

# Molecular mapping of resistance to the Ug99 race group of the stem rust pathogen in spring wheat landrace PI 177906

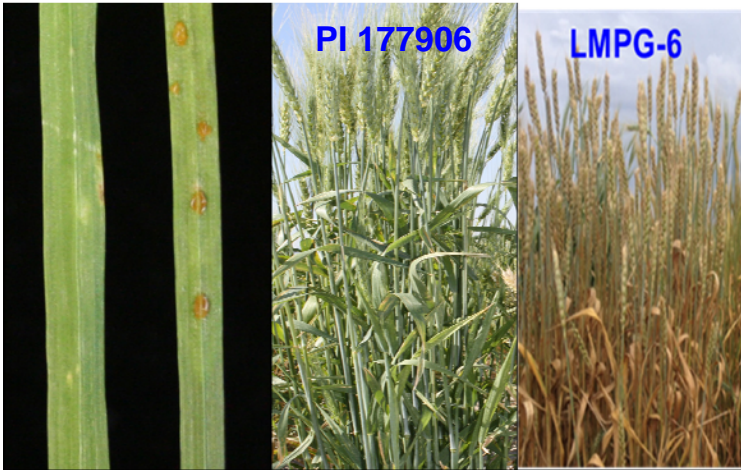
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## Introduction

- Stem rust caused by *Puccinia graminis* f. sp. *tritici*, (*Pgt*) race TTKSK commonly referred to as 'Ug99' is a threat to wheat production worldwide.
  - Wheat landrace accession PI 177906, originating from Turkey, showed seedling resistance to *Pgt* races TTKSK and BCCBC with infection type (IT) '0' and adult plant responses ranging from 5MR to 20MR in field tests at Njoro, Kenya (Fig 1).
  - To improve the efficiency of breeding for resistance to stem rust, molecular markers tightly linked to novel resistance genes are needed to enable marker assisted selection (MAS).
- **Objectives**
- Determine the inheritance of Ug99 resistance in wheat landrace PI 177906.
  - Ascertain if resistance assayed in seedling tests corresponds to resistance assayed under field conditions.
  - Identify SNP markers which could be used in MAS.



**Fig. 1.** PI 177906 and LMPG-6 seedling resistance reactions to TTKSK and adult plant field reactions in Njoro, Kenya.

## Approach

- **Plant materials**
  - PI 177906 was crossed to susceptible wheat line LMPG-6 and F<sub>1</sub> plants were used to generate recombinant inbred lines (RILs), and a doubled haploid population (DH).
- **Seedling evaluation**
  - Parents, 144 RILs, and 138 DH lines were tested as seedlings against *Pgt* race TTKSK.
- **Field evaluation**
  - Parents and the 138 DH lines were evaluated for field resistance on adult plants in three experiments in Kenya
  - Stem rust severity was assessed at the soft dough stage using the modified Cobb scale.
  - For each line, the stem rust severity was multiplied by a constant value for infection response to obtain the coefficient of infection (CI).
- **Map construction and QTL analysis**
  - Parents, 144 RILs, and 138 DH were genetically analyzed using the wheat 90K iSelect SNP platform.
  - A linkage map was constructed using JMP Genomics software.
  - PI 177906 was screened with wPt-7004-PCR and wmc332 diagnostic for the presence of *Sr28* (Rouse et al. 2012).

## Outcome

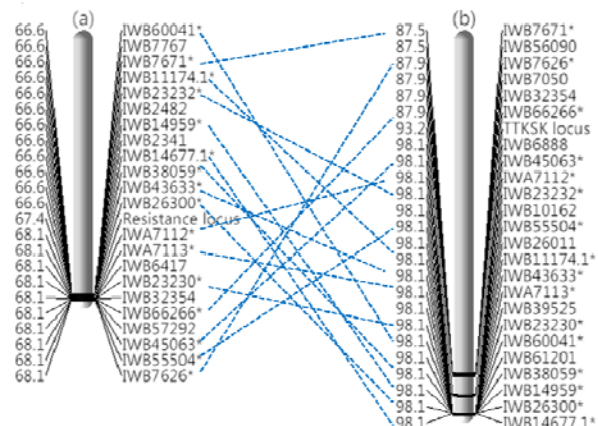
- **Inheritance and mapping of seedling and field resistance**
  - Based on seedling inoculation tests, wheat landrace PI 177906 exhibited low IT '0' when inoculated with *Pgt* races TTKSK and BCCBC, while LMPG-6 exhibited high IT '3+ to 4'. The progeny of the RIL and DH populations segregated for resistance to *Pgt* race TTKSK with resistant seedling exhibiting '0, 1, and 2, and ITs and susceptible seedlings exhibiting ITs of '3+' and '4'.
  - The DH and the RIL population segregation fitted the 3:1 ratio ( $\chi^2=0.78$ ,  $P=0.38$ ;  $\chi^2=0.0027$ ,  $P=0.95$ ) for two dominant genes.
  - In the DH population, two loci for seedling and field resistance were mapped to specific chromosomes: one on the long arm of 2B and one on the long arm of 4B. Using the RIL population, the TTKSK resistance locus mapped to the region on the long arm of chromosome 2B. In the same region major QTL for the field resistance was detected (Table 1, Fig 2).

**Table 1.** QTL for the field resistance to Ug99 in the DH population assessed in Kenya

| SNP ID     | Position (cM) | May-14 |                 |                    | Oct-14 |                 |                    | May-15 |                 |                    |
|------------|---------------|--------|-----------------|--------------------|--------|-----------------|--------------------|--------|-----------------|--------------------|
|            |               | LOD    | Additive effect | R <sup>2</sup> (%) | LOD    | Additive effect | R <sup>2</sup> (%) | LOD    | Additive effect | R <sup>2</sup> (%) |
| IWB69628   | 126.312       | 12.379 | -32.556         | 0.351              | 9.204  | -10.167         | 0.275              | 10.705 | -24.858         | 0.299              |
| IWB69630   | 126.134       | 12.379 | -32.556         | 0.351              | 9.204  | -10.167         | 0.275              | 10.705 | -24.858         | 0.299              |
| IWB55966.1 | 126.530       | 12.379 | -32.556         | 0.351              | 8.553  | -9.823          | 0.258              | 11.433 | -25.715         | 0.315              |
| IWB25869   | 126.530       | 12.379 | -32.556         | 0.351              | 8.553  | -9.823          | 0.258              | 11.433 | -25.715         | 0.315              |
| IWA8534    | 126.143       | 12.379 | -32.556         | 0.351              | 8.553  | -9.823          | 0.258              | 11.433 | -25.715         | 0.315              |
| IWA1821.1  | 126.530       | 12.379 | -32.556         | 0.351              | 8.553  | -9.823          | 0.258              | 11.433 | -25.715         | 0.315              |
| IWB23660   | 126.312       | 12.379 | -32.556         | 0.351              | 8.553  | -9.823          | 0.258              | 11.433 | -25.715         | 0.315              |
| IWB57663   | 126.134       | 12.379 | -32.556         | 0.351              | 8.553  | -9.823          | 0.258              | 11.433 | -25.715         | 0.315              |
| IWB69631   | 126.312       | 12.445 | -32.460         | 0.352              | 9.599  | -10.475         | 0.285              | 11.043 | -25.097         | 0.306              |
| IWB60041   | 130.618       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB7767    | 130.618       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB7671    | 134.712       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB1174.1  | 134.460       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB23232   | 130.291       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB2482    | 134.712       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB14959   | 130.291       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB2341    | 134.460       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB14677.1 | 130.291       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB38059   | 130.618       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB43633   | 129.081       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |
| IWB26300   | 130.618       | 12.706 | -32.687         | 0.358              | 10.469 | -10.762         | 0.306              | 10.665 | -24.280         | 0.298              |

## Conclusions

- Based on the mapping results, race specificity, the seedling infection types observed, and marker results, the TTKSK resistance in 2BL could be due to *Sr28*.
- Resistance at the *Sr28* locus was detected in both seedling and field tests.
- However, seedling test results only partially corresponded with field results, since the locus on 4B detected using seedling data was not detected using field data.



**Fig 2.** Genetic map of Ug99 resistance locus on chromosome 2BL constructed using the (a) DH population and (b) RIL population.