Puccinia graminis: Molecular Analysis

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Life cycle

Puccinia graminis

Leonard & Szabo Mol Plant Pathol 2005
**Puccinia graminis**

- **Broad Host range**
  - 54 genera, 360 species

- **Subdivided**
  - Spore morphology (subspecies, varieties)
  - Host range (formae speciales)
    - *P. graminis* f.sp. *tritici*
      - Wheat, barley
    - *P. graminis* f.sp *secalis*
      - Rye, quack grass
    - *P. graminis* f.sp. *avenae*
      - oat
Phylogenetic analysis

![Phylogenetic Tree]

- *Puccinia triticina* (Triticum aestivum)
- *P. persistens* (Elytrigia intermedia)
- *P. persistens* (E. repens)
- *P. agropyrina* (Elytrigia sp.)
- *P. recondita* (Aegilops ovata)
- *P. recondita* (Secale cereale)
- *P. hordei* (Hordeum vulgare)
- *P. holcina* (Holcus lanatus)
- *P. triseti* (Trisetum flavescens)
- *P. striiformis* (Poa pratensis)
- *P. striiformis* (T. aestivum)
- *P. striiformis* (Dactylis glomerata)
- *P. graminis* (T. aestivum)
- *P. graminis* (S. cereale)
- *P. graminis* (E. repens)
- *P. graminis* (P. pratensis)
- *P. graminis* (D. glomerata)
- *P. hemerocallidis*

10 changes
Race Nomenclature
Wheat Stem Rust Differentials

- Set includes 20 lines
- Most are single R gene lines with a common background
- Infection types (IT) are characteristic of particular R gene

Low IT (R) 0; 1; 1-; 11; 2; 2+; 3; 4

High IT (S)
## Race Nomenclature

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Ug99 - History

First reported in Uganda in 1999
--Pretorius et al. 2000 Plant Dis 84:203

- Virulent on Sr31
- Sr31 is located on 1BL.1RS translocation.
  - 1BL.1RS translocation is widely spread in wheat worldwide due to increased adaptation and higher yield.
  - 1BL.1RS carries Lr26, Yr9.
  - Virulence to Yr9, originated in the eastern Africa in mid 80s, caused worldwide epidemics.
Ug99 - History

- In 2002 and 2004, CIMMYT nursery planted in Njoro, Kenya were severely infected by stem rust.
- Kenyan isolates from 2004 collections were characterized as race TTKS with Sr31 virulence.
  - Wanyera et al., 2006 Plant Dis.
- Ug99 (98UGA1) = race TTKS with Sr31 virulence
- Expanded set of wheat differentials developed to include Sr31 (race TTKS).
  - Jin et al., 2008 Plant Dis.
In 2006, scattered MS to S pustules with low frequency were seen in many Sr24 lines in the Kenyan stem rust nursery. Sr24 virulence was suspected.

- Seedling tests confirmed the variant was virulent on Sr 24 as well as Sr 31
- Race TTKST

Sr36 Virulence

- In 2007, scattered MS pustules were observed on Sr36 lines in Kenya plots.

- Seedling tests confirmed the variant was virulent on Sr 36 as well as Sr 31
- Race TTTKS

Jin et al., 2009. Plant Dis.
## Race Nomenclature: Ug99

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SSR Marker Analysis

PgtSSR47
SSR Marker Analysis

- Compared 18 isolates from Kenya representing races:
  - TTKSK (Ug99)
  - TTKST (Sr24 vir)
  - TTTSK (Sr36 vir)
- 13 SSR markers tested.
- All isolates of these 3 races had identical SSR genotypes.
- SSR genotypes for Ug99 was distinct from North American isolates.

Jin et al., 2009. Plant Dis.
Pgt Genomic Resources
Pgt Genome Sequence

- U.S. Isolate
  - CRL 75-36-700-3

- Standard ABI/Sanger sequence
  - 12X
  - Assembly (7X)
    - 4,557 contigs (81.5 Mb)
    - 392 supercontigs

- http://www.broad.mit.edu/annotation/genome/puccinia_graminis.3
“Ug99” sequencing

- Illumina
  - 75 base read length

- Isolate: 04KEN156/04
  - Race: TTKSK ("Ug99")
    - Approximately 176 million reads (16 lanes)

- Reference isolate: CDL 75-36-700-3 (Ref)
  - Race: SCCL
    - Approximately 21 million reads (2 lanes)
Comparison of DNA sequence data from reference isolate and Ug99 was used to develop a SNP database.

DNA regions containing 2 SNPs within 10 base were chosen as targets for assay development.

Primers and TaqMan probes are being tested for a selected set of target regions.
Summary
Summary

- **Ug99**
  - Lineage contains at least three members:
    - TTKSK Ug99 (vir Sr31)
    - TTKST (vir Sr31 & Sr24)
    - TTTSK (vir Sr31 & Sr36)
  - SSR genotypes of isolates from these three races are identical indicating that the variants (TTKST and TTTSK) are derived by mutation rather than sexual recombination.
Summary

- Genomic resources
  - Standard ABI/Sanger sequence
    - U.S. isolate (CRL 75-36-700-3)
    - Assembled and annotated
  - Illumina
    - Ug99 (04KEN156)
    - Reference U.S. isolate
  - Using SNP data to develop PCR assay specific for Ug99.