitivity to cherimoya is infrequent [2]. To date, few allergy cases have been reported involving different allergens and most of them are related to cross-reaction with latex due the class I chitinases proteins [3].The first report of custard apple allergy revealed the presence of a 20-25 kDa protein as the culprit [1], and the subsequent studies detected cross-reactivity between latex and cherimoya involving allergenic proteins around 40-45 kDa [2.4,5]. The most recent case report describes an IgE-binding band of 14 kDa that displays cross-reactivity with latex, papaya and avocado as a new allergen involved in cherimoya and latex cross-reaction [3].

We present a 11-year old child with the diagnosis of rhinoconjunctivitis due to *Cupressus arizonica* pollen, that during the previous years developed two episodes of oral allergy syndrome after custard apple ingestion. She presented no symptoms with latex and any other fruits, including peach, avocado, papaya and banana.

Skin prick test (SPT) with commercial extracts of latex, common inhalants, palm pollen profilin (Pho d 2), peach Lipid Transfer Protein (LTP) and prick-by-prick with fresh custard apple (peel and pulp) were performed. SPT were positive (mean wheal diameter

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≥ 3 mm) only to *Cupressus arizonica* pollen and to custard apple (peel and pulp). Total and specific immunoglobulin (Ig) E were assessed using the CAP system (Phadia). Total IgE 859kU/L. sIgE test against Pru p 3, profilin, latex and latex recombinants proteins (rHeb v 1, rHeb v 3, rHeb v 5, and rHeb v 6.01) were negative (<0.1kU/L).

Protein extracts from cherimoya pulp and peel (CPE and CLE, respectively)were prepared by homogenization in phosphate-buffered saline, followed by centrifugation and dialyzation. Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) IgE immunoblotting assays revealed IgE reactivity with a band of 50 kDa (Figure 1A) in the CLE. The protein/s from CLE were separated on a series of two-dimensional gel electrophoresis (2D)run with a pH gradient of 4 to 8 (Figure 1B) and, subsequently, transferred to a nitrocellulose membrane for incubation with a serum from allergic patient. A single protein from CLE with a molecular weight of approximately 50 kDa was recognized by IgE with a point isoelectric protein (pI)5-5.8 (Figure 1C). The 50kDa IgE binding spot was manually excised from the Coomasie blue-stained gel for tryptic digestion according to Shevchenko et al [6]. After digestion, peptide mass fingerprinting (PMF) was analyzed by MALDI-TOF MS for protein identification as previously described [6].Additionally, when available and for confirmation of protein identify, peptide fragmentation was performed by MS in tandem MALDI LIFT TOT/TOF. Based on comparison with a database, the resulting peptides were analyzed using mass spectrometry and tandem mass spectrometry, which revealed a high homology with the glycosyltransferase of Capsicum baccatumin the CLE.

Glycosyltransferases comprise a large family of enzymes that catalyze glycosidic bond formation using sugar donors containing a nucleoside phosphate or a lipid phosphate leaving group to a variety of acceptor molecules, including proteins, lipids, polysaccharides, nucleic acids and small organic molecules [7]. As result, these

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enzymes located in the Golgi apparatus generate a significant and diverse amount of glycoconjugates that are important for biological processes, such as cell signaling, cell-pathogen interactions, and the maintenance of the cellular architecture and metabolism [7].Among glycosyltransferases, the enzyme beta1,2-xylosyltransferase adds xylose to glycoprotein, and may play a critical role in allergenicity by regulating the structure of the oligosaccharide chains and the targeting of these proteins to various organelles such as storage bodies [8].*In vitro* reports have shown that IgE from sera of allergic individuals recognize core alpha1-3-fucosyltransferase in the prostate glands of snails, and it has been suggested that this protein could be allergenic [9].Subsequent studies with demonstrated that complex plant *N*-glycans constitute a major class of the so-called carbohydrate cross-reactive determinants reactive with IgE antibodies in the sera of many allergic patients [10]. However, its real implication in clinical practice has not been demonstrated to date.

We report a case of IgE-mediated allergy to custard apple with a glycosyltransferase as the allergen involved. It is important to keep in mind that theoretically low-allergenic foods are always likely to cause problems. In addition, when facing an allergic patient, we must assess different allergenic sources other than those already known, in order to be able to make the appropriate avoidance recommendations.

Conflicts of interest

The authors have no conflict of interest to declare.

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References

1. Gonzalo MA, Moneo I, Ventas P, Polo F, García JM. IgE-mediatedhypersensitivity to custard-apple. Allergy. 1997 May;52(5):597.
2. Sánchez-Guerrero IM, Escudero AI, Tortosa JA, Lombardero M. Anaphylaxis to cherimoya. Allergy. 2000 Oct;55(10):976-7.
3. Santos-Magadán S, Bartolomé-Zavala B, Rodríguez-Jiménez B, González-de-Olano D, Heras-Mendaza F, Conde-Salazar L. Custardapple and latexallergy: a new type of cross-reaction?J Investig Allergol Clin Immunol. 2012;22(3):223-4.
4. Gamboa PM, Sánchez-Monge R, Díaz-Perales A, Salcedo G, Ansótegui J, Sanz ML. Latex-vegetable syndrome due to custard apple and aubergine: new variations of the hevein symphony. J Investig Allergol Clin Immunol. 2005;15(4):308-11.
5. Sánchez-Morillas L, Moneo I, Sedano E, Iglesias A, Caballero ML, Rodríguez M. Oral allergic syndrome after ingestion of custard apple. Allergy. 2003 Mar;58(3):260-1.
6. Martínez-Alonso E, Alcázar P, Camafeita E, Fernández-Lucas M, Ruiz-Roso G, Alcazar A. Proteomic analysis of plasma proteins of high-flux haemodialysis and on-line haemodiafiltration patients reveals differences in transthyretin levels related with anaemia. Scientific Reports. 2020;10:16029.
7. Albesa-Jové D, Cifuente JO, Trastoy B, Guerin ME. Quick-soaking of crystals reveals unprecedented insights into the catalytic mechanism of glycosyltransferases. MethodsEnzymol. 2019;621:261-279.
8. Zeng Y, Bannon G, Thomas VH, Rice K, Drake R, Elbein A. Purification and specificity of beta1,2-xylosyltransferase, an enzyme that contributes to the allergenicity of some plant proteins. J BiolChem. 1997 Dec 12;272(50):31340-7.

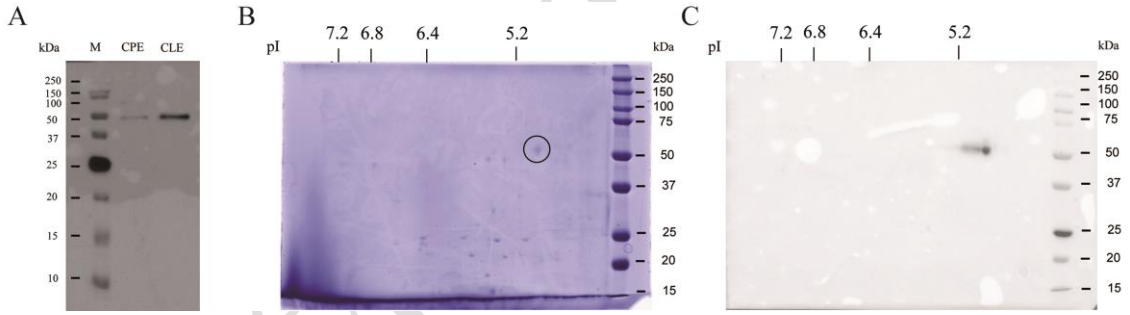
doi: 10.18176/jiaci.0711

9. van Tetering A, Schiphorst WE, van den Eijnden DH, van Die I. Characterization of a core alpha1-->3-fucosyltransferase from the snail Lymnaeastagnalis that is involved in the synthesis of complex-type N-glycans. FEBS Lett. 1999 Nov 19;461(3):311-4.

10. Strasser R, Mucha J, Mach L, Altmann F, Wilson IB, Glössl J, Steinkellner H.Molecular cloning and functional expression of beta1, 2-xylosyltransferasecDNA from Arabidopsis thaliana. FEBS Lett. 2000 Apr 21;472(1):105-8.

Figure Legend

Figure 1. In vitro studies with custard apple extract



A) Inmunoblot. IgE binding to custard apple extract in serum from allergic patient. B) 2D PAGE protein profile of CPE. C) 2D inmunoblot analysis of custard apple with skin extract developed with serum of the patient. Protein spot was used for peptide mass print analysis and protein identification.
CLE: cherimoya peel extract, CPE: cherimoya pulp extract, kDa: kilodalton, M: molecular weight marker.

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