



Letter to the Editor

Food safety: In vitro digestion tests are non-predictive for allergenic potential of food in stomach insufficiency

Abstract

According to the state of the art two classes of food allergens exist: class 1 allergens are capable to sensitize as well as to trigger allergic reactions via the oral route. In contrast, class 2 food allergens do not sensitize orally because they are easily digested and thereby lose their sensitization potential. For the same reason, they trigger IgE mediated reactions only at the primary contact sites. However, our animal and human studies demonstrated that also class 2 food allergens may acquire allergenic potential when gastric digestion is hampered, e.g. by hypoacidity. Therefore, in vitro digestion tests used as decision criteria by international food safety authorities are suitable tools for revealing class 1 food allergens, but they do not permit a safety classification of digestion-labile class 2 proteins in settings of stomach insufficiency.

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Dear Editor,

The concept of digestion-resistance of food allergens has been described first by Astwood et al. [1]. In so-called in vitro digestion assays the protein in question is incubated with pepsin at low pH. Some food proteins persist this treatment for up to several hours. As these stable proteins do not only elicit allergic symptoms, but also have the capacity to sensitize, they were termed “sensitizing elicitors”, alternatively “class 1” or “true” food allergens.

In contrast, class 2 food allergens are referred to as “non-sensitizing elicitors”. Due to the fact that they are degraded during the gastrointestinal transit, they cannot sensitize directly and are capable of triggering allergy just in patients previously sensitized via inhalation. One of the most prominent examples is the birch pollen allergen Bet v 1 and its numerous homologues in plant species being regular constituents of our diet, e.g. Api g 1 in celery, Mal d 1 in apple, Dau c 1 in carrot or homologues in spices. These proteins are degraded within seconds during in vitro digestion tests. However, when conformational epitopes of Bet v 1 were experimentally stabilized, hypersensitivity could be induced via the oral route in mice, leading for the first time to the hypothesis that digestion insufficiency might support food allergy [2].

Interestingly, the quality and extent of clinical reactivity to food allergens correlates with the classification of food allergens. Class 1 food allergens are also potent triggers via the gastrointestinal route, because they are quickly absorbed and distributed conformationally intact. They trigger IgE-armed mast cells and basophils, leading to reactions along the gastrointestinal route like vomiting or diarrhea, or systemic symptoms like asthma, skin rushes, hypotension and anaphylaxis.

In contrast, digestion-labile class 2 food allergens often cause milder symptoms, primarily at the oral contact sites. Upon swallowing, the stomach acts as a gate-keeper and digests these proteins to small peptides. The apple allergen Mal d 1 for example loses its allergenicity through complete degradation. This explains why this molecule has no sensitizing capacity and elicits allergy only indirectly, via crossreaction to the respiratory birch pollen allergen Bet v 1. Consequently, apple consumption induces oral allergy syndromes rather than systemic reactions [2]. However, increasing numbers of severe allergic reactions to “class 2” food allergens, e.g. hazelnut, without primary pollen sensitization have been reported [3].

With regards to potentially life-threatening reactions in food allergic patients, considerable efforts are made to increase food safety, e.g. by obligatory labeling. These

concerns also apply to novel foods in our diet, which are on the rise due to the growing market for imported exotic foods. Moreover, also the number of genetically modified organisms (GMOs) increases continuously. Due to public pressure, regulatory authorities are forced to establish strict and transparent criteria as safety checks for novel foods. To evaluate their allergenic potential a decision tree has been developed [4], including the analysis of the gene source, of sequence homologies, of IgE-binding capacities, and of the physicochemical properties of the protein, such as resistance to peptic digestion. Up to now, this evaluation protocol has been applied for analyzing the allergenicity of novel food compounds like a microbial transglutaminase, or fish gelatine [5].

Among all food safety tests, in vitro digestion assays have become one main pillar of the evaluation process. As outlined above, gastric digestion is imitated using pepsin at pH below 2.0. The protease precursors pepsinogens, produced by stomach chief cells, get optimally activated at a pH between 1.8 and 3.2 in the gastric lumen. This implicates that any elevation of the pH should result in hindrance of peptic degradation. Numerous situations with elevated stomach pH are known, e.g. in early childhood, in elderly, or in chronic atrophic gastritis. Moreover, there are a number of pathologies, like gastritis or ulcer, where acid neutralization or inhibition is an important therapeutic goal. Acid neutralization is state of the art during surgical care, corticosteroid or analgetic treatment. Moreover, anti-acids, H₂-receptor blockers and proton pump inhibitors are increasingly consumed without prescriptions due to liberalization of the market by over-the-counter sale.

Our recent animal studies demonstrated that these potent drugs support IgE induction and sensitization to concomitantly consumed food antigens, by elevating the gastric pH and hindering pepsin activation [6]. The predictions for human food allergy held true as in an observational clinical study 15% of patients on anti-acidic therapy developed de novo IgE towards dietary compounds [7]. The IgE inductions were independent of pollen sensitizations and correlated with positive skin tests. In selected cases of novel hazelnut sensitization, the clinical relevance could be proven by positive food challenges [8]. Moreover, gastric indigestion enhances the triggering potential of food allergens significantly and may, therefore lower the thresholds of foods inducing allergic reactions [9]. Thus, the stomach has not only a controlling function for oral sensitization, but also for the triggering capacity of digestion-labile proteins.

Therefore, we suggest that in vitro digestion tests are reliable tools to select for class 1 food allergens. However, these tests provide limited information on class 2 allergens as these molecules may acquire sensitization and triggering potential when gastric function is impaired.

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Erika Jensen-Jarolim*

Eva Untersmayr

Department of Pathophysiology

Center of Physiology and Pathophysiology

Medical University of Vienna, AKH-EBO.3Q

Währinger G. 18-20, Vienna 1090, Austria

* Corresponding author. Tel.: +43 1 40400 5103

fax: +43 1 40400 5130

E-mail address: erika.jensen-jarolim@meduniwien.ac.at

(E. Jensen-Jarolim)

URL: www.allergology.at.

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