**Grain allergens on the plate – genome mapping delineates the complex mix of proteins contributing to adverse immune responses to wheat**

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Grain storage proteins that are part of the prolamin superfamily of wheat have a distinct role in the plant life cycle to provide the necessary nutrient and energy source for the germinating progeny. They are produced in an orchestrated process during the grain filling by hundreds of genes and stored in a very compact form in protein bodies within the seed. These proteins also play a crucial role when we make food products from the wheat flour as they form a three-dimensional polymer called gluten. Gluten is primarily composed of polymeric glutenins and monomeric gliadins but also includes a complex mix of other proteins from the wheat flour. The versatility of the composing proteins and their expressed amounts provide the balance between the elasticity and extensibility of the dough. The prolamin group of proteins are known to carry immune/allergy-reactive peptides (epitopes) considered to cause food disorders such as celiac disease, gluten intolerance and allergies, and constitute significant public consumer concern associated with modern wheat varieties worldwide.

Using the bread wheat reference genome published by the International Wheat Genome Sequencing Consortium, we established a new annotation and identification of the prolamin superfamily gene models in the wheat genome. The high level of sequence and structural similarities to known food allergens also helped to identify which prolamin proteins belong to food allergens and antigens and also contributed to determining the precise location other non-prolamin food allergens. In the plant these protein families have an active role in a wide range of biological processes; many of them belong to pathogenesis-related (PR) proteins and are related to the plant defence mechanisms against biotic and abiotic stresses of specific benefit to wheat per se.

The collected information led to the construction of a reference map of wheat grain allergens across the genome assembly to accurately locate “problem” genes and establish molecular markers to identify them in wheat-based foods. Additionally, the outcomes of previous clinical results on celiac disease were used to extend the concept of protein toxicity maps for individual proteins to all the proteins with a known relationship to coeliac disease in the wheat genome using the database of peptides scored for immune/allergy-reactivity. These results enabled the identification of where the most immune/allergy-reactive peptides are located within a protein and identified which wheat chromosomes are the primary contributors of these immune/allergy-reactions. By the quantitation of the highly immune-reactive peptides in different cultivars grown under different environmental conditions, we have also explored how genetic variation between the wheat cultivars and environmental changes contribute to the expression variation of these peptides and proteins. The developed method can be used to differentiate cultivars and select for lower immune-response levels.

The high-temperature stress that often affects wheat plants at flowering stage was found to induce the increase in expression of major coeliac disease and some wheat allergy related storage proteins. Expression of seed storage globulins, alpha-amylase/trypsin inhibitors, lipid transfer and non-specific lipid transfer proteins and genes with metabolic functions (e.g., kinases, peroxidases, thioredoxins) are more affected by low-temperature stress, resulting in increased expression of Bakers’ asthma-related genes.

Now we have a better understanding of the number of proteins that might be harmful. We also have some knowledge of their genetic variability as well as their environmental vulnerability. Seed grain protein content strongly depends on the growing conditions; therefore, it is essential for the food industry (especially for specialty foods/healthy foods) if they are targeting products with lower allergen content. Growing conditions with a cool finish increase the accumulation of those proteins that are related to baker’s asthma, food allergies. On the other hand, high temperature has an increasing effect on celiac associated proteins and some proteins related to wheat dependent exercise-induced anaphylaxis.

Now we also understand that the very same epitopes might be present in different proteins and that environmental changes can have a remarkable effect on the overall epitope expression. This knowledge will help to develop cultivars with modified epitope contents and will help to move on from zero tolerance gluten-free diet to food products with different levels of gluten content. It is important to note that many wheat immune-allergy-reactive proteins fulfil a significant role in stress response and adaptation, therefore breeding for low allergen content could potentially affect plant defense capacity.