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Case report

Allergic Reactions to Lysozyme: a Case Report and Literature Review

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SUMMARY

Although lysozyme is classified as a minor egg allergen, it can cause clinically significant allergic reactions in sensitized pediatric patients. We present the case of a child with egg allergy who developed symptoms after consuming cheeses containing lysozyme. A review of the literature indicates that lysozyme, which is also present in various foods, beverages, and medications, can trigger reactions in individuals sensitized to egg. Therefore, accurate and clear labeling is essential to avoid accidental exposure.

KEYWORDS: egg allergy, lysozyme, allergic reaction, case report

INTRODUCTION

Hen's egg allergy is one of the most common food allergies among children and infants. The major allergens reside in egg white fraction and are ovomucoid (Gal d 1), ovalbumin (Gal d 2), conalbumin (Gal d 3), and lysozyme (Gal d 4) [1]. Ovomucoid is recognized as the most allergenic protein due to its heat-stable and gastric acid-stabled properties. Ovalbumin is the main protein in egg white, and together with conalbumin, it is heat-labile, inducing reaction to raw or slightly cooked egg.

Specific IgE to lysozyme can be found in up to 35% of children with hen's egg allergy [2], but is generally not useful in diagnosing or monitoring the course of egg allergy since sIgE against egg white and ovomucoid are better predictors of hen's egg allergy [3].

CASE REPORT

G. is a 5-year-old boy and was first seen at our division when he was 9 months of age for severe atopic dermatitis, which began at 3 months. At first evaluation, his mother reported that he had not yet introduced cow's milk or hen's egg into his diet. Skin prick tests (SPTs) were positive for fresh cow's milk (12 mm) and hen's egg (10 mm), and blood tests confirmed a marked sensitization to milk (> 100 kUa/L), with casein 36.1 kUa/L, and egg (> 100 kUa/L), with ovalbumin >100 kUa/L and ovomucoid 82.9 kUa/L.

Desensitization for milk was initiated and completed by the age of 4, after which he was able to consume cow's milk and dairy products without any reactions. Table I shows the trend of specific IgE levels for cow's milk and its main proteins from the first evaluation to the present.

At age 5, desensitization for egg was started, beginning with a dose of 0.2 ml of pasteurized raw egg white diluted 1:1,000,000, and gradually increased to a maximum dose of 2 ml of a 1:10 dilution. After this dose, he developed abdominal pain, oral itching, and conjunctivitis,

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TABELLA I. Evolution of specific IgE levels (kUa/L) to cow's milk and its main proteins.

IgEs	9 m	13 m	2 y	3 y	4 y	5 y	10 y
Cow's milk	>100	83.1	21.6	9.06	11.5	9.78	3.59
Alpha-lactalbumin	0.75 kUa/L	1.52	8.12	1.22	4.68	4.33	1.72
Beta-lactoglobulin I	>100 kUa/L	82.6	27.8	11.2	6.78	3.34	3.38
Casein	36.1 kUa/L	44.5	10.2	5.76	6.36	2.66	1.9

m: month; y: year.

TABELLA II. Evolution of specific IgE levels (kUa/L) to egg white and its major proteins.

IgEs	9 m	13 m	2 y	4 y	5 y	8 y	10 y
White egg	>100 kUa/L	>100	>100	>100	>100	68.9	29.1
Ovoalbumin	>100 kUa/L	>100	>100	>100	>100	57.5	21
Ovomucoid	82.9 kUa/L	>100	>100	>100k	>100	32.8	14
Lysozyme					10	6.31	4.13

m: month; y: year.

which resolved after oral antihistamine administration. A second attempt was made with the administration of 1 ml of egg white diluted 1:10; however, 20 minutes later he experienced abdominal pain with vomiting, generalized urticaria, conjunctival hyperemia, and rhinitis. Intramuscular adrenaline was administered, and from that moment onward, he remained on an egg-free diet.

Table II shows the trend of specific IgE levels for egg white and its major proteins from the first evaluation to the present.

Several years later, G. experienced oral itching and facial urticaria shortly after eating pasta with tomato sauce and grated parmesan cheese at a restaurant. The symptoms resolved with antihistamine treatment, and no egg contamination was reported. A few months later, a similar reaction occurred after eating the same dish. While G. had previously tolerated Parmigiano Reggiano, his mother discovered that, on both occasions, the cheese used was Grana Padano—a hard, aged cheese that, unlike Parmigiano Reggiano, contains lysozyme as a preservative to prevent unwanted fermentation.

USE OF LYSOZYME AND CONTROVERSIES ON ITS ALLERGIC ACTIVITY

Lysozyme, also known as muramidase, is a thermolabile enzyme naturally found in egg white from which it is commonly extracted. It is a polypeptide consisting of 129 amino acids, with a molecular weight of approximately 14.3 kDa, and represents about 3.5% of the total egg protein content. Lysozyme is widely used in the food industry (particularly in dairy products), in alcoholic beverages (such as wine and beer), and in pharmaceuticals (e.g., antiseptics and nasal sprays).

From a microbiological standpoint, lysozyme is particularly effective against Gram-positive bacteria, including lactic acid bacteria. In brewing, for example, lactic acid bacteria and certain *Clostridium* species can generate undesirable compounds responsible for off-flavors.

Lysozyme can be added at various stages of the production process to limit microbial proliferation: it may be applied to raw materials such as barley and hops, to brewer's yeast before inoculation, or to the wort before or during fermentation [4].

In cheese production, lysozyme is primarily used to prevent the phenomenon known as "late blowing", which is mainly caused by the growth of *Clostridium tyrobutyricum*, a common milk contaminant. This microorganism ferments lactate present in cheese, leading to CO_2 production. The accumulation of gas during the later stages of ripening — especially in pressed cheeses — can cause structural deformations.

In winemaking, lysozyme is employed to control the growth of Gram-positive spoilage bacteria, to modulate malolactic fermentation and to stabilize wine after alcoholic or malolactic fermentation. Its use can reduce the need for high levels of sulfur dioxide and may help minimize the concentration of biogenic amines in the final product.

However, lysozyme has recently been recognized as a food allergen

due to its origin from egg. Stricter food allergen labeling regulations have led the dairy industry to develop lysozyme-free cheese varieties and have encouraged public funding of research into the health impact of food allergens.

Despite these regulatory efforts, the presence of egg-derived lysozyme is not always clearly indicated on product labels; it may appear as"lysozyme", "E1105", or simply as"preservative". As a result, individuals with egg allergies may unknowingly be exposed to trace amounts of lysozyme through various food products.

Lysozyme in dairy production

In 1992, the FAO-WHO committee approved the use of lysozyme for dairy production in the range of 10-35 g lysozyme/100 L of milk or 10-35 g lysozyme/100 kg of cheese [5].

Grana Padano is an Italian PDO (Protected Designation of Origin) hard cheese made from raw milk and aged for a long period. It is the only cheese of its kind permitted to include lysozyme, up to a maximum of 2.5 mg per 100 kg of milk.

The allergenic potential of lysozyme remains controversial. Hoffman et al [6] found that ovalbumin, ovomucoid, and ovotransferrin were the most clinically relevant egg allergens, while lysozyme showed only weak allergenicity. In contrast, Holen and Elsayed [7] demonstrated that lysozyme strongly bound to IgE in all serum samples from egg-allergic patients in their study, concluding that lysozyme should be regarded as a major egg white allergen.

In recent years, only a limited number of studies have addressed the allergenic potential of lysozyme. In a 1997 study by Fremont et al. [2], specific IgEs were measured using the CAP-RAST system in 52 patients aged 6 months to 45 years with egg allergy. Among them, 35% had anti-lysozyme specific IgEs. Moreover, 3 of 7 oral challenges with lysozyme were positive: one after ingesting 3 mg of lysozyme, and two following a labial challenge with one drop of lysozyme solution (1 mg/mL).

Similarly, in a study by Marseglia et al. [8], 54 pediatric patients allergic to egg proteins were enrolled and given either Grana Padano (containing lysozyme) or TrentinGrana (lysozyme-free), both with two different aging periods (12 and 24 months). Twenty-one children had specific anti-lysozyme IgEs, and among them, 9 experienced rash, nausea, vomiting, or abdominal pain when challenged with Grana Padano; one child reported an anaphylactic reaction. The authors found a correlation between the severity of allergic reactions and serum-specific IgE levels to lysozyme. Vomiting, hypotension, and abdominal pain were particularly associated with IgE levels higher than 7 kU/L.

Iaconelli et al. [9] studied the immunologic response to ingestion of Grana Padano cheese ripened for 18 months, containing 0.8, 2.3, and 9.3 mg of lysozyme in 5, 15, and 60 g of cheese, respectively, in 10 healthy adults and 20 subjects with egg white allergy. No allergic reactions were observed, but higher serum levels of lysozyme were found in egg-allergic individuals.

Rossi et al. [10] conducted a controlled clinical trial involving 25 adult individuals (aged 20-50 years) with confirmed egg sensitization based on positive skin prick tests. The study employed a rigorous randomized, double-blind, crossover design. Each subject consumed, with a one-week washout period between sessions, 30 grams of Grana Padano cheese (containing lysozyme) and 30 grams of TrentinGrana cheese (free from lysozyme), both aged for either 16 or 24 months. Serum concentrations of total IgA, total IgE, and specific IgE against ovalbumin, ovomucoid, and lysozyme were measured before and after ingestion. No clinically evident allergic reactions were observed following the consumption of either cheese. Furthermore, the intake of lysozyme-containing cheese did not elicit significant changes in specific IgE levels compared to baseline values. However, in lysozyme-sensitized individuals, a statistically significant correlation was identified between IgA levels and lysozyme-specific IgE, suggesting a possible translocation of the enzyme across the intestinal barrier, albeit without triggering a clinically meaningful immunological response.

Lysozyme in pharmaceutical preparations

Lysozyme is also present in certain, typically at concentrations exceeding those found in cheese. This finding further supports the notion that dietary exposure to lysozyme — at levels commonly used in cheese production — does not represent a significant clinical risk for the majority of individuals with egg allergy.

Perez-Calderon et al. described in 2007 [1] a 54-year-old woman allergic to raw egg but tolerant to cooked egg (with negative IgEs for egg white, yolk, ovalbumin, and ovomucoid; lysozyme IgEs 0.45 kUa/L). After taking a Lizipaina® tablet (3 mg of bacitracin, 2 mg of papain and 5 mg of lysozyme) for the first time, she developed dyspnea and edema of the eyelids, tongue, and lips. She experienced a similar reaction after consuming aged cheeses containing lysozyme. Skin prick tests (SPT) were clearly positive for egg white and lysozyme, and negative for yolk, ovalbumin, and ovomucoid. Subsequent oral challenge tests with bacitracin and papain were well tolerated, as was the challenge with lysozyme-free cheese. The clinical reaction was therefore very likely attributed to the presence of lysozyme in Lizipaina®.

In 2014, Infante et al. [10] reported on a 10-year-old girl with egg allergy who was already desensitized to cooked egg but still sensitized to raw egg. She experienced symptoms like oral itching and difficulty swallowing after taking Lizipaina® tablets for pharyngodynia. SPT and IgE were positive for egg white, yolk, ovalbumin, ovomucoid, and lysozyme, while sensitization to papain was ruled out (negative SPT and IgEs); allergy to bacitracin was considered unlikely, given the previous tolerance to medication containing this ingredient.

One of the most recent reports, from 2024, describe an 8-year-old boy allergic boy who, during the desensitization process for egg allergy, took a Lysopaine® tablet for pharyngodynia and immediately developed lip and tongue edema and dysphonia. The medication contained 20 mg of lysozyme, 1.5 mg of cetylpyridinium chloride, and various excipients [3] (SPT Lysopaine®: 8 mm; lysozyme IgEs: 29.4 kUa/L).

In 2020, Tan et al. described the case of a 3-year-old girl who developed

TABELLA III. The main case reports on drug allergy to lysozyme.

Author, year, journal	Age, gender	Egg	Medication	Concentration of lysozyme contained	SPT lysozyme	IgEs lysozyme	Reaction
R Pérez-Calderón et al. [11]	54 years, F	Allergic to raw eggs	• Lizipaina® • Cured cheese	5 mg	20 mm	0.45 kUa /L	Anaphylaxis: dyspnea associated with edema of the eyelids, tongue, and lips
Infante S et al. [12]	10 years, F	Allergic to raw eggs	Lizipaina®	5 mg	12 mm	/	Oral and pharyngeal pruritus, including difficulty swallowing
Elbany C. et al. [3]	9 years, M	Egg allergy in oral immu-no- therapy	Lysopaine®	20 mg	SPT Lysopaine® 8 mm	29.4 kUa /L	Edema of the lips and tongue, along with dysphonia
Tan L et al. [13]	3 years, F	Egg allergy	Leftose®	5 mg/ml	/	/	Generalized urticaria
Artesani MC et al. [14]	20 months, M	Egg not in-troduced	Narlisim®	5 mg/ml	SPT Narlisim® 8 mm	1,82 kUa /L	Anaphylaxis: rhinitis, bronchial asthma, and dysphonia
Ledesma Benítez I et al. [15]	Infant	Egg not in-troduced	Nasal decongestant	/	/	/	Anaphylaxis: urticaria, breathing difficulties and angioedema
Muramatsu R, et al. [16]	6 years, M	Egg allergy	Deodorant spray	/	/	/	Anaphylaxis: dyspnea, vomiting, urticaria in the trunk and neck area.

generalized urticaria within 30 min of the first administration of Leftose® — a mucolytic containing lysozyme [4].

Medical history included atopic dermatitis, milk-induced anaphylaxis, and egg allergy. SPT and IgEs to egg white were positive. (SPT Leftose®: 14 mm).

In 2021, an anaphylactic reaction was described in a 6-year-old boy with egg allergy following the use of a spray deodorant containing lysozyme hydrochloride. The reaction was characterized by dyspnea, vomiting, and localized urticaria on the trunk and neck [5].

Clinical adverse reactions to lysozyme have also been described in children who had not yet introduced egg into their diet. One documented case in Spain in 2007 [6] involved a newborn healthy girl who experienced an episode of anaphylaxis (urticaria, respiratory distress, and angioedema) after administration of a nasal decongestant containing neomycin, dexamethasone, chlorphenamine, and lysozyme, prior to the introduction of egg into her diet. SPT were positive for egg white, yolk, ovalbumin, and ovomucoid.

Finally, a case report from 2008 [7] described a 20-month-old boy with severe atopic dermatitis since the age of 2 months and no prior exposure to egg who developed rhinitis, respiratory distress, and dysphonia shortly after the nasal administration of Narlisim ® (containing phenylpropylamine, tonsilamine, and 500 mg of lysozyme). SPT were positive for yolk, egg white, and Narlisim® (8 mm). IgEs were elevated for yolk, egg white, ovalbumin, ovomucoid, and lysozyme (1.82 kUa/L). Table III summarizes the main case reports on drug allergy to lysozyme

Lysozyme in wine

In winemaking, lysozyme is used to control lactic acid bacteria and is considered essential for achieving consistent quality in the final product. A study by Kirschner et al. [17] investigated 5 egg-allergic individuals, 4 of whom tested positive to lysozyme by skin prick test, while one tested negative. Three of these patients underwent skin prick testing with two lysozyme-treated wines. Two patients reacted to the treated wines; however, one also reacted to the corresponding untreated (unfined) wines, limiting the interpretability of the findings. Contrary to what was stated in the application, this study did not include a double-blind placebo-controlled food challenge involving lysozyme-treated wine. Although the results confirm the presence of significant

residual lysozyme in treated wines, they do not provide conclusive evidence of clinical reactivity in egg-allergic subjects to oral consumption of such wines.

Weber et al.[18] investigated whether the use of lysozyme as an additive in wine could pose a clinical risk for egg-allergic consumers. The concentrations applied during wine treatment ranged between 250 and 500 mg/L. The researchers analyzed various samples of white and red wines treated with lysozyme, evaluating residual content using both enzyme-linked immunosorbent assay (ELISA) and high-performance liquid chromatography (HPLC).

The results revealed that residual lysozyme levels varied significantly depending on the type of wine and post-fermentation treatments. In red wines, the presence of polyphenols facilitated the removal of lysozyme, resulting in generally low residual concentrations. In contrast, in white wines, and particularly the absence of fining treatments (especially with bentonite), residual lysozyme levels could be considerably higher, reaching up to 327 mg/L. Fining with bentonite proved to be highly effective in removing lysozyme, reducing its concentration to nearly undetectable levels (< 1 mg/L).

Oral challenge tests with lysozyme-containing wine were conducted in egg-allergic individuals and did not elicit any clinical reactions. This suggests that, despite immunological sensitization, the actual clinical risk associated with the consumption of lysozyme-containing wine is very low, especially in products that undergo proper fining procedures.

The authors concluded that, under typical commercial winemaking conditions — where wines are usually fined before being released to the market — the risk to egg-allergic consumers is negligible. However, due to the potential presence of lysozyme as a hidden allergen, mandatory labeling remains justified, as established by European Directives 2003/89/EC and 2007/68/EC.

In 2011, the European Food Safety Authority (EFSA) Panel notes that published reports have documented allergic reactions to lysozyme and lysozyme-containing foods in egg-allergic individuals. However, evidence from studies involving cheeses containing lysozyme remains inconclusive, making it difficult to draw firm conclusions about the risk of allergic reactions following oral exposure.

The Panel also emphasized that allergic reactions to wine, including those involving immunological mechanisms, are well documented (Armentia, 2008; Vally and Thompson, 2003 [19,20]). Since consumers and healthcare providers may not be aware that egg-derived substances such as lysozyme are used in winemaking, allergic reactions to lysozyme may go unrecognized and underreported following wine consumption.

CONCLUSIONS

Lysozyme, an enzyme naturally present in hen's eggs, is widely used as a food additive and preservative in products such as long-ripened cheeses, wines, and pharmaceutical preparations. Although it is not commonly perceived as an allergenic protein by patients with egg allergy, lysozyme can represent a hidden source of exposure for sensitized individuals, especially in products whose lysozyme content is not always clearly indicated on the label.

The EFSA confirmed the persistent risk of allergic reactions in individuals sensitized to egg proteins, emphasizing the importance of transparent and comprehensive labeling to manage this risk. Despite ongoing debate regarding its allergenic potential, numerous clinical cases have demonstrated that ingestion of lysozyme — either in isolated form or via pharmaceutical preparations — can trigger severe allergic reactions, particularly in the presence of cofactors such as infections, which may modulate the severity of the allergic response [21].

Anaphylactic reactions related to lysozyme are rare but represent an event that should be considered when faced with unexplained allergic reactions. Moreover, these cases highlight the need for appropriate education of allergic patients and their families to ensure careful reading of labels on both food and pharmaceutical products.

In conclusion, although habitual exposure to lysozyme through foods and beverages is generally well tolerated by most egg-allergic individuals, increased awareness of its potential role as a hidden allergen is necessary.

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Author's contributions

All authors have read and approved the final version of the manuscript and agree with its submission to the journal.

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