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Improving wheat for nutrition and processing

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SUMMARY – Wheat is the most widely grown food crop in the world being consumed after processing into a range of products. Although the major emphasis in the past has been on processing quality, there is increasing interest in the use of wheat to delivery nutritional benefits. We are therefore using new approaches to determine the molecular basis for the processing properties and nutritional quality of wheat, combining transcriptomic analysis of gene expression with the identification and exploitation of variation in natural and mutant populations. The results of these studies should provide breeders with sources of variation and tools to improve grain quality for processing and health.

Introduction

Wheat has been a central part of the European diet for many centuries, being consumed in a range of food products, including bread, other baked goods, pasta and noodles. Much of the research carried out on the wheat grain has therefore been focused on understanding the requirements for these products and producing varieties with improved end-use qualities. However, wheat is also a major source of calories and protein for human nutrition and can play an important role in a healthy diet, particularly when consumed in wholegrain products. Interest in the nutritional quality of wheat has increased over the past decade as nutritionists have become concerned about the possible association of conditions such as type 2 diabetes and obesity with the increased consumption of highly refined, processed foods. We have therefore initiated a new research programme focusing on grain components which contribute to the health benefits associated with whole grain foods.

Establishment of a technology platform for wheat improvement

We have established a suite of technologies to allow us to explore and manipulate aspects of wheat grain composition and quality. A population of approximately 4,200 lines of bread wheat cv. 'Cadenza' has been grown to the M3 generation after treatment with the chemical mutagen ethyl methane sulphonate (EMS) at either 0.6% or 0.9%. This population is being screened for visual phenotypes but will also form the basis for identifying mutations in specific genes using PCR (by TILLING, targeting induced local lesions in genomes [Colbert *et al*., 2001; Slade *et al*., 2005]). Efficient transformation systems have also been established in a range of genotypes (including Cadenza), using biolistics and *Agrobacterium*-based systems. These systems provide a basis for testing the functions of candidate genes identified by mutagenesis and bioinformatics as well as an opportunity to develop improved lines for incorporation into plant breeding programmes.

Three types of profiling are being used to study grain development and composition.

Transcriptome analysis has been carried out using a cDNA microarray of 9,246 unigene sequences (http://www.cerealsdb.net.uk/index.htm; Wilson *et al*., 2004) (Baudo *et al*., 2006; Kan *et al*., 2006) with more recent studies using the public Affymetrix wheat microarray, which includes sequences corresponding to about 55,000 genes. Metabolic profiling of mature and developing grain is being carried out by NMR spectroscopy and mass spectrometry (Baker *et al*., 2006) and proteomic comparisons by 2-D electrophoresis and mass spectrometry.

Finally, bioinformatics is being used to integrate data from these different approaches and to exploit information from public databases of wheat ESTs and the genomic sequences of rice and *Arabidopsis*.

Application to processing quality

The quality of wheat for breadmaking is determined by the genetic makeup of the plant and by the interactions of this with the environment. Stability to environmental variation is particularly important in the UK because of its variable maritime climate and is predicted to become even more important in the future as year-to-year variation is predicted to increase as a consequence of climate change (Richter and Semenov, 2004).

We have adopted a multidisciplinary approach to dissecting GxE impacts on wheat grain development and end-use quality in a major UK programme that includes eight academic partners and two companies. Five cultivars ('Spark', 'Rialto', 'Soissons', 'Beaver' and 'Hereward'), and selected doubled haploid lines from a cross between Spark and Rialto (SxR), have been grown in pots in polythene tunnels and subjected to two growth regimes from 14 days after flowering: at ambient temperature with full irrigation or at 5°C above ambient temperature with daily watering to maintain 10-40% field capacity. These conditions were based on a previous experiment carried out on cv. 'Hereward' grown in controlled environment cabinets (Gooding *et al*., 2003). Grain samples were harvested between 14 days and maturity for transcriptome profiling using the Affymetrix platform and for biochemical analyses while mature grains were used for test milling and baking and to prepare gluten for spectroscopic studies. The composition of the gluten proteins and the size distribution of glutenin polymers have also been determined by SDS-PAGE and SE-HPLC, respectively.

Preliminary analyses have shown that a total of 446 genes are differentially expressed between the seven DH lines from Spark x Rialto (Fig. 1). Other differences have been observed between the two stages of development which were selected for analysis and between the two environments. The expression of a number of transcripts is also statistically correlated with grain quality traits.

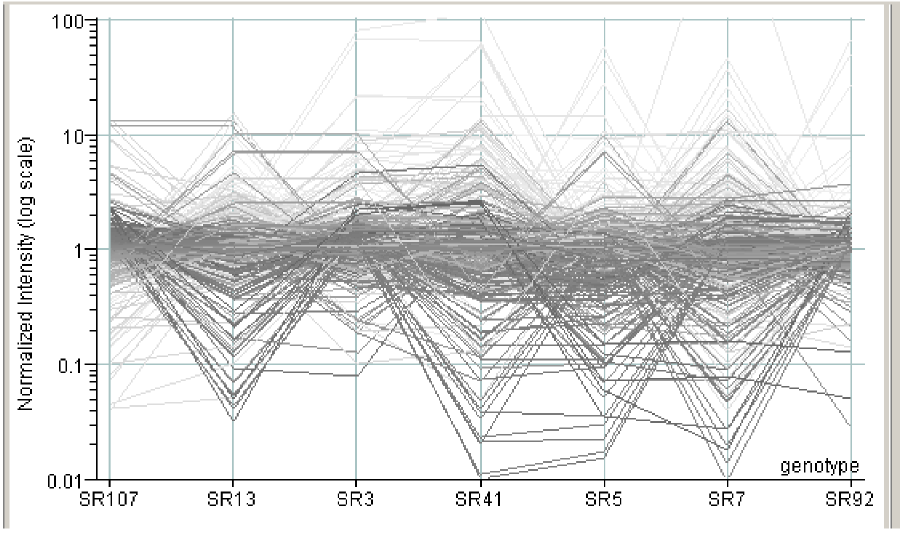

Fig. 1. Average expression of 446 transcripts determined on Affymetrix array that are significantly differentially expressed between seven DH lines from the cross 'Spark' x 'Rialto'.

Application to nutritional quality

There is a substantial body of evidence that the consumption of whole cereal grains is associated with health benefits in humans, including reduced coronary heart disease (Jacobs *et al*., 1999; Truswell, 2002). In addition, the high content of dietary fibre in whole grain products has positive effects on bowel function (Gråsten *et al*., 2003) and may be fermented by the gut flora to yield short chain fatty acids which inhibit the growth of tumour cells (Glei *et al*., 2006). In addition, "phytochemicals" that are enriched in wholegrain products (e.g. tocols, phytosterols, alkylresorcinols and phenolics) may act as antioxidants or have other specific health benefits.

The enrichment of fibre and phytochemicals in wholegrain and wholemeal products results from the fact that these components are largely concentrated in the aleurone and/or pericarp and testa. Hence, in order for consumers to benefit it is necessary to either incorporate these parts of the grain into a wider range of food products or develop new types of wheat in which they are enriched in the white flour derived from the starchy endosperm.

HEALTHGRAIN is a major research programme supported by the European Union under the 6th Framework Programme. It has 43 partners (including 10 industries) with a total budget of approximately €15 million over five years (2005-2010).

HEALTHGRAIN includes a wide range of studies, from consumer perception and expectations through grain processing and technology to nutrition and metabolism. However, an important part of the programme is focused on crop improvement, including the identification and/or generation of germplasm containing enhanced levels of bioactive components and the development tools and technologies to facilitate the exploitation of this variation by plant breeders and grain processors.

The first part of this study has included an extensive "diversity screen" of over 150 wheat lines and about 50 other cereal lines grown at Martonvasar (Hungary) in 2004/05. These lines were selected to provide a wide range of diversity and included modern varieties, historical varieties and landraces from Europe, the Americas and Asia.

Wholegrain, wholemeal, white flour and bran fractions have been analysed for a range of major (starch, fibre, protein, oil) and minor (sterols, phenolics, tocols, alkylresorcinols, folate) components, allowing differences in the amount and composition of individual groups of components to be established. Some of the lines identified may therefore be appropriate for incorporation into plant breeding programmes to produce new "nutrient dense" varieties for specific end uses.

In addition, two approaches are being taken to increase the range of variation available for plant breeders. Firstly, as described above, a mutagenised population of bread wheat has been established using ethyl methane sulphonate as a mutagen. This population will be screened for mutations in genes known to affect grain composition using TILLING and mutations in homoeologous genes combined by crossing to give "non-GM" mutant lines suitable for incorporation into plant breeding programmes. Secondly, work is in progress to identify genes encoding enzymes of arabinoxylans synthesis and to manipulate their expression by transgenesis.

At Rothamsted we are focusing on two targets, the content and composition of dietary fibre and the amount, composition and distribution of phenolic compounds.

Conclusions

We consider the interactions between genotype and environment and the delivery of health benefits as two of the most important targets for wheat research in the UK. However, both require multidisciplinary approaches, combining studies of grain development at the molecular and biochemical levels with detailed analyses of end use (nutritional and functional) properties. Improvement of these two targets will not only contribute to competitive cereal production but also to the health and well-being of EU consumers.

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