What Is the Limit? Anaphylaxis After Intake of a Whey Sport Supplement Shake by a Cow's Milk–Tolerant Patient

Villalobos-Vilda C[1]*, Lendínez MA[2]*, Lorente-Sorolla C[2], Rodrigo-Muñoz JM[2,3], Del Pozo V[2,3]
[1]*Department of Allergy, Fundación Jiménez Díaz, Madrid, Spain*
[2]*Department of Immunology, Instituto de Investigación Sanitaria (IIS) Fundación Jiménez Díaz, Madrid, Spain*
[3]*CIBER de Enfermedades Respiratorias (CIBERES), Madrid, Spain*
[*]*Both authors contributed equally as first authors.*

J Investig Allergol Clin Immunol 2023; Vol. 33(3): 228-229
doi: 10.18176/jiaci.0836

Key words: Anaphylaxis. Cow's milk. Whey protein. Immunotherapy. Sport supplementation.

Palabras clave: Anafilaxia. Leche de vaca. Suero de leche. Inmunoterapia. Suplemento deportivo.

Cow's milk protein allergy (CMPA) is one of the most common food allergies worldwide. It is a frequent disease in childhood and usually appears in the first 2 or 3 months of breastfeeding supplemented with artificial formula. Prevalence ranges between 0.5% and 3% in children under 1 year of age [1], with most cases resolving spontaneously. Cow's milk (*Bos domesticus*) is composed of lactose, simple lipids, and proteins, of which 80% are casein and 20% whey proteins (eg, β-lactoglobulin [BLG], α-lactalbumin [ALA], bovine serum albumin [BSA]) [2].

We report the case of a 20-year-old man with a history of CMPA treated with oral immunotherapy (OIT) from age 4 to 6 years who currently tolerates daily doses of 200-300 mL (7-8 g) of cow's milk and presented with an episode of anaphylaxis after ingesting a whey-rich protein shake (composed of 30 g of whey protein supplement [WPS], creatine monohydrate, and 200 mL of milk). Ten minutes after taking the mix, the patient developed epigastric pain, generalized urticarial lesions, facial angioedema, and rhinoconjunctivitis, as well as dyspnea, which began after 40 minutes. The clinical manifestations were managed with adrenaline (0.5 mg), methylprednisolone (60 mg), and dexchlorpheniramine (5 mg), and the symptoms resolved in less than 1 hour. Since then, the patient has continued to consume dairy products at the doses stated above, avoiding ingestion of the shake that caused the symptoms, without developing new episodes. The patient gave his informed consent to participate in this study.

He was sent to the allergology department, where he denied the influence of cofactors such as physical exercise and anti-inflammatory drugs. He also commented that 20 minutes before this episode he had had a meal based on vegetable cream, gilthead bream, strawberries, and nuts (all of which were previously tolerated).

Skin tests were performed with milk, the components of the protein shake (milk, WPS, and creatine monohydrate), the abovementioned foods, and a commercial *Anisakis* extract. A very positive prick-prick result was recorded for the commercial milk extract, casein, ALA, BLG, BSA, and WPS, with negative results for the remaining allergens.

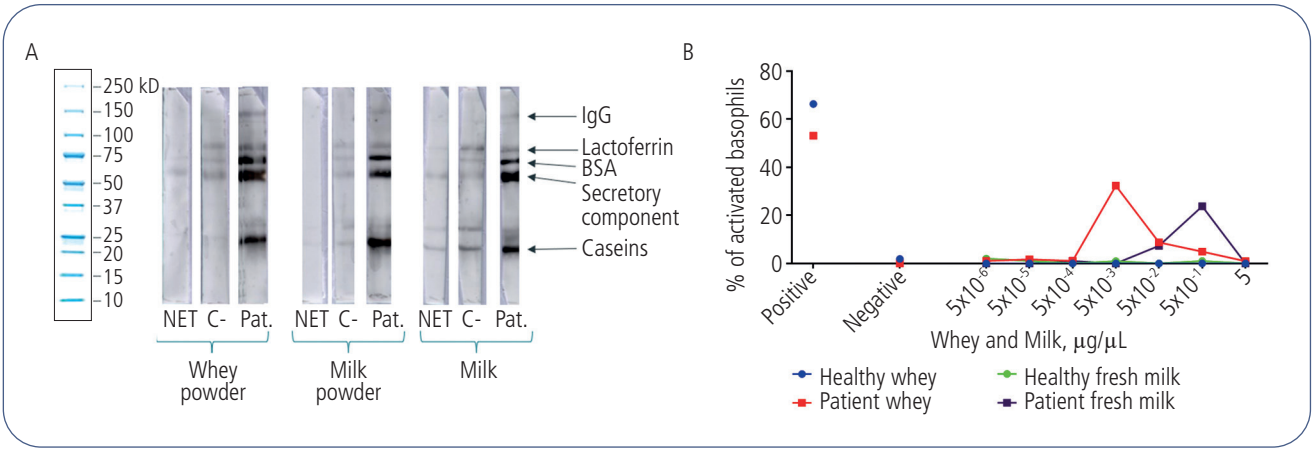

Figure. A, Western blot results. Electrophoresis performed with SDS-PAGE under nonreducing conditions. Milk and supplement were dissolved in PBS1X. Binding of the immune complexes was performed using anti-IgE-HRP. B, Basophil activation test performed with whey protein supplement and fresh milk. The blood of a patient and the blood of a healthy control were confronted with decreasing concentrations of milk and whey protein powder. The result was considered positive with activation ≥15% (as established by the protocol). Results are expressed as the percentage of $CD63^+$ basophils (activated basophils). NET indicates Net 1X (negative technique control); C-, control serum; Pat, patient serum.

© 2023 Esmon Publicidad

Given these findings, we performed prick-to-prick tests with serial dilutions of milk and WPS, obtaining positive results for doses ranging from 5 mg/mL to 0.1 mg/mL. Likewise, 2 in vitro studies were carried out (Western blot and basophil activation test [BAT]).

Specific IgE (sIgE) to milk components was assessed using ImmunoCAP (Thermo Fisher Scientific), considering values >0.35 kU/L as positive. Values were also recorded for total IgE (25.5 kU/L), cow's milk sIgE (0.47 kU/L), BSA (0 kU/L), ALA (0.38 kU/L), BLG (0.35 kU/L), and casein (0.04 kU/L). The results were significantly lower than those previously recorded (Supplementary Table 1).

Western blot revealed a positive response by the patient's serum to various proteins from both the supplement and milk (Figure, A), which by molecular weight could correspond to lactoferrin (80 kDa), BSA (66 kDa), secretory component (≈60 kDa), caseins (21-27 kDa), and IgG (≈155 kDa).

BAT was performed using the BasoFlowEx reagent kit (EXBIO) following the manufacturer's protocol. The basophil population was defined as $CD203c^{+}/SSC^{low}$ by flow cytometry. As shown in the Figure (B), maximum activation was reached after stimulation with 0.5 µg/µL of whey protein powder (24% activation) and with 0.005 µg/µL of milk (33% activation).

As mentioned above, skin-prick testing with milk and WPS was positive at very low doses (0.1 mg/mL), suggesting a high degree of sensitization. Regarding immunoblotting, the possible proteins against which the patient's serum reacted were caseins, BSA, lactoferrin, secretory component, and whey protein, as described in the literature [3].

Furthermore, BAT yielded a positive result for both WPS and whole milk. It is striking that the concentrations at which maximum activation is achieved are higher for milk (0.5 µg/µL) than for WPS (0.005 µg/µL). Although graphs from various studies that show the degree of basophil activation in patients with CMPA is heterogeneous, since basophil reactivity differs from patient to patient [4,5], sensitized people's curves grow as the concentration of the allergen increases. Our BAT results, which were recorded in a clinically asymptomatic but sensitized patient, are similar to those recorded for an allergic person. The absence of conclusive results leads us to hypothesize that our findings could be the result of OIT, with the patient currently tolerating higher doses than recommended in the SEAIC immunotherapy guideline [6], or of basophil inactivation due to exposure to high doses of the allergens involved (inhibition by overactivation).

According to the results, the patient we report has sensitivity to milk without clinical repercussions if the dose is 200-300 mL; however, he develops an anaphylactic reaction if he ingests milk protein at higher amounts, eg, in a sport supplement shake.

In conclusion, the patient may have developed the anaphylactic reaction because the ingested dose of whey protein (24 g per serving [serving = 31 g]) is much higher than the usual tolerated amounts of milk (200 mL/d). The results of our extensive laboratory evaluation support the notion that OIT is a process of desensitization that does not predictably result in tolerance; it is not clear that patients with negative skin prick test results, undetectable sIgE, and nonreactive BAT results are equivalent to those who acquire natural tolerance. However, in the present case, the patient did achieve desensitization successfully, and even maintains tolerance to CMP after the reported episode.

To our knowledge, this is the first clinical case of anaphylaxis without cofactors due to ingestion of a sports supplement in a patient with CMPA. The condition was resolved by immunotherapy, consistent with findings reported elsewhere [7]. Given that the anaphylactic reaction seems to be associated with the dose, it would be necessary to carry out a review that offers conclusive recommendations on the maximum tolerated dose of CMP suitable for patients treated with OIT. It would be interesting to warn affected patients of the possible risks of taking sports supplements based on hydrolyzed whey protein.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

VdP reports having served as a consultant for AstraZeneca and GSK and having been paid lecture fees by AstraZeneca and GSK. She is also Associate Editor of JIACI. The remaining authors declare that they have no conflicts of interest.

References

1. Flom JD, Sicherer SH. Epidemiology of Cow's Milk Allergy Gastrointestinal Symptoms. Nutrients. 2019;11(1051):2-14.
2. Linhart B, Freidl R, Elisyutina O, Khaitov M, Karaulov A, Valenta R. Molecular approaches for diagnosis, therapy and prevention of Cow´s milk allergy. Nutrients. 2019;11(7):1-24.
3. Farrell H, Jimenez-Flores R, Bleck G, Brown E, Butler J, Creamer L, et al. Nomenclature of the Proteins of Cows' Milk—Sixth Revision. J Dairy Sci. 2004;87(6):1641-74.
4. Paranjape A, Tsai M, Mukai K, Hoh RA, Joshi SA, Chinthrajah RS, et al. Oral Immunotherapy and Basophil and Mast Cell Reactivity in Food Allergy. Front Immunol. 2020;11(December):1-12.
5. Ruinemans-Koerts J, Schmidt-Hieltjes Y, Jansen A, Savelkoul HFJ, Plaisier A, van Setten P. The Basophil Activation Test reduces the need for a food challenge test in children suspected of IgE-mediated cow's milk allergy. Clin Exp Allergy. 2019;49(3):350-6.
6. Martorell A, Alonso E, Echeverría L, Escudero C, García-Rodríguez R, Blasco C, et al. Oral immunotherapy for food allergy: A Spanish guideline. Immunotherapy egg and milk Spanish guide (ITEMS guide). Part I: Cow milk and egg oral immunotherapy: Introduction, methodology, rationale, current state, indications, contraindications, and oral immunotherapy build-up phase. J Investig Allergol Clin Immunol. 2017;27(4):225-37.
7. Sousa MJCS, Ferreira ALR, da Silva JPM. Bodybuilding protein supplements and cow's milk allergy in adult. Eur Ann Allergy Clin Immunol. 2018;50(1):42-4.

Manuscript received May 26, 2022; accepted for publication June 10, 2022.

Victoria del Pozo
Hospital Universitario Fundación Jiménez Díaz
Avenida de los Reyes Católicos 2
28040 Madrid, Spain
E-mail: VPozo@fjd.es

 © 2023 Esmon Publicidad