References

1. Frey L. Le syndrome du nerf auriculo-temporal. Rev neurol. 1923; 2:97.
2. Escudero-Canto MC, Cuartero-del P, I, Ruiz-Cano R, Balmaseda-Serrano E, Gil- delivery. Auriculotemporal nerve syndrome in children secondary to a forceps delivery. Rev Neurol. 2007; 44:186.
3. Elliott RM, Weinstein GS, Low DW, Wu LC: Reconstruction of Complex Total Parotidectomy Defects Using the Free Anterolateral Thigh Flap: A Classification System and Algorithm. Ann Plast Surg. 2011 May;66(5):429-37.
4. Singh N, Kohli M, Kohli H. Innovative Technique to Reduce Incidence of Frey's Syndrome after Parotid Surgery. Am Surg. 2011;77:351-4.
5. Thoma-Uszynski S, Mahler V. Incomplete auriculotemporal nerve syndrome--mimicry of oral allergy syndrome. Eur J Dermatol. 2007;17:157-9.
6. Martínez-Baylach J, Aragó T, Galdós H, Herrera C, Rubio de Abajo I. Frey's sydrome secondary to an obstetrics trauma: Presentation of 2 cases and a review of the literature. An Pediatr (Barc). 2010 Apr;72(4):272-7.
7. Sethuraman G, Mancini AJ. Familial auriculotemporal nerve (Frey) syndrome. Pediatr Dermatol. 2009 May-Jun;26(3):302-5.
8. Listernick R, Legius E, Charrow J. Gustatory flushing (auriculotemporal nerve syndrome) in children with neurofibromatosis type 1 and facial plexiform neurofibromas. J Pediatr. 2011 Jun;158(6):1034-34
9. Farman M, Zaitoun H. Auriculotemporal nerve syndrome in association with congenital haemangiopericytoma: a case report. Eur J Paediatr Dent. 2010 Dec;11(4):213-5.
10. Sampson HA. Differential diagnosis in adverse reactions to foods. J Allergy Clin Immunol. 1986;78:212-219.

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Anaphylaxis Due to Orange Soft Drinks

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Citrus sinensis is a tree that belongs to the Rutaceae family; its fruit, the sweet orange, is widely consumed throughout Europe. Valencia, which is the largest producer of sweet oranges in Spain, is one of the most important orange-growing areas in the world. Spain alone produced 5.7 million tons of this fruit in 2011. Sweet orange is not considered to be a common allergenic fruit, although 3 allergens have been described to date: Cit s 1 (germin-like protein), Cit s 2 (profilin), and Cit s 3 (nonspecfic lipid transfer protein) [1,2]; this last allergen has been found to show cross-reactivity with the major peach allergen Pru p 3 [3]. However, no cases of allergy to orange soft drinks have been reported in the literature.

We report the case of a 23-year-old woman who experienced an episode of anaphylaxis 10 minutes after handling sweet oranges in a fruit warehouse (an orange storage network), where she had been working for a month. Because of this episode, she left the job and some months later experienced contact urticaria when squeezing an orange; she also experienced oral allergy syndrome after drinking several orange soft drinks (Sunny Delight and TriNa orange) that she had previously tolerated.

Surprisingly, despite living in Valencia, she had not eaten sweet oranges for 15 fifteen years as she did not like them. She denied having clinically allergic rhinitis or asthma and tolerated various types of fruits, including citric fruit such as lemon, which she tolerated in ice cream and juice form before and after the allergic episodes described above.

Skin prick tests (SPTs) were negative for mites, fungi, and pollen (*Parietaria judaica*, *Olea europaea*, Gramineae family, *Cupressus arizonica*, *Platanus acerifolia*, and *Artemisia vulgaris*) and a prick test with profilin from *Gramineae* pollen was also negative. Extracts from sweet orange pulp and peel were prepared as follows. The pulp and peel were ground into small pieces, defatted, and extracted by magnetic stirring in 50 mM of phosphate-buffered saline (PBS) at pH 7.5 for 3 hours at room temperature. The samples were centrifuged at 5600×g for 30 minutes, and the supernatants dialyzed against water and freeze-dried. The orange soft drink extracts (Sunny Delight and TriNa) were prepared by dilution in phosphate buffer (1:2), followed by magnetic stirring for 15 minutes at room temperature, dialysis, and freeze-drying. Positive SPT

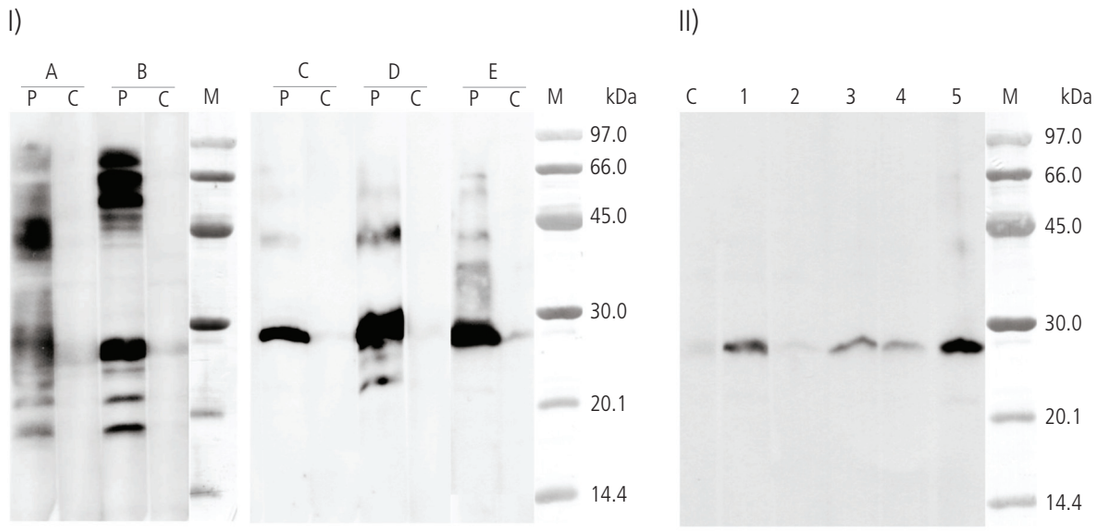

Figure. I) Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) immunoblotting results under reducing conditions. A: orange peel extract; B: orange pulp extract; C: Sunny Delight soft drink extract; D: TriNa soft drink extract; E: KAS soft drink extract. Lane P: patient serum (dilution 1:1); lane C: control serum (pool of sera from nonatopic individuals); lane M: molecular mass markers. II) SDS-PAGE immunoblotting-inhibition results with Sunny Delight extract in solid phase. Lane C: control serum (pool of sera from nonatopic individuals); lane 1: patient serum previously incubated with Sunny Delight extract; lane 2: patient serum previously incubated with orange pulp extract; lane 3: patient serum previously incubated with TriNa extract; lane 4: patient serum previously incubated with KAS extract; lane 5: patient serum previously incubated with lamb extract; lane M: molecular mass markers.

results were obtained for the pulp extract (4 × 4 mm) and the peel extract (44 × 5 mm); the results for similar tests with lemon pulp and peel extracts (all at 10 mg/mL) were all negative. The SPTs with the soft drink extracts were negative but the corresponding intradermal tests were positive in both cases (>64 × 6 mm). Furthermore, 1 hour after the intradermal test with orange extract (pulp and peel), the patient developed facial edema, dyspnea, and wheezing that required urgent medical attention.

Total serum immunoglobulin (Ig) E (UniCAP, Phadia) was 6.7 IU/mL and serum specific IgE by means of an enzyme allergosorbent test against orange peel and pulp extracts was <0.35 kU/L (class 0).

Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) immunoblotting was performed with sweet orange pulp extract. IgE immunoblotting with the patient's serum revealed IgE-binding bands of approximately 75 kDa, 66 kDa, 57 kDa, 28 kDa, 22 kDa, and 18 kDa. The immunoblotting profile of 3 orange soft drink extracts (Sunny Delight, TriNa, and KAS) showed a strong band of 28 kDa. SDS-PAGE immunoblotting-inhibition assays showed the capacity of the orange pulp extract to inhibit the IgE binding to the Sunny Delight extract; partial inhibition was observed with the extracts from the 3 orange refreshments (Figure). This difference in IgE binding–inhibition activity must be due to the higher concentration of Cit s 1 in the orange pulp sample than in the soft drink samples.

The 28-kDa IgE binding protein was identified by MALDI-TOF (matrix-assisted laser desorption/ionization-time-of-flight mass spectrometry) and LC-ESI-MS/MS (liquid chromatography electrospray ionization tandem mass spectrometry), and by searching the NCBI non-redundant protein sequence database using the Mascot program (http://www.matrixscience.com), as previously described [4]. The resulting peptides analyzed by MS or MS/MS corresponded to Cit s 1.

To clarify the implication of this allergen, we suggested performing an oral provocation test with orange, but this was rejected by the patient due to the symptoms she had developed after the intradermal test.

Ahrazem et al [3] showed a high frequency of Cit s 1 sensitization in individuals who did not develop symptoms on consumping oranges; this was attributed to the monovalent nature of this protein based on its N-glycan epitope. For this reason, Cit s 1 has been labeled an "equivocal allergen" [5].

We have presented an interesting case of a nonatopic patient who showed clinically relevant monosensitized allergy to Cit s 1, a sweet orange allergen, which appeared as an active allergenic component in orange soft drinks. Our findings are consistent with the IgE reactivity exhibited by Cit s 1 in heat-processed orange juice described by Crespo et al [1].

To conclude, we believe that the clinical relevance of Cit s 1 should be studied more carefully, with an independent investigation of each case.

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References

1. Crespo JF, Retzek M, Foetisch K, Sierra-Maestro E, Cid-Sanchez AB, Pascual CY, Conti A, Feliu A, Rodriguez J, Vieths S, Scheurer S. Germin-like protein Cit s 1 and profilin Cit s 2 are major allergens in orange (Citrus sinensis) fruits. Mol Nutr Food Res. 2006 Mar;50(3):282-90.
2. López-Torrejón G, Ibáñez MD, Ahrazem O, Sánchez-Monge R, Sastre J, Lombardero M, Barber D, Salcedo G. Isolation, cloning and allergenic reactivity of natural profilin Cit s 2, a major orange allergen. Allergy. 2005 Nov;60(11):1424-9.
3. Ahrazem O, Ibáñez MD, López-Torrejón G, Sánchez-Monge R, Sastre J, Lombardero M, Barber D, Salcedo G. Orange germin-like glycoprotein Cit s 1: an equivocal allergen. Int Arch Allergy Immunol. 2006;139(2):96-103.
4. Pastor C, Cuesta-Herranz J, Cases B, Pérez-Gordo M, Figueredo E, de las Heras M, Vivanco F. Identification of major allergens in watermelon. Int Arch Allergy Immunol. 2009;149(4):291-8.
5. Pöltl G, Ahrazem O, Paschinger K, Ibañez MD, Salcedo G, Wilson IB. Molecular and immunological characterization of the glycosylated orange allergen Cit s 1. Glycobiology. 2007;Feb;17(2):220-30.

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Nasal Challenge Test in the Diagnosis of Latex-Related Systemic Reactions

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Palabras clave: Anafilaxia. Prueba de provocación nasal. Látex.

Natural rubber latex (NRL) allergy is recognized as a major health problem. Around 12% of allergic reactions occurring during anesthesia are caused by latex [1], although the incidence of latex allergy seems to have decreased in recent years [2]. None of the available diagnostic tests has 100% sensitivity.

We report the case of a 30-year-old woman who experienced anaphylactic shock during a myomectomy under general anesthesia (serum tryptase, 18.6 ng/mL at 1 hour after the reaction). The patient had a personal history of mild rhinitis and house dust mite sensitization, but neither drug-related nor food-related allergic reactions had been reported. She worked as a dental assistant and had never experienced problems with NRL gloves or occupational respiratory symptoms.

In vivo study: The results of skin prick tests (SPT) and intradermal tests with the drugs used during anesthesia (propofol, fentanyl, midazolam, cisatracurium, and cefminox) were all negative. We performed an intramuscular challenge test with cefminox, although no allergic reaction was observed. SPT using commercial latex extract (100 IR/mL, Stallergènes) was not conclusive (wheal 3 × 3 mm without erythema). Therefore, to confirm the suspected NRL allergy, we performed a prick-by-prick test with a latex glove, a rubbing test, and a glove use test, although the results were negative in all cases. In order to rule out a possible lack of efficacy of the first commercial latex extract, SPT was repeated using another commercial extract with a known latex protein concentration of 0.5 mg/mL (ALK-Abelló). We tested several dilutions (1/100 weight/volume [w/v], 1/10 w/v, and 1/1 w/v), although only the undiluted SPT (0.5 mg/mL) was slightly positive, with a wheal of 4 × 4 mm (histamine, 9 × 8 mm). We mimicked mucosal allergen exposure by performing a nasal challenge test (NCT) with the NRL extract used in the second SPT. The challenge was performed according to guidelines [3] and monitored using acoustic rhinometry. Although SPT had only been positive at 1/1 w/v, NCT was performed at 2 different concentrations (0.5 mg/mL and 0.05 mg/mL). The result was positive (decrease of >25% in nasal volume between 0 and 7 cm) only at 0.5 mg/mL. No systemic reactions were observed. A healthy control was also tested after providing written informed consent, with negative results. A thorough investigation into food sensitization related to NRL allergy (banana, kiwi, avocado, and chestnut) was negative (both history and SPT).

In vitro study: Specific immunoglobulin (Ig) E to NRL