IDENTIFICATION, VALIDATION, AND PYRAMIDING OF QUANTITATIVE TRAIT LOCI FOR RESISTANCE TO CROWN ROT IN WHEAT

A thesis submitted in fulfilment of the requirements for the degree of Doctor of Philosophy at the University of Southern Queensland

BY

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ABSTRACT

Crown rot (causal organism: *Fusarium pseudograminearum*) is a significant disease affecting wheat in Australia. Although first reported over 60 years ago, the disease has become more prevalent in recent years due to the adoption of minimum tillage and stubble retention practices. Breeding for resistance to crown rot is difficult – phenotypic selection, which is usually done at harvest, is time-consuming, expensive, and subject to between year variability due to sensitivity to environmental conditions. For these reasons, the coupling of molecular techniques with conventional plant breeding (marker-assisted selection) has the potential to more rapidly and reliably identify genomic regions that contribute to resistance. The objective of this study was to identify, validate, and pyramid quantitative trait loci (QTL) for resistance to crown rot present in a W21MMT70 x Mendos doubled haploid wheat population.

Replicated seedling trials were conducted in 2001, 2003, and 2005. In each seedling trial, W21MMT70 displayed partial resistance to crown rot whereas Mendos seedlings were susceptible. A bulked segregant analysis (BSA), using 390 simple sequence repeat (SSR) markers chosen for their coverage of the wheat genome, was initially conducted based upon the 2001 seedling trial data in an attempt to rapidly identify genomic regions associated to resistance. The BSA did not reveal any markers associated with resistance to crown rot. As a result, a full mapping study was conducted. One hundred and twenty eight (128) SSR markers were mapped across the population to produce a framework map. Previously screened AFLP markers were added to the map. Composite interval mapping revealed eight QTL associated with resistance. Of these, three (located on chromosomes 2B, 2D, and 5D) were consistently detected in each of the three seedling trials. Two QTL (on chromosomes 1A and 3B) were detected in two of the three trials. The 2D, 3B, and 5D QTL were inherited from W21MMT70, whereas the 1A and 2B QTL were inherited from Mendos.

Two software programs were used to identify epistatic interactions between QTL. While the results of the two programs differed markedly, both programs detected a highly significant interaction between the W21MMT70 inherited 5D
QTL and a locus on chromosome 2D inherited from Mendos. The overall effect of the epistatic interactions was not as great as the additive effects of non-epistatic QTL. Nonetheless, the presence of epistasis may indicate that, particularly in the case of 5D, the effect of this QTL may be dependent on the background into which it is introgressed.

Validation of three W21MMT70-inherited QTL (on chromosomes 2D, 3B, and 5D) was conducted on three F₂ populations with W21MMT70 as one of the parents. While the 5D QTL was validated in two of the three crosses, neither the 2D nor the 3B QTL were detected in any of the F₂ validation populations. It is likely that the size of the F₂ populations (the largest composing of 94 individuals), in conjunction with the variability that is inherent when screening for resistance to crown rot, precluded validation of these regions. Validation of the 2B Mendos-inherited QTL was conducted on a Sunco x Batavia doubled haploid population because Sunco possesses the same *Triticum timopheevi* 2B introgression that is present in Mendos. This validated QTL (designated *Q.Cr.usq-2B2*) explained 11 % of the phenotypic variance in the Sunco x Batavia population.

To assess the effectiveness of pyramiding QTL for resistance to crown rot, a 2-49 x W21MMT70 population was examined. A number of lines of this population performed significantly better than each of the parents in the replicated seedling trial that was conducted. Four QTL, located on chromosomes 1A, 1D, 2D, and 3B, were detected. The 1A and 1D QTL were inherited from 2-49 whereas the 2D and 3B QTL were inherited from W21MMT70. The 1A QTL from 2-49 has not been previously validated, and this QTL has been designated *Q.Cr.usq-1A1*. The 3B QTL (designated *Q.Cr.usq-3B1*) had the highest effect (LRS 42.1; explaining 21.0 % of the phenotypic variance) in the 2-49 x W21MMT70 population. The 2D QTL (*Q.Cr.usq-2D1*) was shown to have a minor effect. The 5D QTL that was inherited from W21MMT70 in the W21MMT70 x Mendos population was not detected in the 2-49 x W21MMT70 population. A number of possible explanations for the inability to detect this QTL in the 2-49 x W21MMT70 population are discussed.
CERTIFICATION OF DISSERTATION

I certify that the ideas, experimental work, results, analyses, and conclusions reported in this dissertation are entirely my own effort, except where otherwise acknowledged. I also certify that the work is original and has not been previously submitted for any other award, except where otherwise acknowledged.

__________________________________________    ________________________________
Signature of Candidate                           Date

ENDORSEMENT

__________________________________________    ________________________________
Signature of Principal Supervisor                Date
ACKNOWLEDGEMENTS

There are many people I would like to thank for a variety of reasons. Firstly, I’d like to thank my supervisor, Professor Mark W. Sutherland - the “Research Trainer”. Mark’s seemingly unlimited knowledge of most things plant and his clear passion to gain more, have been essential for both the guidance and motivation required to complete this study. On a personal level, I would like to thank Mark for his understanding and compassion when not just one, but two distractions entered my world, bringing with them an era of uncertainty. It has, almost always (!), been a pleasure to be supervised by Mark.

My knowledge of general molecular biology techniques was raw at the commencement of this work – I am thankful to Dr. Raechelle Grams for “teaching me the ropes”. Dr. Bert Collard’s enthusiasm instilled a real interest for QTL mapping in me (even though he himself may still be a sceptic), and Dr. Anke Lehmensiek’s German efficiency helped me to appreciate the difference between a map and a good map. The majority of the phenotyping presented in this thesis was conducted under the supervision of Dr. Graham Wildermuth and Mr. Matt Davis, and I am grateful for their leadership in this area.

I have spent more years than I care to admit trying to complete a study worthy of receiving a PhD. I have no doubt that I would not have made it to the end without the community feel that is generated by the Biological and Physical Sciences Department at USQ. There are too many people to name, but Cassy Percy, Bene Watson, Grant Daggard, Eric Storlie, Joan Vickers, Vic Schultz and Pat McConnell deserve special mentions.

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I briefly mentioned two distractions that came into my life during the course of this study - but really there were three. My wife, Jessica, has been the greatest distraction of them all. I know that I haven’t always been easy (and you know my finishing this PhD probably won’t make that much difference), but I know that I’m a happier person with you. Thankyou Jess - your support, encouragement, and optimism were, and continue to be, a source of inspiration.

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