

Genetic and molecular dissection of stem rust (*Ug99*) resistance in bread wheat

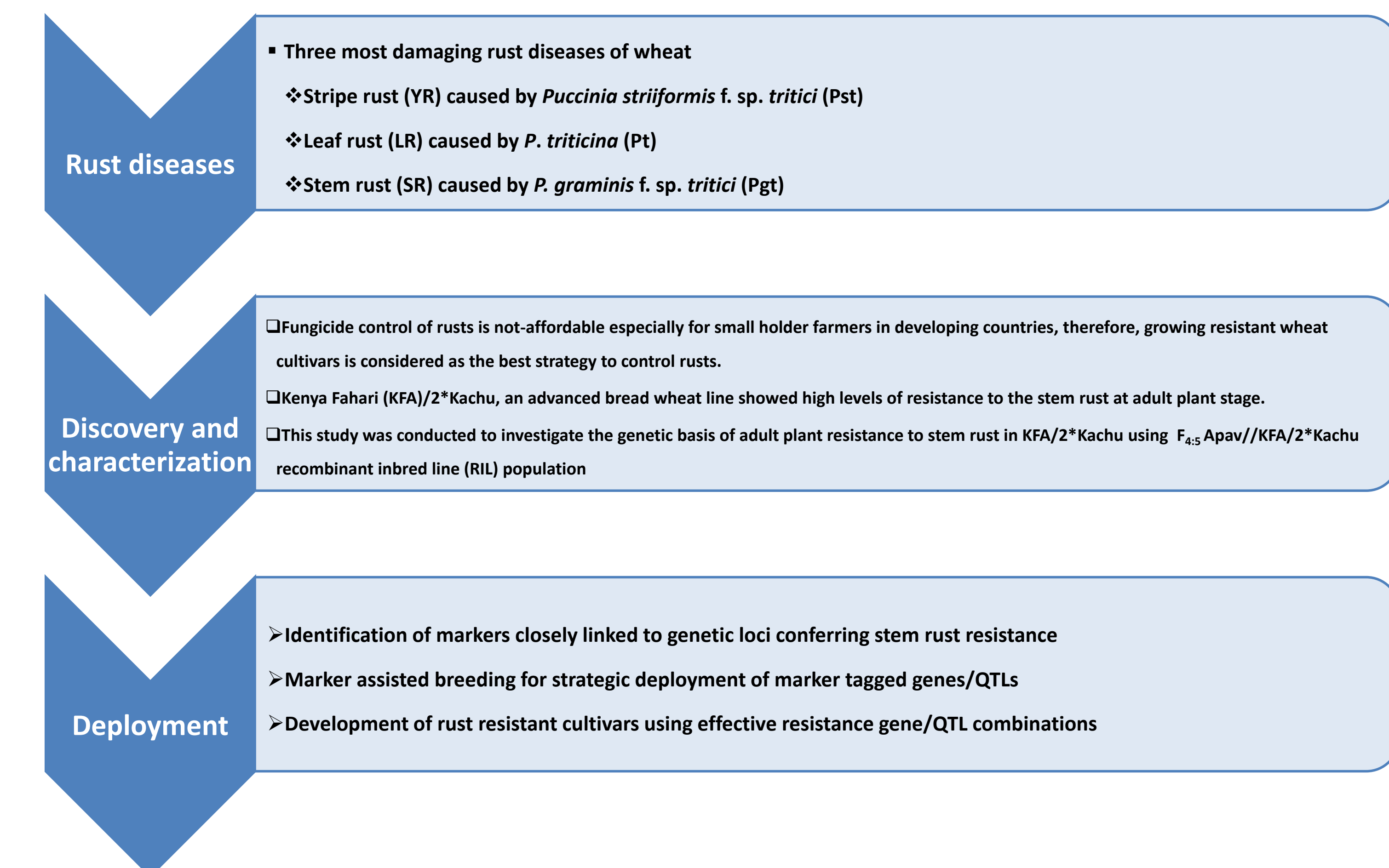
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Introduction



Materials and methods

Host and pathogen materials

❖ $F_{4:5}$ RIL population Apav//KFA/2*Kachu (198 lines)

❖ Stem rust races: RTR in Mexico; TTKST and TTKSK in CDL, USDA; TTKSK, TTKST, TTKTT, TTTTF in Njoro, Kenya

Phenotyping

❖ Field tests in Mexico for SR during season 2017-18 (2018Y)

❖ Field tests for SR (*Ug99*) in Njoro, Kenya for two seasons in 2016 (2016KO and 2016KM) and main season in 2018 (2018KM)

❖ Seedling tests against races TTKSK and TTKST at CDL, USDA

Genotyping

❖ Genotyping using DArT-GBS and gene linked markers for *Sr2/Yr30*, *Sr38/Lr37/Yr18*, *Sr58/Lr46/Yr29*, *Sr57/Lr34/Yr18*, *Sr55/Lr67/Yr46*, and *Sr8155-B1*

QTL mapping

❖ Composite interval mapping (CIM) using IciMapping software; Pearson's correlation coefficient using SAS software

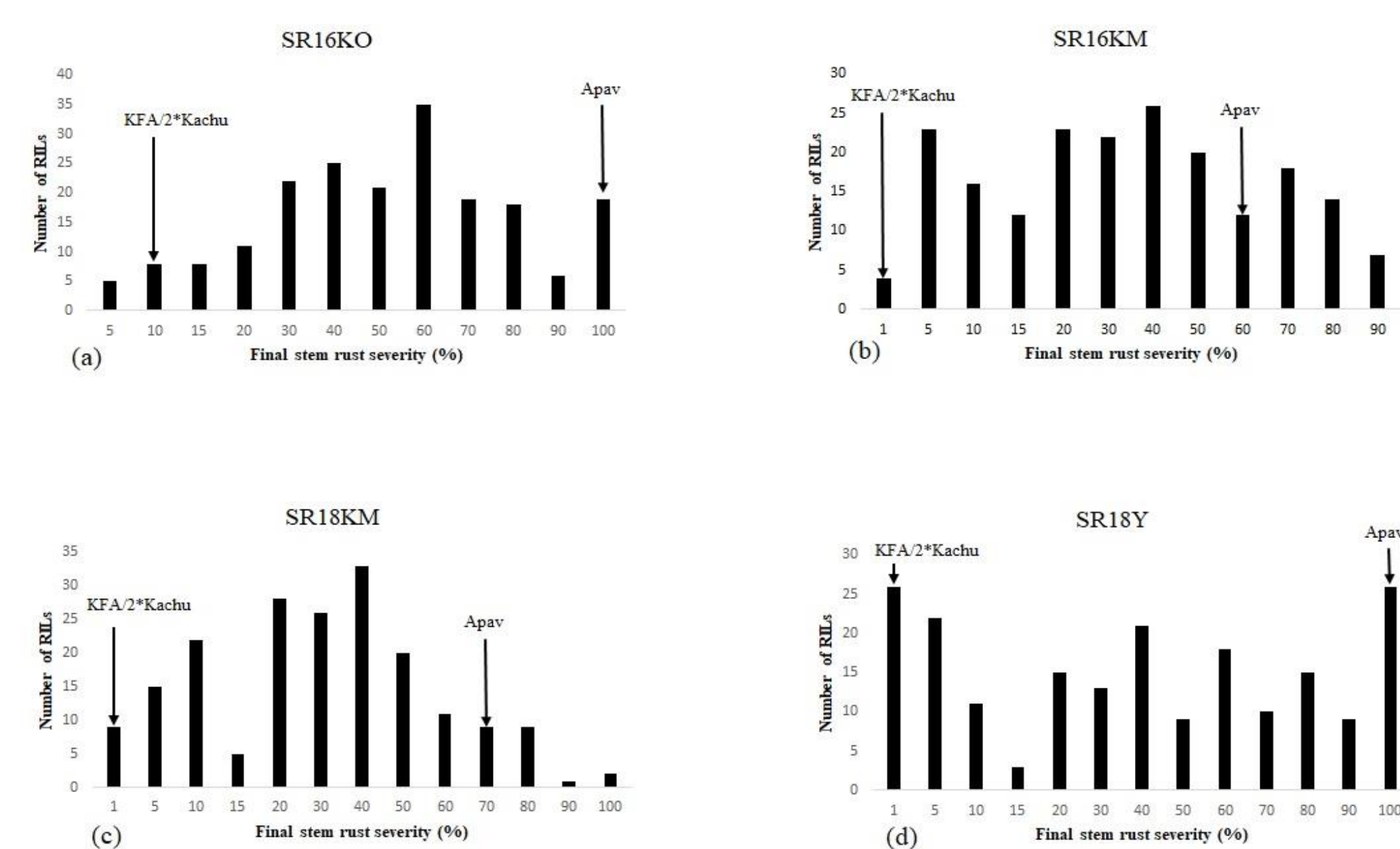


Fig. 1 Frequency distribution of final stem rust severity of Apav//KFA/2*Kachu RILs at Njoro, Kenya during 2016 off season (a), 2016 main season (b), 2018 main season (c), and at Ciudad Obregon during season 2017-18 (d). Arrows indicate the mean rust severity values of parents, Apav and KFA/2*Kachu.

	SR16KO	SR16KM	SR18KM
SR16KM	0.81**		
SR18KM	0.67**	0.74**	
SR18Y	0.60**	0.66**	0.61**

Table 1 Pearson correlations (*r*) between final disease severities of $F_{4:5}$ Apav//KFA/2*Kachu RILs in the four environments for stem rust (SR16KO, SR16KM, SR18KM and SR18Y) ***P*<0.0001

Reference

Nirmala et al. (2017) Discovery of a novel stem rust resistance allele in durum wheat that exhibits differential reactions to *Ug99* isolates. *G3* 7:3481-3490. <https://doi.org/10.1534/g3.117.300209>

Results

❖ Fig. 1 shows continuous distributions of final stem rust severities indicating polygenic inheritance.

❖ Significant correlations (medium to high) were observed between different testing environments (Table 1).

❖ Two to 3 stem rust resistance loci were identified to be segregating in the RIL population (Table 2).

❖ The QTL on chromosome 1BL, mapped to the same location as pleiotropic resistance gene *Sr58/Lr46/Yr29*, explained 14.2-27.6% of the phenotypic variation for stem rust severity (Table 3).

❖ The second QTL mapped on chromosome 2AS (*Sr38/Lr37/Yr17*), explained 18.8% of the stem rust variation during season 2017-18 at Yaqui valley, Mexico (Table 3).

❖ A consistent QTL, mapped on chromosome 6AS, *Qs.cim-6AS*, explained 13.1 to 35.4% of the stem rust variation across all SR experiments (SR16KO, SR16KM and SR18KM) conducted in Njoro, Kenya (Figure 2, Table 3).

Table 2 Estimation of the number of stem rust resistance genes segregating in the $F_{4:5}$ Apav//KFA/2*Kachu RIL population using Mendelian segregation analysis based on the final disease severity in each environment

Environment	Apav	KFA/2*Kachu	Mean	Range	HPTR ^b	HPTS ^c	OTHER ^d	Missing	Total	<i>p</i> -value ^e	No. of genes
SR16KO ^a	100 S	10 MS	53.1	5-100	13	19	165	1	198	0.21	3
SR16KM	60 S	1 MS	37.3	1-100	27	51	119	1	198	0.02	2
SR18KM	70 S	1 MS	33.4	1-100	9	21	160	8	198	0.04	3
SR18Y	100 S	1 MS	43.7	1-100	26	26	146	0	198	0.14	3

^bHPTR : homozygous parental type resistant; ^cHPTS : homozygous parental type susceptible; ^dOTHERS : lines with responses different from the two parents

Table 3 Position and effects of QTL for adult plant resistance to stem rust (SR) using final disease severity values in the $F_{4:5}$ Apav//KFA/2*Kachu RIL population

QTL/gene	Environment/race	Position ^a	Marker interval	LOD ^b	PVE (%) ^c	Additive ^d
<i>Sr58</i>	SR16KO	654.0	1122155-53:C>G-1B <i>Lr46</i> -SNP-1BL	13.8	16.9	10.1
	SR16KM	654.0	1122155-53:C>G-1B <i>Lr46</i> -SNP-1BL	23.5	27.6	14.2
	SR18KM	655.0	<i>Lr46</i> -SNP-1BL 5324108-18: A>G-1B	16.5	20.4	11.4
	SRM	654.0	1122155-53:C>G-1B <i>Lr46</i> -SNP-1BL	17.2	14.2	10.5
<i>Sr38</i>	SR18Y	397.0	1088430-2A 1229997-2A	18.8	46.4	15.1
	<i>Qs.cim-6AS</i>	SR16KO	46.0	1229292-60:C>T-6A 4405227-6A	24.0	35.4
<i>Qs.cim-6AS</i>	SR16KM	47.0	4405227-6A 1712639-6A	19.3	21.2	12.4
	SR18KM	47.0	4405227-6A 1712639-6A	15.0	17.7	10.6
	SRM	48.0	4405227-6A 1712639-6A	15.8	13.1	10.1
<i>SrKFA</i>	TTKST	43.0	3023396-6A 4910854-27: A>G-6A	16.2	29.5	1.2

^dScale for the additive effect for the seedling trait is 0-9 whereas it is 0-100 for the other traits

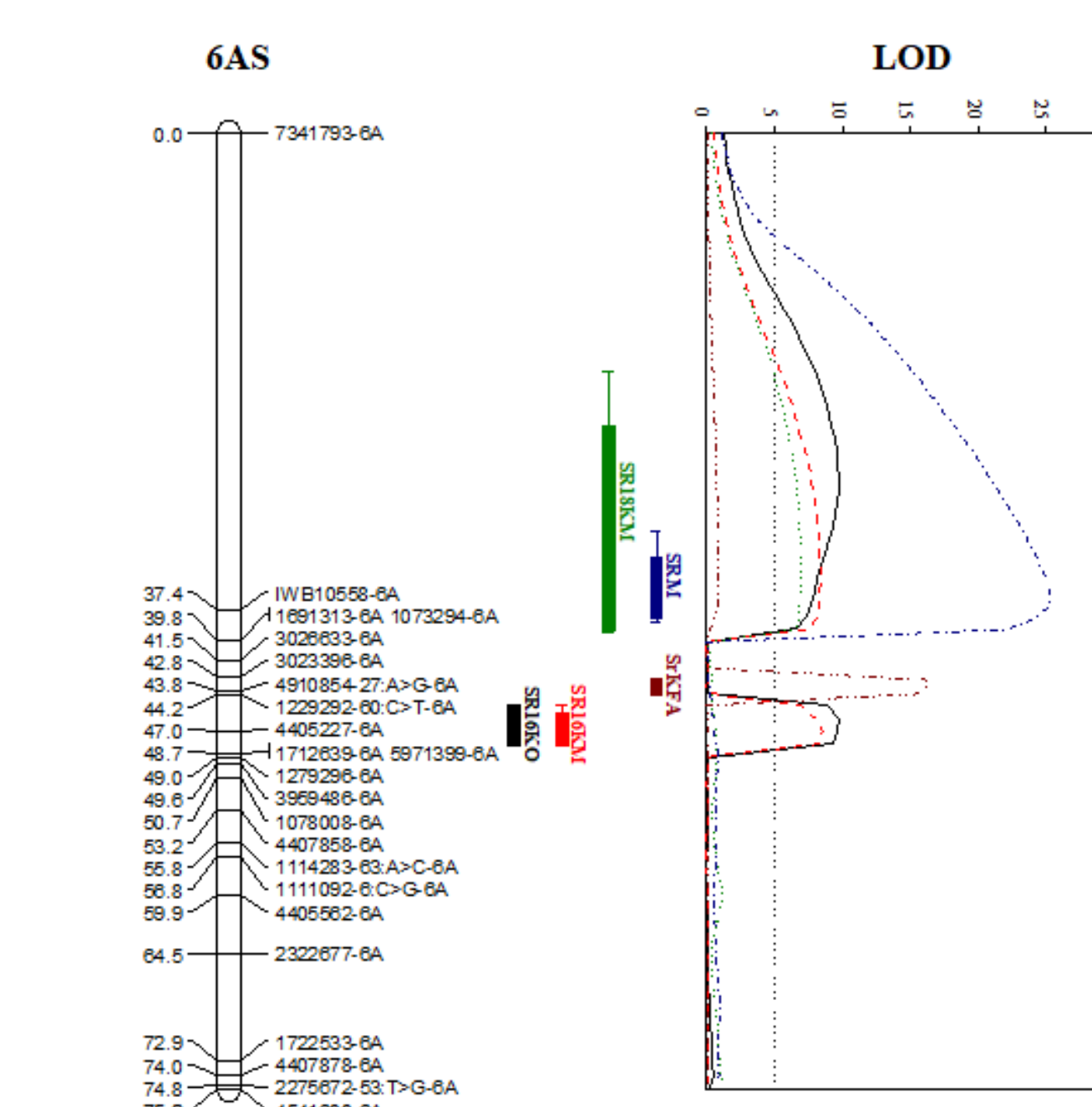


Fig. 3 Genetic map of chromosome 6AS generated using Apav//KFA/2*Kachu RIL population. Genetic distance between markers are given in centimorgan (cM). LOD, Logarithm of odds.

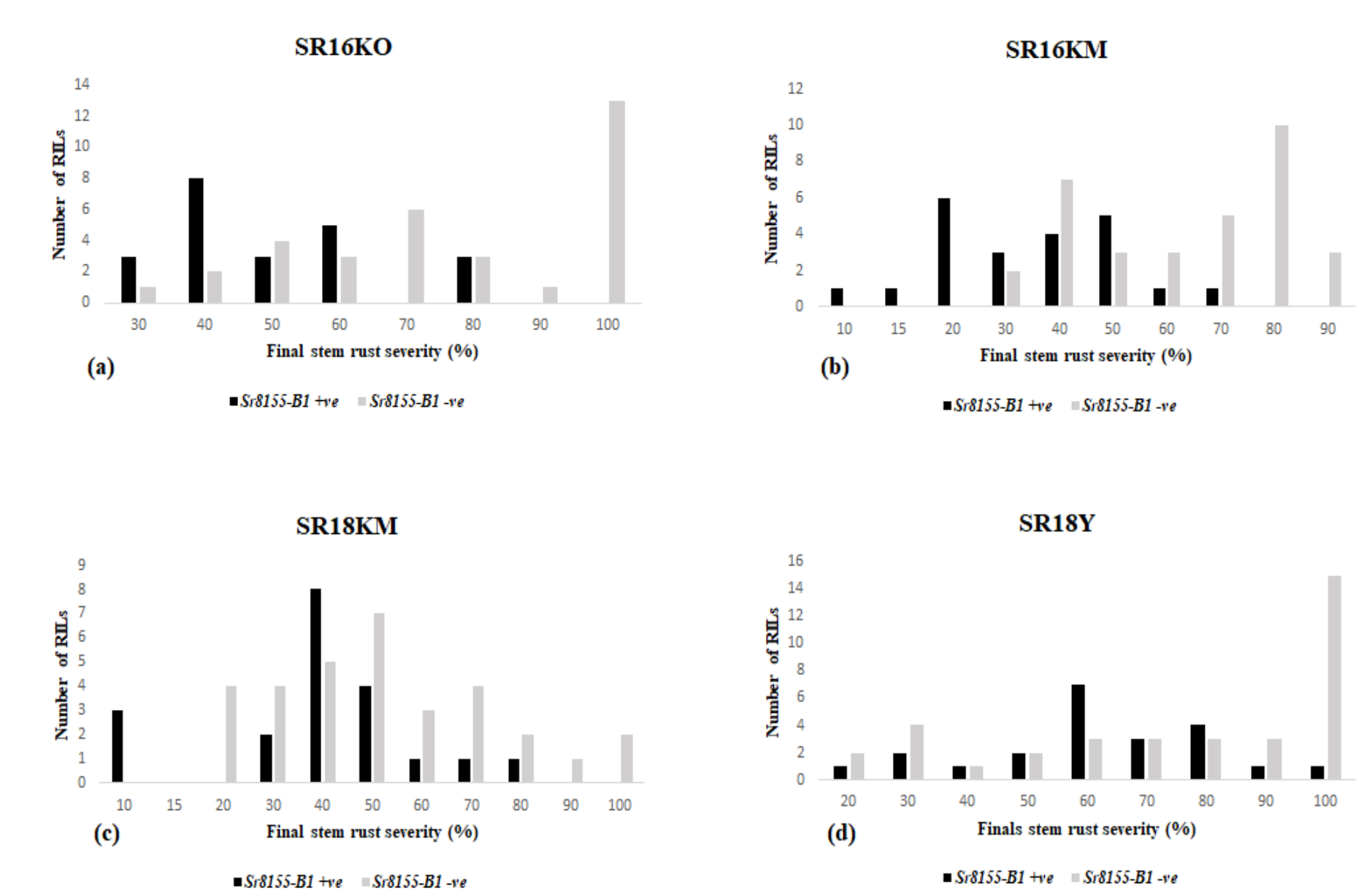


Fig. 4 Distribution of stem rust disease severity among Apav//KFA/2*Kachu RILs with (+) or without (-) stem rust resistance gene *Sr8155-B1*. RILs were classified based on *Sr8155-B1* linked marker *IWB10558-6A*.

Conclusion

➢ Genes *Sr58/Lr46/Yr29* and *Qs.cim-6AS* are derived from old Kenyan wheat variety Kenya Fahari whereas *Sr38/Lr37/Yr17* located on chromosomal segment 2AS, contributed by Kachu.

➢ QTL *Qs.cim-6AS* and *SrKFA* are mapped in the same region on chromosome 6AS as *Sr8155-B1* (Nirmala et al. 2017) from 8155-B1 durum wheat.

➢ Alike *Sr8155-B1*, QTL *Qs.cim-6AS/SrKFA* is effective against Pgt race TTKST whereas ineffective against TTKSK at the seedling stage. Both loci, *Sr8155-B1* and *Qs.cim-6AS/SrKFA*, appear identical based on similar map position and phenotypic assays, although response vary from low IT in durum (*Sr8155-B1*) to mesothetic IT in bread wheat (*Qs.cim-6AS/SrKFA*) against race TTKST.

➢ Unfortunately, susceptible pustules collected from wheat lines (e.g. Kasuko) in Njoro supported emergence of a new variant within race TTKTT with corresponding virulence.