FINAL REPORT

GRDC Grains Research & Development Corporation

National screening for barley grain defects including black point, staining and pre-harvest sprouting

DAQ00094

Project Details

- **Project Code:** DAQ00094
- **Project Title:** National screening for barley grain defects including black point, staining and pre-harvest sprouting
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Summary

Three defects on barley grain can impact on the price paid to grain growers. Black point (BP), kernel staining (KS) and pre-harvest sprouting (PHS) can result in malting barley being downgraded to feed. Resistance to these defects is the best option, and in this project hundreds of breeding lines grown over three years were screened for these traits. A number of lines exhibited resistance to each defect but very few had resistance to all defects. The results from the screening program have been provided to the Australian barley breeders through the Barley Breeding Australia (BBA) program.

Conclusions

The purpose of this project was to screen lines from the BBA nodes and identify breeding lines with resistance to barley grain defects. A number of barley lines have shown good levels of resistance to these defects, with two lines showing resistance to all three defects. Having commercial barley varieties with resistance to barley grain defects is a very important future direction for the Australian barley industry. A major portion of Australian barley is exported as malting barley, malt or feed barley, and there is an expectation among international clients that the quality of Australian barley includes "clean and bright" grain. One additional and important quality attribute of Australian malting barley is the level of dormancy. Obtaining the balance between resistance to PHS (dormancy) and having grain with a high level of germination within a couple of months after harvest for malting is a difficult target for breeders. However, the right level of PHS resistance is important to protect the quality of the barley crop from rain at harvest.

Apart from the occurrence of sprouting during wet harvest periods, weathered barley grain can also take on a mouldy or stained appearance. Moderate to good levels of resistance to this defect have been achieved over recent years due to the use of resistant parents, such as Kino Nijo 7 from Japan or MN Brite from the United States (US). Current varieties such as Commander[A] and Baudin[A] have good levels of resistance to KS.

Black point is a defect with varying expression over the germ and no definitive cause has yet to be identified in barley. Environmental effects (heat and humidity during the latter stage of grain fill), as well as the presence of fungal agents, can induce a similar visually discoloured appearance. Current studies cannot confirm or eliminate the effect of any fungal pathogen on the expression of black point. However, the presence of fungal mould on the grain may well confound any environmental trigger such as heat or humidity. A preliminary analysis of five years data has indicated that higher average temperatures during grain fill results in higher levels of the expression of black point.

Recommendations

The barley breeders from all GRDC programs have indicated support for the continuation of grain defect screening and the introduction of PHS assessment. PHS in barley is responsible for the downgrading of malting barley to feed due to the negative effect that pre-germination will have on the grain when it is being malted. It addition PHS renders barley undesirable to the intensive livestock industry. Breeding material with resistance to PHS is desirable to all Australian breeding programs. It is expected that breeding lines will be identified with resistance to PHS as well as KS which is caused by pre-harvest rainfall. Preliminary results from project DAQ00031 have shown molecular regions for KS resistance to be associated with genetic regions linked to dormancy genes. This relationship will be explored further through the screening of crosses with PHS and KS resistant parents. The relationship between PHS and KS is important as breeding for PHS resistance without KS resistance would have a significant impact on the quality of stored barley. The main reason being, that if grain has any fungal staining, then it is currently assumed that a pre-harvest weathering event has occurred and germination tests can be carried out. Therefore to avoid having clean, but sprouted grain, it is necessary to breed for both traits simultaneously. The PHS screening can be provided by the Barley Quality Laboratory, which carried out this service for the DPI&F wheat breeding program during the 1980s and early 1990s.

A unique opportunity exists to investigate the possible linkage between genetic regions associated with resistance to the three defects already mentioned and fusarium head blight (FHB). The link between FHB and the three barley grain defects was initially identified from data produced from DAQ00031, where genetic regions were identified as associated with resistance to KS and husk thickness. These regions aligned with published data on resistance to FHB. In addition, the preliminary assessment of breeding lines from the BBA-North Program which were screened for and exhibited resistance to FHB have also shown to be resistant to KS. The new research opportunity would be to screen a population from BBA-North (which has a parent from the USA with known resistance to FHB) for FHB and grain defects. The outcomes from this particular research component would be molecular markers and germplasm with resistance to both FHB and KS. This material could be made available to Australian barley breeding programs, while additional populations could be screened through a defect program by DEEDI and the FHB program carried out by Industry & Investment NSW (I&INSW).

Investment in this research would ensure data availability on the level of resistance to multiple grain defects of Australia's new barley varieties. In addition, knowledge on the genetic linkage between FHB and other barley grain defects would provide a critical platform to carry out molecular screening for all of these defects in much earlier generations than present. The benefit of producing barley varieties with resistance to BP, KS, PHS and FHB will be a continued demand for Australian barley over other international competitors that continue to have FHB and KS as perennial problems.

Outcomes

The expected outcome in the project proposal was: "Germplasm with improved barley quality (in respect to the grain defects BP, KS, PHS and possibly FHB resistance) available to all participants in the BBA Program. This improvement in grain quality will be achieved through effective annual screening of germplasm supplied by breeding programs, and will lead to Australian barley varieties maintaining a market advantage over international competitors." The project has delivered on this outcome with lines from over 500 breeding lines screened for the defects. A number of the lines screened have shown a high level of resistance to these defects. In addition, as these lines are quite advanced in their breeding cycle, if they do achieve release and commercialisation, future varieties will have increased resistance to these defects. Through the release of varieties with increased resistance such as Buloke[A] and Commander[A], growers have greater opportunities to achieve malting status.

Economic outcomes

The economic benefits of the project will be evident through the eventual release of defect resistant varieties. Varieties with resistance will provide increased incomes for growers through the delivery of malting grade varieties.

Social outcomes

Improved barley quality will assist the productivity of grain growers by allowing growers to achieve malting status more frequently. It will also provide greater opportunities to export markets, thereby sustaining the reputation of Australian barley as a premium quality commodity.

Achievement/Benefit

Australian barley is exported based on its quality, including the fact that Australian barley is clean, safe and free from fungal contamination, unlike northern hemisphere crops. The objective of this project was to provide field screening of breeders' lines for three barley grain defects (BP, KS and PHS) to identify lines with resistance. The longer term outcome of this research was to ensure that future varieties contain the necessary level of resistance to these defects to meet industry standards, thereby maintaining world market share for Australian barley and marketing options for Australian barley producers.

Australia has provided high quality barley for the world malt and feed markets for many years. A number of our client countries buy grain on visual quality - its appearance, colour and aroma. This implies they are sourcing grain that appears clean and free from fungal contamination. The most recently released varieties, such as Fitzroy[A] and Flagship[A] have only moderate levels of resistance to these defects. However, grain defect screening has identified a number of Australian breeding lines with high levels of resistance to the grain defects. These lines are now progressing through Australian breeding programs. In addition, several overseas introductions including Baronesse, Valier, Chevron and MN-Brite have also exhibited a high level of resistance and are being used as parents in Australian breeding programs. A specific cross, WA5034xLindwall[A] (where WA5034 is a resistant line identified previously in project DAQ433 ‘Objective Measurement and Biochemical Basis of Malting Barley Colour’), with Lindwall (an accepted malting variety), has been found to produce a number of resistant lines. In addition, a new population (TR251xGairdner[A]), developed through BBA-West, has a high level of resistance to BP and KS inherited through its ancestral parentage.

Molecular markers

A population, Valier x Binalong[A], was screened at Hermitage and Bundaberg Research Stations over a number of seasons. The range of BP expression was similar to the breeding material tested for the BBA nodes, suggesting the field screening protocol used was effective for consistent BP expression. In addition, the results from the marker study are supported by previous studies; identifying genetic regions on chromosome 2H linked to maturity as well as regions for the expression of peroxidases and other oxidative enzymes as identified through project UA00003.

Objective measurement

Objective assessment at delivery remains an important issue. BP and KS are currently assessed manually i.e. visually. To improve the assessment of grain, the previous project DAQ00031 and this current project have trialled two image analysis systems. The first, an Australian system, worked with a moderate level of success for wheat, but failed to have sufficient levels of precision and repeatability for barley. The second system, an overseas software-based system, showed considerable improvement over the Australian system for both wheat and barley. This second system analyses an electronic image and counts the number of black or dark pixels. The system can be set up to count specific colours, so it could be possible to assess for grain colour and BP. This software underwent extensive assessment during the 2006-07 and 2007-08 seasons but failed to be seen to be quicker or more accurate that manual assessment. Near infrared reflectance spectroscopy (NIRS) will be assessed as an objective in the new barley defects project. However, preliminary results suggest single kernel screening will be more reliable than bulk screening through current NIR instrumentation. Additional research into optimising the technology and calibrations is therefore required, as well as industry investment in yet to be proven technology, to screen and separate defective kernels.

Defects associated with Bipolaris sorokininia (spot blotch)

Kernel staining is directly linked to the presence of saprophytic fungi, present due to moist conditions prior to and during harvest. However, any link between BP and fungi has not been proven, although some studies have suggested fungi, such as spot blotch, could be responsible for BP. Results from trials conducted in previous seasons have shown no link between fungal infection and BP. In 2007 the high humidity site at Bundaberg had a low level infection with spot blotch (leaf lesions noticed on a few lines), although these infected plants showed no significant increase in BP expression. To remove spot blotch as a possible causal agent, however, the 2008 trial was relocated from Bundaberg to Biloela. A comparison using check varieties across seasons showed no link between spot blotch at Bundaberg and the 2008 trial in Biloela. In addition, BP data from varieties grown in specific spot blotch screening nurseries also suggested there was no direct link. A specific set of probe lines will be used in the 2009 trials to confirm any link to spot blotch.

Black point and environmental effects

A preliminary MET analysis from the previous five seasons' trials (2003- 2007) suggests that the average temperature correlates well with the expression of BP. This relationship, as well as the effect of humidity and maturity, will be explored in the new barley defects project.

Pre-harvest sprouting

The screening carried out in this project for PHS resistance used the intact head wetting test. This method is used routinely by international researchers in screening barley. However, germination tests offer a possible alternative for assessing grain dormancy. Dr. Mares from the University of Adelaide (UA) uses this method for screening wheat. A preliminary test from the 2008 trials suggests this method may provide useful information on dormancy in barley, but testing of the hundreds of lines screened through the barley defects project would require additional resourcing and the data would not be available for many months after harvest.

Other Research

An opportunity for a new research and development (R&D) activity would be to collaborate with Dr. Jim Helm from Alberta Agriculture Food and Rural Development (AAFRD), Canada. Dr. Helm was recently in Australia for the 14th Australian Barley Technical Symposium. The Alberta barley program also carries out screening for resistance to PHS. Dr. Helm is keen to collaborate in the area of sharing markers and germplasm to identify new sources and genomic regions of resistance.

Intellectual Property Summary

Data generated from this project is provided to the BBA breeders who supply the seed for screening. The release of varieties is conducted by the respective breeding nodes and not through this project.

Collaboration Organisations

No international collaborations were part of this project, however as mentioned in 'Other research', an opportunity for a new research and development activity is to develop a collaboration with Dr. Jim Helm from AAFRD, Canada. However, any new activity would require investment to match the Canadian investment.

In addition, Dr. Steve Ullrich from Washington State University has worked for a number of years assessing for PHS resistance using the Steptoe x Morex population. From preliminary discussions, Dr. Ullrich is happy to share data so as to build a consensus map using diversity arrays technology (DArT) markers to align the Steptoe x Morex population with a number of Australian populations. Both Dr. Helm and Dr. Ullrich use the same intact head wetting test to screen for resistance. However, a germination test can be used to easily assess grain dormancy, as shown through the research conducted by Dr. Mares from the University of Adelaide when studying PHS resistance in wheat.

Additional Information

Tah P, Lehmensiek A, Fox G, Mace E, Sulman M, Bloustien G & Daggard D (2009). Identification of genetic regions associated with black point in barley. Field Crops Research. (in press).

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