Customer Case Study:
Colorado State University Wheat Breeding and Genetics Program

The Colorado State University Wheat Breeding and Genetics Program is a long standing initiative run out of Colorado State University (CSU), USA. Lead by Scott Haley, Professor of Plant Breeding at CSU, this ground breaking program is using traditional breeding techniques combined with modern plant genomics to breed the next generation of wheat varieties for the Colorado wheat growing industry and beyond. Their locally-developed winter wheat varieties are specifically adapted to Colorado's tough climate.

Over 60% of all the wheat grown in Colorado is supplied by this wheat breeding group, as well as Kansas, Nebraska and Montana, and there is a very close symbiosis between them and the growers they supply. Scott Haley and his team also conduct basic research into genetic and environmental factors that affect wheat yield and end-use quality.

The work undertaken by Scott Haley and his team of extremely dedicated and experienced research associates involves:

- Plant genomics research
- Greenhouse propagation
- Double haploid production
- Screening
- Extensive field testing.

This combined effort produces new wheat varieties specifically bred, selected and tested for yield, drought tolerance, disease resistance, quality and other essential traits.

Using molecular biology for plant breeding:

Scott Haley has used molecular biology as part of his wheat breeding program at CSU for many years, in particular marker-assisted selection (for intergeneric introgressions) and wheat-maize doubled haploid development (for breeding line development and parent building).

Scott Haley's research team sample the leaf tissue, extract the DNA and then genotype the samples in order to identify and track single nucleotide polymorphisms (SNPs) within a set wheat population. By tracking SNPs they are able to link specific genes to desirable crop characteristics or 'traits'. When a gene of interest is found, the plant carrying this gene is then backcrossed into the population in the greenhouse to fully isolate it or 'clean it up'.

"From October to early June we are gene jockeys, working in the lab generating and analysing data; from mid-June to early October we put our overalls on and become farmers. It is a very holistic process". - Tori Anderson, Research Associate on Scott Haley's team.

The team takes advantage of an extensive network of local trial locations. These trial locations include eighteen main field plot locations around Colorado and west Kansas, over 17,000 wheat plots and 30,000 early generation headrows where the team carry out pilot studies and initial analysis.

The challenge: Time and money!

Narrow windows between harvest in July and planting in September in the University's wheat breeding program schedule mean that the program's researchers need to process a large number of samples for genotyping with a very quick turnaround.

Initially the team were using assays for SSR markers but discovered that SNPs were much more plentiful.

The team has long collaborated with the USDA Hard Winter Wheat Genotyping laboratory in Manhattan KS for routine marker analyses. These tests have included SSR markers and more recently KASP assays for SNP marker's linked to key traits in the breeding program. The staff at the USDA has been especially helpful in optimisation of new markers for selection and sharing information on development of new KASP markers.

As part of her work on Scott's team, Tori Anderson investigates new genotyping technologies to integrate into their wheat breeding program. The group started looking into KASP genotyping a couple of years ago when it was mentioned by the USDA genomics facility who were talking about using KASP markers for their work.

A TILLING program at CSU is working to identify novel variants for traits of interest in wheat production or utilisation. Sanger sequencing allowed a greater degree of customisation but they found that sequencing in order to genotype every SNP mutant that they wanted to look into was very expensive and took time.

The group need to perform SNP assays on a large number of samples quickly to allow them to select variants. In the back crossing program this is normally a one month window but can be more or less depending on the wheat strain they are growing, how fast it grows, and greenhouse conditions.

Discovering KASP

Tori Anderson contacted our genomics technical support team to find out what we could do for them and found that KASP assays lend themselves very well to exactly the type of work that CSU is doing. KASP genotyping allowed assays for new SNPs identified by the team at CSU to be designed and made available very quickly; they have had great success with how easy KASP genotyping is to use and are now trying to convert as many markers as possible to KASP assays.

KASP SNP genotyping is based on a universal detection system where the fluorescence detection system is in the KASP Master mix and generation of an assay against a new target is only a matter of ordering and assembling three short unlabelled oligonucleotides designed using LGC's software. For each new SNP marker, new KASP assays can easily be designed, ordered and supplied at very little cost.

After an initial period trialling KASP genotyping to see whether it would work for their projects, the team has now been using KASP in earnest for full blown production work for over a year. Adoption of KASP genotyping has enabled the team to do more where previously costs and time prohibited them.

They order new KASP assays as KODs (sometimes KBDs) and run them in their lab on BIO-RAD CFX real-time machines. In this way the team in the past year conducted over 16,000 total KASP assays to enable selection for 15 different trait-associated SNPs.

The team started with simple SNP assays representing novel mutations that they want to track, and are now working with more complex assays analysing small and now larger InDels (insertation/deletions) as well as haplotypes.

Desirable traits – Human health & pest resistance

As well as breeding crops tailored for the Colorado climate, traits for human health are now a focus, including traits related to starch production and antioxidant properties. Another example is selection for strains of wheat with specific aleurone layer colour, which could potentially improve product color in whole grain products made from hard white wheat. Consumers like to have the health benefits of wholewheat flour without the brown colouring that it gives to the bread, and improved strains allow the more of the healthy bran to survive the milling process without carrying with it the brown colour.

Wheat produces a volatile compound which is attractive to crop-damaging pests such as the wheat stem sawfly. Wheat varieties are being selected that produce less volatiles and are therefore less likely to be attacked by pests.

Funding

The Wheat Breeding and Genetics Program is funded in a very unique way. The program has a very strong relationship with wheat industry groups in Colorado and the majority of funding for their work comes from the *Colorado Wheat Administrative Committee (CWAC)* and the Colorado Wheat Research Foundation (CWRF). These funds are used to complement and leverage the vital base support that comes from the University through the Agricultural Experiment Station (the US Land Grant University funding mechanism for agricultural research).

A small percentage from every bushel of wheat grown by the varieties CSU supply is paid back into the group to fund their work. All or parts of the salaries of 5 of the program's workers are paid in this way, plus laboratory instruments and consumables.

"They fund our work and our equipment and in return we are able to work better…to produce a better product for them. It is a real symbiotic relationship". - Tori Anderson

This unique funding mechanism gives the group freedom to explore and find the technologies that work, but with that responsibility to deliver for their sponsors. The consortium also funds postdoctoral positions which have allowed the team to look more in depth at double haploid wheat varieties, TILLING mutants and genomic selection.

'In our sixth decade of continuous wheat breeding activity, we continue to enjoy the excellent support provided through the partnership between CSU and the wheat industry in Colorado. Funding for our program comes from a combination of state and federal funds provided by the Colorado Agricultural Experiment Station (CSU AES) and Colorado wheat industry groups, including the Colorado Wheat Administrative Committee (CWAC) and the Colorado Wheat Research Foundation (CWRF).'

CSU developed wheat varieties

- Since inception of the CSU Wheat Breeding Program in 1963, over 35 improved wheat varieties have been developed and released by the program
- Over 60% of the Colorado wheat acreage is planted to CSU-developed wheat varieties.
- Colorado growers have made substantial yield gains with new CSU varieties like Hatcher, Ripper and Bill Brown, proven in 2010, when Colorado set a new yield record of 45 bushels per acre. Recent releases such as Snowmass, Byrd, Brawl CL Plus, and Denali promise to bring even greater benefits to the wheat industry in Colorado and surrounding states.
- Varieties are marketed under the PlainsGold Brand.
- Over $3 million has been returned to CSU under the CWRF royalty program. This is money that will then fund further improved varieties for the Colorado growers.

About LGC

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With headquarters in Teddington, South West London, LGC employs over 2,000 staff, operating out of 22 countries worldwide. Its operations are extensively accredited to international quality standards such as ISO/ IEC 17025.

Set up in 1842 as the Laboratory of the Government Chemist, for more than 100 years LGC has held the unique function of the Government Chemist in the UK. LGC was privatised in 1996 and is now majority-owned by funds managed by Bridgepoint.

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