Allergen determination towards thresholds of allergic diseases

Project Team

Stefanie Allgöwer
Thomas Holzhauser
Stefanie Randow
Elke Völker

Project Summary

The knowledge about so called threshold doses, the minimum amount of an allergen which is able to cause an allergic reaction is limited. Quantitative thresholds can be determined with appropriate challenge preparations to establish clinical baseline data on sensitivity prior to allergen specific immunotherapy, and to evaluate efficacy after intervention due to change in sensitivity. Moreover, hidden allergenic compounds in processed foods, food supplements and drugs can cause serious allergic reactions. Specific and sensitive methods for the detection and quantification of allergenic compounds are required to allow the characterisation of challenge preparations with regard to consistency, and to determine allergen component specific thresholds of allergy elicitation.

The latter contributes to a better understanding of manifestation of the allergic disease. In addition, such methods allow allergen monitoring with regard to the implementation of labelling directives and to reduce the risk of allergen contamination, thus protecting the allergic consumer. For determination of data related to individual allergen components, protein fractions and protein extracts of allergenic sources, protein specific methods are preferred. In cases of risk-based approaches that are based on the identification and quantification of the presence of the allergenic source, such as total peanut, additional specific methods, e.g. DNA-based methods, are complementary.

Peanut, soybean, lupine, hazelnut, almond, Brazil nut and celery were selected as model allergens for our initial studies: We have established and in-house validated tests for quantification of these allergen sources. The assays are based on the specific detection of either protein by antibodies (ELISA, Dipstick-ELISA, Lateral Flow Devices) or DNA by PCR (Real-time PCR, PCR-ELISA). Most methods have detection limits of 0,0001 - 0.001% (1-10 mg/kg) of the potentially allergenic compound. To prove the allergological relevance of such traces, a mediator release assay based on passively sensitised rat basophilic leukaemia cells was established and applied.

In addition, ELISA methods to quantify individual allergen components, such as soybean Gly m 4 and carrot Dau c 1 and Dau c 4, were developed and allowed the quantification of relevant allergens towards the determination of component specific threshold data. Few allergen assays have been validated in ring trials, and validated allergenic reference materials are generally not available. To overcome these drawbacks novel strategies for the normalisation of the quantitative detection of potentially allergenic foods are investigated to make quantification of allergens in challenge meals, that are used in double blind placebo controlled food challenges as well as hidden allergens in processed foods, reproducible and accurate.