

QTL mapping for adult plant resistance to stripe rust in the AAC Cameron/P2711_spring wheat population

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BACKGROUND

Stripe rust, caused by *Puccinia striiformis* (*Pst*), is one of the major biotic threats to wheat production in western Canada. Breeding resistant cultivars represents the most cost effective and ecologically sustainable solution to control disease. Exploring and utilizing new sources of resistance is essential for breeding resistant wheat cultivars. The wheat germplasm line P2711 possesses effective stripe rust resistance under field conditions, however, the genetic basis of this resistance is unknown. The objective of this study was to identify and locate QTL controlling adult stripe rust resistance in the AAC Cameron/P2711 recombinant inbred line (RIL) mapping population.

MATERIALS AND METHODS

- A mapping population comprising of 252 RILs was developed from the cross between wheat cultivar AAC Cameron and germplasm line P2711. The RILs were produced by the single seed descent (SSD) method and seeds of F7:8 generation were then multiplied and used for all genotyping and phenotyping filed experiments in this study.
- Parents and RILs were screened for stripe rust severity in disease nurseries at Creston, BC in 2018, 2019 and 2020, as well as at Lethbridge, AB in 2018 and 2020.
- A random selection of 190 RIL and two parents, representing two 96-well plates, were genotyped with the wheat 90K Infinium iSelect SNP assay. Genome Studio 2.0 (Illumina Inc., San Diego, USA) software was used to filter high quality SNPs.
- Linkage map was constructed using MSTMap software which was further refined at *r* max threshold of 0.35 and LOD min threshold of 3.0 using MapDisto software. Two or more linkage groups created for the same chromosome were merged into a single linkage group using less stringent cut off values (*r* max > 0.35).
- Composite Interval QTL mapping was carried out using software QTL Cartographer.

KEY RESULTS

- Disease nurseries had good disease development. Parents of RIL population, AAC Cameron and P2711, showed significant differences for stripe rust severity at adult plant stage. Histograms showed skewed distribution towards greater resistance in Creston 2018, 2019 and 2020 and Lethbridge 2020 environments (Figure 1a, c, d, and e), while skewed towards greater susceptibility in Lethbridge 2018 environment (Figure 1b).
- In total, 9,312 high quality SNP markers showing polymorphism between two parents were used for the construction of genetic map. A total of 8,914 out of 9,312 SNPs were incorporated into 29 linkage groups covering all 21 wheat chromosomes. Single linkage group were observed for almost three-fourth of the chromosomes, two linkage group for each of chromosome 1D, 3D and 7D, and three linkage groups for each of chromosome 5D and 6D, respectively (Figure 2). The total map length for all linkage groups was 4075.8 cM with an average distance of 2.2 between markers.
- A total of seven stripe rust resistance QTLs including *QYr.lrdc-1A.1*, *QYr.lrdc-1A.2*, *QYr.lrdc-2A.1*, *QYr.lrdc-2A.2*, *QYr.lrdc-2B*, *QYr.lrdc-3B* and *QYr.lrdc-5A* were identified in this study (Table 1). These QTLs were mapped on chromosomes 1A, 2A, 2B, 3B and 5A, respectively (Figure 2).

DISCUSSION

- The high density AAC Cameron/P2711 linkage map covering all wheat chromosomes provides enough coverage to dissect the genetic variation of stripe rust resistance present in this population.
- Resistance in AAC Cameron/P2711 was modulated by a combination of seven stripe rust resistance loci of varying effectiveness.
- The three most stable QTLs (*QYr.lrdc-2A.1*, *QYr.lrdc-2B* and *QYr.lrdc-5A*) had a moderate effect on stripe rust severity when used individually but showed larger effects when deployed with other stable QTLs identified in this study (Figure 3). Our findings suggest that these QTLs need to be pyramided to achieve a high level of resistance.
- QTLs *QYr.lrdc-2A.1*, *QYr.lrdc-2B* and *QYr.lrdc-5A* can easily be manipulated within a breeding program using the flanking SNP markers reported in this study.

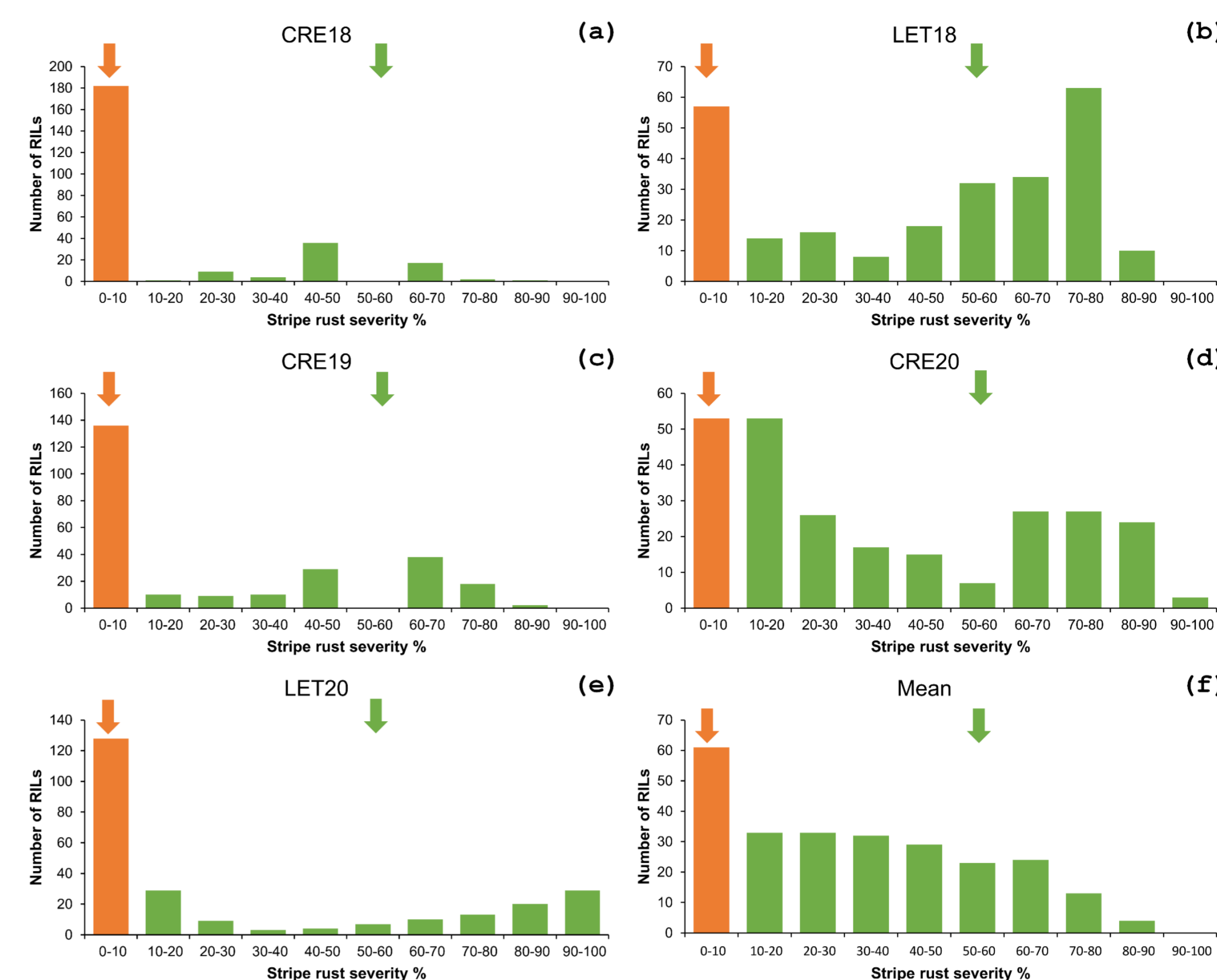


Figure 1. Frequency distribution of stripe rust severity among recombinant inbred lines of AAC Cameron/P2711 mapping population in trials at Creston 2018 (a), Lethbridge 2018 (b), Creston 2019 (c), Creston 2020 (d) and Lethbridge 2020 Environments (e) and Mean (using average data of all environments) (f). Arrows indicate the mean percentage disease severity of parents, with orange arrow representing resistant parent P2711 and green arrow representing moderately susceptible parent AAC Cameron.

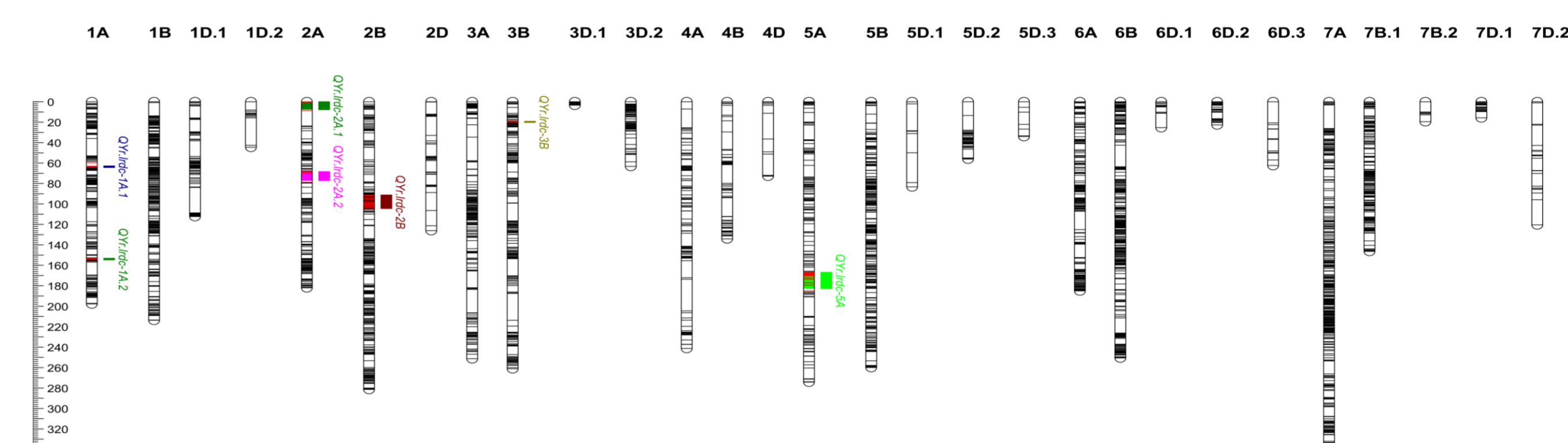


Figure 2. AAC Cameron/ P2711 recombinant inbred line mapping population genetic map. The 29 linkage groups (LGs) are labeled as 1A.1 to 7D.2 according to their relatedness to homeologous group chromosomes 1-7 and three genomes A, B and D of hexaploid spring wheat. A scale ruler for marker positions (cM) are shown on the left side of chart. Marker loci are shown as horizontal lines of either black or red colour on LGs. Red colour lines for marker loci represent linked markers of quantitative trait loci (QTL) located in respective chromosome segment, while black colour lines represent loci outside of QTL regions. Coloured LG blocks represent QTL intervals.

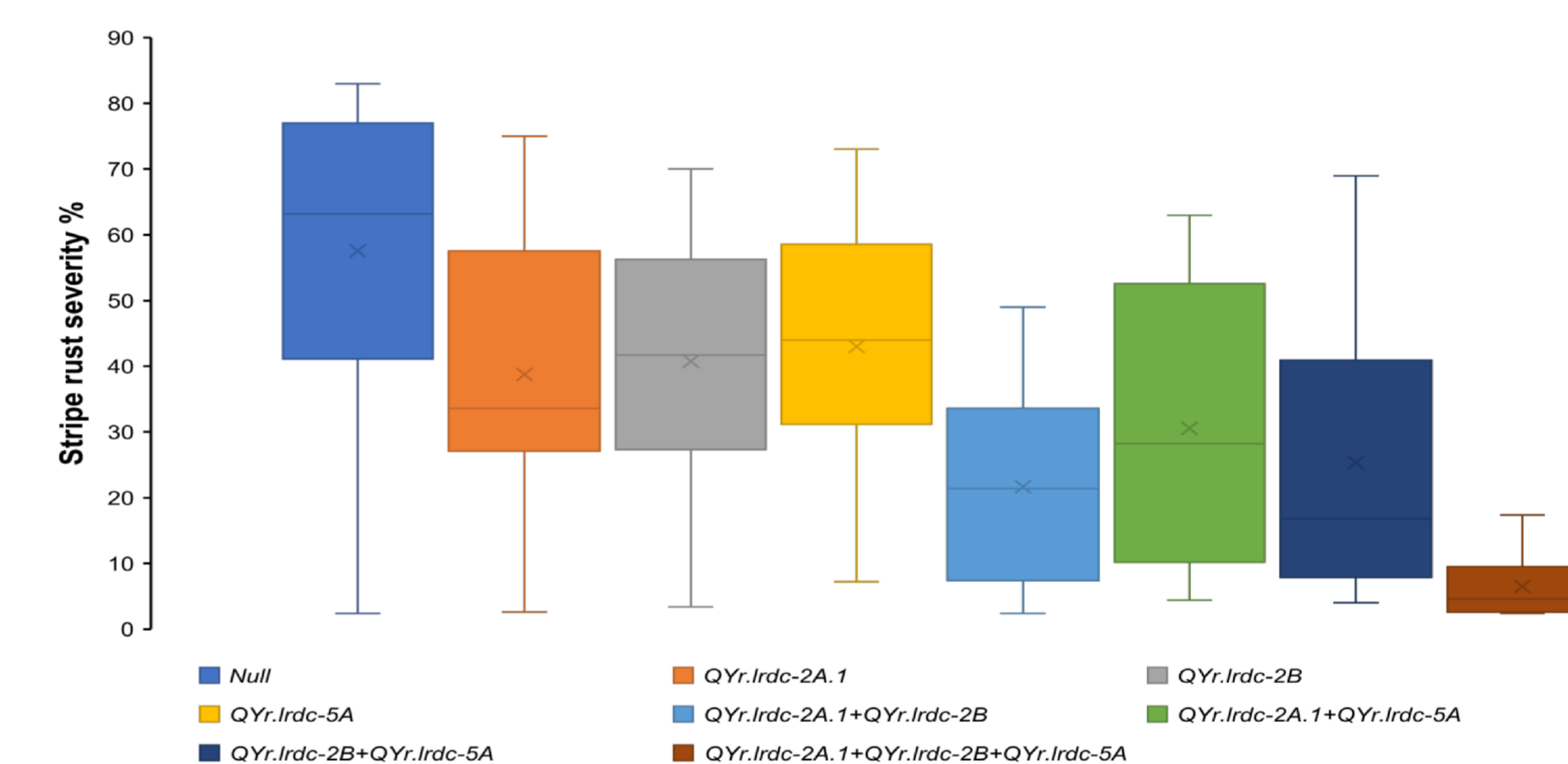


Figure 3. Boxplot distributions of AAC Cameron/P2711 recombinant inbred line mapping population. Effects of single QTL (*QYr.lrdc-2A.1*, *QYr.lrdc-2B* and *QYr.lrdc-5A*) and their combinations on stripe rust severity using pooled phenotypic data (average of all environments). Quartiles and medians are shown by boxes and continuous lines, respectively. Whiskers extend to the farthest points that are not outliers.

Table 1. Details of quantitative trait loci (QTL) identified for stripe rust (*Yr*) resistance on different wheat chromosomes in AAC Cameron/P2711 recombinant inbred line mapping population.

QTL	LG	Env	Position (cM)	Confidence Interval (cM)	LOD	Additive Effect	%R ²	Closest Marker	Physical Position (Mb)	Donor Parent
<i>QYr.lrdc-1A.1</i>	1A	CRE20	63.6	62.9-64.3	4.5	-7.94	7.2	<i>Tdurum_contig47183_205</i>	41.9-47.2	C
		CRE19	153.8	153.4-154.6 ^a	3.3	-6.18	4.9	<i>Kukri_c52420_112</i>	560.5-572.0	C
<i>QYr.lrdc-1A.2</i>	1A	CRE20	153.8	153.2-154.6 ^a	3.2	-6.44	4.8	<i>Kukri_c52420_112</i>		
		Combined	153.8	153.4-153.7	3.7	-5.72	6.0	<i>Kukri_c52420_112</i>		
<i>QYr.lrdc-2A.1</i>	2A	CRE18	0.9	0.0-7.3	6.5	8.57	12.6	<i>BS00010087_51</i>	31.1-36.9	P
		LET18	0.0	0.0-7.8	6.5	10.93	10.9	<i>GENE-0137_1660</i>		
		CRE19	0.0	0.0-6.7	5.8	8.40	8.9	<i>GENE-0137_1660</i>		
		LET20	0.0	0.0-4.6	3.9	9.83	7.4	<i>GENE-0137_1660</i>		
		Combined	0.0	0.0-5.0	5.2	7.03	8.6	<i>GENE-0137_1660</i>		
<i>QYr.lrdc-2A.2</i>	2A	CRE20	69.2	68.4-77.2	4.1	7.35	6.2	<i>BS00022301_51</i>	597.5-675.0	P
		LET18	94.8	91.8-98.6	7.1	-11.27	12.1	<i>Tdurum_contig54704_176</i>	47.4-68.2	C
<i>QYr.lrdc-2B</i>	2B	CRE19	94.8	91.8-98.6	6.0	-8.60	9.4	<i>Tdurum_contig54704_176</i>		
		LET20	103.4	102.5-104.5	4.3	-10.31	8.1	<i>BobWhite_c31129_60</i>		
		Combined	94.8	91.2-98.0	8.0	-8.80	14.0	<i>Tdurum_contig54704_176</i>		
		CRE20	69.2	68.4-77.2	4.1	7.35	6.2	<i>BS00022301_51</i>		
<i>QYr.lrdc-3B</i>	3B	CRE19	20.1	19.2-20.3	4.0	6.90	6.0	<i>RFL_Contig4531_1195</i>	6.3-13.8	P
		CRE20	169.6	168.6-171.8 ^a	2.8	5.45	5.2	<i>RFL_Contig316_572</i>	593.3-611.6	P
<i>QYr.lrdc-5A</i>	5A	CRE18	169.6	177.5-182.7 ^a	3.5	6.51	5.3	<i>IAAV108</i>		
		CRE19	179.3	167.7-170.3	11.7	13.26	20.0	<i>wsnp_Ex_rep_c109532_92292121</i>		
		Combined	169.6	166.9-176.1	5.7	7.06	8.4	<i>RFL_Contig316_572</i>		

Note: C: parent AAC Cameron; P: parent P2711; ^a Intervals determined at LOD score 2.5.

FUTURE PERSPECTIVES

- Fine mapping of identified QTLs/genes from RIL population.

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