QTL identification in a slow-rusting population reveals complex inheritance patterns of resistance

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INTRODUCTION

Slow-rusting or adult plant resistance genes that are effective against leaf and stripe rust can have significant impacts on disease severity in the field. Being quantitative and additive in nature, the combination of three or more of these genes into elite germplasm can provide very high levels of protection under epidemic conditions (Singh et al. 2000). To more effectively combine and deploy these resistance genes, it is important to know their chromosomal location. Many quantitative trait loci (QTL) have been identified as being important in reducing disease severity. There has been at least 18 loci identified as being effective against leaf rust and at least 15 effective against stripe rust (Rosewarne et al. 2008 and references therein).

Despite the identification of many slow-rusting loci, the majority have very minor effects against the diseases. However the loci, *Lr34/Yr18* on chromosome 7D and *Lr46/Yr29* on 1B, have reasonably significant effects against both leaf and stripe rust. Furthermore, the development of molecular markers to these genes has facilitated their identification in a range of germplasm. Although significant in their effect in reducing disease severity, alone these genes do not provide enough protection. Even when combined together, the resistance is usually not enough to combat the diseases to a sufficient level, unless combined with other background loci.

This study aims to identify slow-rusting loci in a F_2 derived F_5 SSD population from a cross between the susceptible Avocet and the slow-rusting line Attila. Field testing was completed over three seasons each for leaf rust and stripe rust (separately) under severe epidemic conditions. Partial linkage mapping was completed through a bulked segregant analysis. A multi-environment QTL analysis was conducted with QTLNetwork 2.0 (Yang et al. 2005).

RESULTS

Genetic analysis of this population indicated that at least two loci were involved in leaf rust resistance and at least three were involved in stripe rust resistance (Rosewarne et al. 2006). Furthermore, a significant QTL for both leaf rust reaction and stripe rust reaction was observed across all environments and located to chromosome 1BL (Fig 1). This was shown to be *Lr46/Yr29* through its co-location with leaf tip necrosis and several molecular markers diagnostic for this locus. This QTL had the highest additive effect for rust reaction against both leaf rust (27.2) and stripe rust (13.2) Table 1 and 2.

There were two other loci that had minor but significant effects on leaf rust reaction (Fig. 1, Table 1). These were located on chromosome 2BS and 7BL. These loci, along with the *Lr46/Yr29* locus, accounted for nearly 70% of the phenotypic variation observed for leaf rust.

There were six QTL associated with stripe rust reaction (Table 2), one of which showed a significant genotype × environment interaction. Loci were identified on chromosome 1BL (Lr46/Yr29), 2BS, 2BL, 7BL and two unidentified regions. The 1BL, 2BL, 7BL and an unmapped locus had consistent effects across all environments. The 2BS locus had a significantly larger effect on reducing disease severity in the second year of stripe rust data. Another unmapped locus had an epistatic effect by reducing stripe rust reaction associated with the *Lr46/Yr29* locus.

DISCUSSION

Slow-rusting resistance to leaf rust in this population was relatively straight forward with three loci contributing to resistance. The *Lr46/Yr29* locus was the dominant contributor and accounted for an average reduction in disease severity of over 54%. Two other loci had minor but consistent effects of leaf rust reaction. This is interesting as genetic inheritance analysis predict a minimum of two genes being involved in resistance. A flaw of this type of analysis is that it assumes all loci contribute equally to resistance but this is clearly not the case. It cannot be ruled out that other minor genes may also be involved in resistance.

Previous studies have shown QTL for leaf rust reaction on 2B and 7B (see numerous references in Rosewarne et al 2008). This indicates that these regions are important sites for slow-rusting resistance to leaf rust and may be widespread in a range of germplasm.

Stripe rust resistance was much more complicated in this population. Again the *Lr46/Yr29* locus had a significant contribution, giving an average reduction in disease

severity of over 26%. However, four other loci also contributed to resistance. Of the other loci, the 2BL is of interest as no previous studies had identified this region as being important for stripe rust. This locus and one other that could not be chromosomally localised were derived from the susceptible parent, Avocet. Singh et al. (2005) had identified resistance on 6A from Avocet. A possible reason why neither locus was identified by both Singh et al (2005) and in this study, could be due to the use of partial linkage mapping. This commonly results in incomplete genome coverage, so that QTL with minor effects may be overlooked.

The other locus derived from the Avocet parent was not able to be mapped as it was only identified through AFLP based markers. Therefore, although it was not linked to the 2BL linkage group, conclusions as to whether it was associated with the 6A locus of Singh et al (2005) cannot be drawn.

The QTL on 2BS was most intriguing as it showed a significant environmental interaction. In 2002, it accounted for an additive effect of 9.3%. However, this effect was much lower in the other 2 years (4.7% and 5.5%). Field notes taken in 2002 revealed an early incursion of an endemic *Yr27* avirulent stripe rust pathotype, followed up by the spread of the inoculated *Yr27* virulent pathotype. As Attila is known to contain *Yr27*, it seems likely that this gene was involved in the QTL observed in 2002. Interestingly, the smaller additive effects in 2000 and 2003 may be indicative of low levels of this endemic rust pathotype being present.

The 7BL QTL for stripe rust reaction was consistently observed in all three years. Suenaga et al. (2003) also identified a QTL in this region and this may be a useful locus in helping to generate high levels of resistance based on slow-rusting.

Finally one other locus that came from an AFLP based linkage group had an epistatic interaction with stripe rust reaction from *Lr46/Yr29*. When both loci were present this locus accounted for a decrease in additive effect of 7%. This was significant and present in all three environments. The presence of such genes in slow-rusting lines indicates the value of breeding and selecting material under epidemic conditions, as was the case with the development of Attila.

In conclusion, the inheritance of slow-rusting was relatively simple for leaf rust reaction with only a few genes being involved. Resistance to both rusts was generally dominated by the effect of the *Lr46/Yr29* locus. However, stripe rust resistance was much more complicated with a total of 7 loci being involved. Some of these loci were derived from the susceptible parent and one was epistatic in function. With the advent of molecular markers for the two dominant slow-rusting genes of *Lr34/Yr18* and *Lr46/Yr29*, the incorporation of these two loci in breeding material will be easier to achieve. However these are unlikely to provide adequate protection for stripe rust unless other minor genes are incorporated.

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Table 1 Multi-environment QTL analysis for leaf rust (LR) reaction in the Avocet-S × Attila population. Estimated additive effects are given for significant leaf rust reaction QTL in all environments and deviations from this value for individual environments. Negative values indicate the resistance allele is derived from the susceptible parent

QTL interval/marker	Chromosome location	Lr All	Lr 2000	Lr 2002	Lr 2004
XP84/M78-LTN	1BL	27.2	-0.5	1.5	-1.1
Xgwm682-XP32/M62	2BS	4.4	0.0	0.0	0.0
Xwmc273-Xgwm146	7BL	3.0	-0.4	1.4	-1.0

Table 2 Multi-environment QTL analysis for stripe rust (YR) reaction in the Avocet-S × Attila RIL population. Estimated additive effects are given for significant stripe rust reaction QTL in all environments and deviations from this value for individual environments. Negative values indicate the resistance allele is derived from the susceptible parent. * significantly different from mean additive effect at P<0.1 and ** at P<0.01; nd = not determined

QTL Interval/Marker	Location	Yr All	Yr 2000	Yr 2002	Yr 2003
LTN-*XP35/M55*	1BL	13.2	-0.7	-1.1	1.8
XP32/M62-XP88/M64	2BS	6.5	-1.8	2.8*	-1.0
Xgwm1027-Xgwm619	2BL	-4.5	-0.6	1.5	-1.0
XP32/M59-Xgwm344	7BL	3.1	0.0	0.0	0.0
XP87/M68b-XP85/M67b	nd	-3.1	0.0	0.0	0.0
XP33/M61	nd	4.8	2.3*	-3.3**	1.0

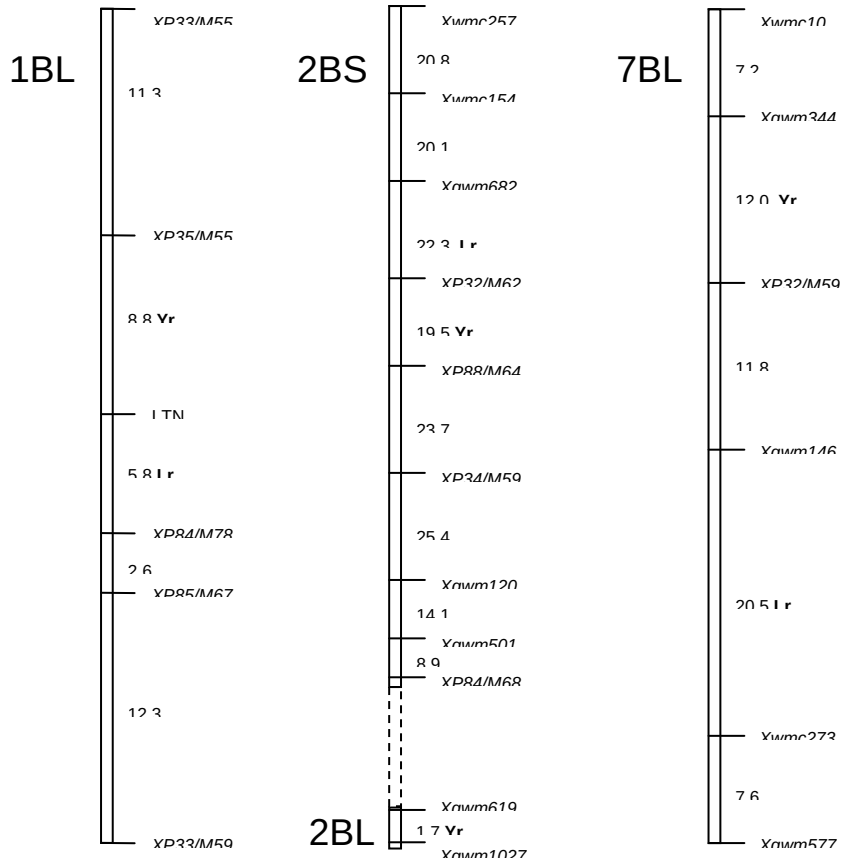

Figure 1 Loci identified as having effects against leaf rust reaction (Lr) stripe rust reaction (Yr).