

Opinion of the Scientific Panel on Dietetic Products, Nutrition and Allergies on a request from the Commission related to a notification from GME on fish gelatine pursuant to Article 6 paragraph 11 of Directive 2000/13/EC

(Request EFSA-Q-2004-135)

(adopted on 2 December 2004)

SUMMARY

Fish is one of the most important allergenic foods, and allergic reactions to fish can be severe. The major allergen of fish is the muscle protein parvalbumin. Gelatine is made by denaturation of collagen. Fish gelatine is used in foods and pharmaceuticals.

The applicant states that fish gelatine is produced from fish skins and bones according to existing European regulations. No analytical data regarding possible residual levels of the major fish allergen parvalbumin in fish gelatine preparations are provided. No consumer exposure data specific for fish gelatine are given by the applicant. However, exposures of up to about 10 g per serving may occur under certain conditions of use.

In a double-blind placebo-controlled food challenge (DBPCFC) study of 30 patients with clinical allergy to fish, one patient reacted to 7.6 g of fish gelatine.

The scientific data provided by the applicant are insufficient to predict the likelihood of adverse reactions in fish allergic individuals. Nevertheless, the Panel considers that fish gelatine could cause an allergic reaction in fish allergic individuals under certain conditions of use.

Appropriate analytical methods to determine residual levels of parvalbumin in fish gelatine preparations are needed. More data from clinical challenge studies with gelatine of fish allergic individuals having specific IgE or positive skin prick tests to fish collagen or gelatine are needed.

KEY WORDS

Fish gelatine, food allergy.

BACKGROUND

In November 2003, the European Parliament and the Council adopted Directive 2003/89/EC¹ amending Directive 2000/13/EC, as regards indication of the ingredients present in foodstuffs.

Annex IIIa of the Directive specifies a list of ingredients that are known to trigger allergic reactions or intolerances for which no labelling exemptions are allowed. Whenever the listed ingredients or their derivatives are used in the production of foodstuffs, they must be labelled.

¹ Directive 2003/89/EC of the European Parliament and of the Council amending Directive 2000/13/EC as regards indication of the ingredients present in foodstuffs. OJ L 308. 25.11.2003, p. 15.

Article 1, paragraph 11 of the Directive establishes a procedure allowing for temporary labelling exemption of derivatives from ingredients listed in Annex IIIa for which it has been scientifically established that it is not possible for them to cause adverse reactions. In accordance with this provision, submissions of request for temporary labelling exemption were notified to the Commission before 25 August 2004. The Commission shall, not later than 25 November 2004, and after consultation with the European Food Safety Authority, adopt a list of those ingredients which shall be temporarily excluded from Annex IIIa, pending the final results of the notified studies, or at the latest until 25 November 2007. Therefore, the European Food Safety Authority is asked to provide scientific opinions on the submissions in accordance with the present terms of reference.

TERMS OF REFERENCE

In accordance with Article 29 (1) (a) of Regulation (EC) N° 178/2002, the European Commission requests the European Food Safety Authority to evaluate the scientific data submitted by GME (the Gelatine Manufacturers of Europe) in the framework of the procedure laid down for temporary labelling exemptions in Article 6 paragraph 11 of Directive 2000/13/EC. On the basis of that evaluation, EFSA is requested to issue an opinion on the information provided, and particularly, pending the final results of the studies undertaken, to consider the likelihood of adverse reactions triggered in susceptible individuals by the consumption of the following ingredients/substances used under the conditions specified by the applicant: Fish gelatine.

ASSESSMENT

1. Manufacturing process

Fish gelatine is produced by extraction and hydrolysis of fibrous, insoluble collagen from skins and bones. It is manufactured from fish raw materials in compliance with the requirements of the Commission Decision 1999/724/EC. The manufacturing process includes treatment with acid and/or alkali, followed by one or more rinses, pH adjustment, extraction by heating one or several times in succession, followed by purification by means of filtration and/or ion exchange and sterilisation. Further details are not given.

2. Characterisation of the products and their use

2.1 Characterisation of fish gelatine

Gelatine is denatured collagen, modified by the separation of three alpha chains comprising the triple helix of collagen and by some degree of hydrolysis of the resultant alpha chain polypeptides. The applicant states that according to Commission Decision 1999/724/EC gelatine is a natural, soluble protein, gelling or non-gelling, obtained by the partial hydrolysis of collagen produced from bones, hides and skins, tendons and sinews of animals (including fish and poultry). For the purpose of the present application, gelatine is produced exclusively from fish raw materials.

2.2 Conditions of use and exposure levels

2.2.1 Conditions of use

Gelatine is a widely applied protein in foods and pharmaceuticals. Fish gelatine has different technological properties compared to beef and pork gelatine. Gelatine used directly by the consumers is applied by oral consumption.

2.2.2 Exposure levels

According to the applicant, the average intake of all gelatines (mammalian and fish) in Germany is about 300 g per individual per year. This is said to be valid for Europe as well, which has the highest gelatine consumption worldwide. No data specific for fish gelatine are provided.

No information on specified conditions of use is provided by the applicant. Under certain conditions of use, exposure to fish gelatine is up to about 10 g per serving (NDA, 2004a).

3. Evidence of non-allergenicity

3.1 History of non-allergenicity of the product

3.1.1 Literature search strategy

No literature search is reported in the application.

3.1.2 Historical evidence of safe use

This issue is not raised by the applicant.

3.2 Laboratory-based tests

3.2.1 Residual amounts of known fish allergen (parvalbumin) in gelatine

The major fish allergen is the protein parvalbumin (NDA, 2004b), a protein structurally different from collagen and gelatine. At least 14 fish parvalbumins have been described, and mostly they have been isolated from fish muscle. However, there is one report in which parvalbumin was detected in the brain and kidney, and it is uncertain whether parvalbumin has been looked for in fish skin and fish scales. Other minor fish allergens have also been reported.

The applicant states that the extensive production processes eliminate the allergens detected in fish from the final gelatine. However, no analytical data are provided to support this claim regarding residual levels of parvalbumin. Moreover, the allergenicity of fish parvalbumin is not easily destroyed by heat, proteolytic activity or denaturation with chemicals.

3.2.2 Allergenicity of fish gelatine

3.2.2.1 Serological and immunochemical studies of fish collagen and gelatine

3.2.2.1.1 Collagen and gelatine

Experimental studies (Hamada *et al.*, 2001 and 2003) found that the IgE reactivity of fish collagen was very thermostable and was preserved also in peptide fragments. When collagen was denatured to gelatine by heating in boiling water for 120 minutes, the collagen (gelatine) retained 90% of its original binding ability to the IgE in three human sera (Hamada *et al.*, 2001). Collagen allergenicity must, therefore, be considered in the discussion of gelatine allergenicity.

3.2.2.1.2 Mammalian and fish gelatines

Allergic reactions to mammalian (bovine, porcine) gelatines used in vaccines and medical devices are well documented. Also, a few cases of food allergic reactions to mammalian gelatine can be found in the literature. These cases may often be a consequence of sensitisation due to medical applications of gelatine.

Mammalian and fish gelatines have some similarities, and the possibility of allergic cross-reactivity must be considered. However, several published studies support the absence of serological cross-reactivity between mammalian and fish gelatines. In contrast, fish collagens from different species appear to be broadly cross-reactive, and it seems reasonable to treat fish collagens from different species as one entity. No data have been found regarding cross-reactivity between collagens from different organs (e.g. skin and muscle) from the same species of fish.

3.2.2.1.3 Serological and immunochemical evidence for fish collagen allergenicity

A number of scientific papers report that serum IgE from some fish allergic individuals binds to fish collagen and gelatine. However, this aspect is not discussed by the applicant. The presence of IgE-binding to gelatine does not mean that clinical allergy is present. Food challenges with gelatine or collagen were not performed in the studies reporting fish collagen and gelatine IgE binding, except for the two clinical provocation studies discussed below.

3.3 Clinical studies

3.3.1 DBPCFC (double-blind placebo-controlled food challenge) study with fish gelatine

The applicant refers to a DBPCFC study performed in Denmark (Hansen *et al.*, 2004).

Patients: Thirty fish allergic patients age 9 to 50 years were included. All were fish allergic according to the European Academy of Allergology and Clinical Immunology (EAACI) Guidelines. Fifteen patients had reacted in DBPCFC with codfish (no information regarding how long before testing with gelatine), 12 of the remaining patients had experienced “classical systemic type 1 reactions to ingestion of very small amounts of codfish within few weeks to less than 24 months prior to this study”, and the last 3 patients all had experienced “relevant reaction to fish more than two years ago” and “still had a large skin prick test (SPT) to codfish and all had recently experienced rhinoconjunctivitis and asthma due to vapour from preparation

of fish meals”. These latter patients considered highly sensitive had not been challenged with fish for ethical reasons.

Protocol: SPT and histamine release tests (HR) were performed with fish gelatine made from codfish skin and with fresh raw codfish. Codfish specific IgE was measured. All patients underwent DBPCFC (seven dose steps increasing from 10 mg to 7 g with a cumulative dose of 14.61 g fish gelatine).

Results: In all 30 patients SPT, HR, and specific IgE to codfish were positive. In SPT with fish gelatine 3/30 were positive, and in HR with fish gelatine 7/30 were positive. One patient showed a mild objective reaction to placebo and no reaction to active substance challenge. Two patients reported mild subjective reactions to active challenge. They were re-challenged, and one patient with positive SPT/HR to fish gelatine and a high level of codfish-specific IgE (477 kUA/L) described subjective symptoms to active challenge with no reaction to placebo, while the other patient experienced mild subjective symptoms to placebo and nothing to active challenge. Thus, one strongly fish allergic patient showed a mild subjective reaction (“irritation in the throat” of which the patient according to the investigators was unsure [1st challenge], and “itching in the mouth” [2nd challenge]) to a cumulative dose of 7.61 g of fish gelatine, but without reappearance of the symptoms with the final dose (cumulative 14.61 g dose) in any of the two challenges. None of the 30 patients reacted adversely to the ingestion of 3.61 g cumulative dose of fish gelatine.

Conclusions: One patient had a confirmed subjective reaction at a cumulative dose of 7.61 g of fish gelatine at DBPCFC, without reaction to the following higher dose. The investigators conclude that the NOAEL for fish gelatine was a cumulative dose of 3.61 g. However, the applicant claims with reference to the study protocol (“reaction to fish gelatine not present at the maximal dose given” to be regarded as “no reactions”) that the NOAEL observed was 14.61 g fish gelatine. Statistically, the results according to the investigators indicate that there is 90% certainty that 95% of fish allergic consumers will not react to the ingestion of a cumulative dose of 3.61 g of fish gelatine. The applicant claims that “(the) results demonstrate that fish gelatine is unlikely to trigger adverse reactions”.

3.3.2 *Other clinical studies*

A study by André *et al.* (2003) analysed serum samples from 100 consecutive adults and children (age 1 to 76 years) with documented fish allergy or sensitisation without clinical allergy and tested for IgE antibodies to hydrolyzed and non-hydrolyzed (gelforming) tuna (yellowfin) skin-derived gelatine, tuna skin, tuna flesh, and bovine and porcine gelatines. In SDS-PAGE and immunoblotting three of 100 serum samples showed evidence of IgE reactivity to tuna skin-derived gelatine. Pre-incubation of the serum with gelatine, tuna skin or tuna flesh all removed the band corresponding to gelatine in immunoblotting. There was no evidence for cross-reactivity between fish gelatine and bovine and porcine gelatines. The three patients with IgE binding to tuna skin-derived gelatine had negative skin prick tests for the gelatine, and did not react clinically upon ingestion of 5 g of tuna skin-derived gelatine (single-blind testing with one dose).

3.4 *Proposed clinical studies*

The applicant does not indicate any ongoing or planned studies.

CONCLUSIONS AND RECOMMENDATIONS

No analytical data regarding possible residual levels of the major fish allergen parvalbumin in fish gelatine preparations are provided. No consumer exposure data for fish gelatine are provided by the applicant. However, exposures of up to about 10 g per serving may occur under certain conditions of use. In a DBPCFC study of 30 patients with clinical allergy to fish, one patient reacted to 7.6 g of fish gelatine.

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DOCUMENTATION PROVIDED TO EFSA

Dossier submitted by the Gelatine Manufacturers of Europe (GME) to the European Commission pursuant to Article 6 Paragraph 11 of Directive 2000/13/EC as amended by Directive 2000/89/EC, on 24 August 2004.

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