CRISPR for ultra-low acrylamide wheat

Supervisory team:

Rothamsted supervisor: Prof Nigel Halford (Rothamsted Research)
Academic supervisor: Dr Gary Barker (University of Bristol)
Non-academic (CASE) supervisor: Dr Dhan Bhandari (Agriculture and Horticulture Development Board) with placement to be undertaken with Dr Chris Tapsell (KWS UK Ltd)
Dr Vladimir Nekrasov (Rothamsted Research), Dr Andy Bailey (University of Bristol), Ms Caroline Sparks (Rothamsted Research), Dr Mark Wilkinson (Rothamsted Research)

Other CASE partners: Dr Richard Jennaway (Saaten Union UK Ltd), Dr Chris Burt (RAGT Seeds Ltd), Dr David Feuerhelm (Syngenta UK Ltd), Dr Ian Foot (Limagrain UK Ltd)

Host institution: Rothamsted Research (Harpenden)
CASE partner: Agriculture and Horticulture Development Board

Project description:

Acrylamide is a 'probably cancer-causing' contaminant that is produced during food processing and cooking. Its presence in popular foods has become one of the most difficult problems facing the food industry, which has to comply with increasingly stringent regulations. Wheat products are major sources of dietary acrylamide, with biscuits, breakfast cereals, bread (particularly toasted), batter, cakes, pies and snack products all affected. Acrylamide forms from an amino acid, asparagine, in its free (non-protein) form, and sugars such as glucose, fructose and maltose, with free asparagine concentration the limiting factor in cereal products.

Asparagine is produced by an enzyme called asparagine synthetase, and wheat has four asparagine synthetase genes, ASN1-4, of which ASN2 is the most highly expressed in the grain. We have produced very low asparagine lines by editing this gene with CRISPR and by crossing lines in which the gene has been mutated using a chemical mutagen. This project will target a second asparagine synthetase gene, ASN1, that is also expressed at relatively high levels in the grain and is responsive to both nitrogen (N) fertilization and sulphur (S) deprivation. The student will investigate a strong candidate regulatory motif in this gene using cultured wheat embryos supplied with different levels of N and S. If the motif is indeed involved in N/S responses, the student will use CRISPR to disrupt it to produce a line in which the ASN1 gene is expressed but no longer responsive to nutritional triggers. If the motif is not involved, the student will use CRISPR to produce full knockouts of ASN1 in a wild-type and asn2- background. Wheat lacking both ASN1 and ASN2 would still retain two active asparagine synthetase genes, making it similar to rice. The student will analyse the edited plants for phenotypic differences with the wild-type, including grain development, grain weight and composition, yield and germination.

The student will also be able to exploit the asn2- lines that have already been produced to investigate the effects of the edits and chemically-induced mutations on the wider network of genes that we have shown to be involved in the regulation of asparagine metabolism and turnover. If possible, the analysis of the asn2- lines will involve field trials. Otherwise, all the material will be generated in a glasshouse.

The student will gain expertise in designing and making gene constructs, the application of CRISPR in wheat, and the biochemical and molecular analysis of genome-edited plants.