

Online Resource 5

Article title: “Mapping and validation of QTL which confer partial resistance to broadly virulent post-2000 North American races of stripe rust in hexaploid wheat”

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Table S1 (page 1 of 3)

Reference Haplotypes (A = homozygous for UC1110 allele; B = homozygous for PI610750 allele):

Line	3BS (A = resistant)			5AL (B = resistant)			2B (A = resistant)		Combined 3BS 5AL	Average Reaction Type	Average Disease Severity
	Distal <i>BE446462</i>	Peak <i>gwm533.1</i>	Proximal <i>barc133</i>	Proximal <i>wmc727</i>	Peak <i>cfa2149</i>	Distal <i>wPt-9800</i>	Peak <i>gwm474</i>				
RIL149	B	B	B	A	A	A	B	BBB AAA	10.9	87%	
RIL167	B	B	B	B	B	B	B	BBB BBB	7.0	46%	
RIL233	A	A	A	A	A	A	B	AAA AAA	4.3	33%	

Validation of 3BS QTL

Population: RIL149 / RIL233, only homozygous F2 plants tested (37)

F2 3BS 5AL Haplotypes	Resistant	Susceptible
AAA AAA	18	0
BBB AAA	0	19

Fisher's Exact Probability Test *
p < 0.0001

Validation of 5AL QTL

Population: RIL149 / RIL167, only homozygous F2 plants tested (66)

F2 3BS 5AL Haplotypes	Resistant	Susceptible
BBB AAA	0	32
BBB BBB	34	0

Fisher's Exact Probability Test *
p < 0.0001

* see <http://faculty.vassar.edu/lowry/fisher.html>

Table caption is on the following page.

Table S1 (continued, page 2 of 3)

Table S1. The table on the previous page summarizes the results of the independent validation tests for the 3BS and 5AL QTL regions. At the top are shown reference haplotypes for all the lines used in these crosses: RIL149 (a completely susceptible line), RIL167 (a line carrying only the 5AL resistance), and RIL233 (a line carrying only the 3BS resistance). Shorthand "3BS|5AL" haplotypes are defined for easy reference in the two contingency tables of results.

1. To validate the 3BS QTL region, a population was developed from the cross RIL149/RIL233. Within this population, 19 F₂ plants were found to be homozygous for the RIL149 (i.e. susceptible) haplotype in the 3BS QTL region and 18 F₂ plants were found to be homozygous for the RIL233 (i.e. resistant) haplotype in the 3BS QTL region. These 37 plants were then inoculated in the greenhouse at booting stage with field spores harvested during the 2009 growing season (Davis site). 21 days after inoculation, each plant was evaluated for its reaction and classified as either resistant or susceptible (see images on next page). As the contingency table shows, reaction type segregated perfectly with haplotype, confirming the presence of an effect of the 3BS QTL region on stripe rust resistance.

2. To validate the 5AL QTL region, a population was developed from the cross RIL149/RIL167. Within this population, 32 F₂ plants were found to be homozygous for the RIL149 (i.e. susceptible) haplotype in the 5AL QTL region and 34 F₂ plants were found to be homozygous for the RIL167 (i.e. resistant) haplotype in the 5AL QTL region. These 66 plants were then inoculated in the greenhouse at booting stage with field spores harvested during the 2009 growing season (Davis site). 21 days after inoculation, each plant was evaluated for its reaction and classified as either resistant or susceptible (see images on next page). As the contingency table shows, reaction type segregated perfectly with haplotype, confirming the presence of an effect of the 5AL QTL region on stripe rust resistance.

Images of typical reactions from these screens are on the following page.

Table S1 (continued, page 3 of 3)

Typical Reactions

3BS validation screen

SUSCEPTIBLE



3BS RESISTANT



5AL validation screen

SUSCEPTIBLE



5AL RESISTANT



Table S2

Reference Haplotypes (A = homozygous for UC1110 allele; B = homozygous for PI610750 allele):

Line	3BS (A = resistant)			5AL (B = resistant)			2B (A = resistant)			Average Reaction Type	Average Disease Severity
	Distal BE446462	Peak gwm533.1	Proximal barc133	Proximal wmc727	Peak cfa2149	Distal wPt-9800	Proximal gwm429	Peak gwm474	Distal barc230		
e.g. RIL233	A	A	A	A	A	A	B	B	B	4.3	33%
3BS QTL Recombinants											
Evidence for BE446462 as distal flanking marker											
RIL199	A	B	B	B	B	B	A	A	A	7.3	48%
RIL178	A	B	B	B	B	-	B	B	B	6.7	43%
RIL204	A	B	B	B	B	B	A	A	A	6.5	27%
Evidence for barc133 as proximal flanking marker (see progeny test in Table S3, next page)											
RIL160	A	A	B	B	B	B	B	B	B	1.8	1%
5AL QTL Recombinants											
Evidence for wmc727 as proximal flanking marker											
RIL150	B	B	B	B	A	A	A	B	B	10.9	91%
RIL243	B	B	B	A	B	-	B	B	B	7.0	38%
Evidence for wPt-9800 as distal flanking marker											
RIL86	B	B	B	B	B	A	B	B	B	6.2	38%
2B QTL Recombinants											
Evidence for gwm429 as proximal flanking marker											
RIL149	B	B	B	A	A	A	A	B	B	10.9	87%
RIL150	B	B	B	B	A	A	A	B	B	10.9	91%
RIL193	B	B	B	A	A	A	B	B	B	10.9	99%
RIL194	B	B	B	A	A	A	B	A	A	7.0	68%
Evidence for barc230 as distal flanking marker											
RIL72	B	B	B	A	A	A	B	B	A	10.8	98%

Table S2. This table summarizes and presents a schematic visualization of the QTL dissection information described in the text. Specifically, presented here are the QTL haplotypes and average phenotypic scores of all RILs from the mapping population that provide evidence for the flanking markers of the three QTL regions.

Table S3

Reference Haplotypes (A = homozygous for UC1110 allele; B = homozygous for PI610750 allele):

Line	3BS (A = resistant)			5AL (B = resistant)			2B (A = resistant)		Combined 3BS 5AL	Average Reaction Type	Average Disease Severity
	Distal <i>BE446462</i>	Peak <i>gwm533.1</i>	Proximal <i>barc133</i>	Proximal <i>wmc727</i>	Peak <i>cfa2149</i>	Distal <i>wPt-9800</i>	Peak <i>gwm474</i>				
RIL233	A	A	A	A	A	A	B	AAA AAA	4.3	33%	
RIL149	B	B	B	A	A	A	B	BBB AAA	10.9	87%	
RIL160	A	A	B	B	B	B	B	AAB BBB	1.8	1%	

Validation of *barc133* as proximal marker for 3BS QTL

Population 1: RIL149 / RIL160, only homozygous F2 plants tested (6)

Population 2: RIL 233 / RIL160, only homozygous F2 plants tested (12)

F2 3BS 5AL Haplotypes	Resistant	Susceptible
AAB AAA	11	0
BBB AAA	0	2
AAA AAA	5	0

Fisher's Exact Probability Test
(Freeman-Halton extension to 2x3 tables) *
p = 0.007

* <http://faculty.vassar.edu/lowry/fisher2x3.html>

Table S3. To validate *barc133* as the proximal flanking marker for the 3BS QTL region, two populations were developed (RIL149/RIL160 and RIL160/RIL233). In the first population, 2 F₂ plants were found to be homozygous 5AL-susceptible and homozygous for the RIL149 haplotype in the 3BS QTL region; and 4 F₂ plants were found to be homozygous 5AL-susceptible and homozygous for the RIL160 haplotype in the 3BS QTL region. In the second population, 5 F₂ plants were found to be homozygous 5AL-susceptible and homozygous for the RIL233 haplotype in the 3BS QTL region; and 7 F₂ plants were found to be homozygous 5AL-susceptible and homozygous for the RIL160 haplotype in the 3BS QTL region. These 18 plants were then inoculated in the greenhouse at booting stage with field spores harvested during the 2009 growing season (Davis site). 21 days after inoculation, each plant was evaluated for its reaction and was classified as either resistant or susceptible (see images below). As the contingency table shows, reaction type segregated perfectly with haplotype, confirming *barc133* as a proximal marker.