## Impact of oxidative stress on plant proteins modifications: relevance for plant allergens

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ovel tools are needed in order to address an important issue of how environmental pollution affects the structure and functionality of plant proteins. The impact of pollution and environmental oxidative stress, as well as heating at the molecular level have been studied by means of quantitative proteomics and global profiling of post-translational and chemical modifications by mass spectrometry and immunoblotting.

An increased phenolic content and release of sub-pollen particles was found in pollen samples from the polluted area, including a significantly higher content of mercury, cadmium, and manganese, with irregular long spines on pollen grain surface structures. Antioxidative defense-related enzymes were significantly upregulated and seven oxidative PTMs were significantly increased (methionine, histidine, lysine, and proline oxidation; tyrosine glycosylation, lysine 4-hydroxy-2-nonenal adduct, and lysine carbamylation) in pollen exposed to the chemical plant and road traffic pollution sources. Oxidative modifications affected several Timothy pollen allergens; Phl p 6, in particular, exhibited several different oxidative modifications. The expression of Phl p 6, 12, and 13 allergens were downregulated in polluted pollen, and IgE binding to pollen extract was substantially lower in the 18 patients studied, as measured by quantitative ELISA. Quantitative, unrestricted, and detailed PTM searches using an enrichment-free approach pointed to modification of Timothy pollen allergens and suggested that heavy metals are primarily responsible for oxidative stress effects observed in pollen proteins.

High temperature of roasting of peanut proteins results in several modifications of amino-acid residues that could be detected by mass spectrometry and immunoblotting. The most prominent allergen in peanut found to undergo several modifications was Ara h 1, a storage protein of peanut.

Theresults show that there are qualitative and quantitative differences in modifications of amino—acid residues of relevant plant allergens (relevant in respiratory and food allergens) in response to external stimuli and in relation to the level of inorganic and organic pollutants, as well as high temperature heating (in case of food roasting). The most frequently found modifications are oxidative modifications of amino—acid residues. Those modifications can affect IgE binding, as well as digestibility, processing and uptake of plant proteins in human digestive tract.

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