Comparison of isolates of *Rhizoctonia solani* and development of a DNA probe to detect *Rhizoctonia solani*

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Abstract

Rhizoctonia solani is the causative organism of sheath blight disease of rice. In addition to rice, the fungus infects a large number of crop plants all over the world. Early detection of the pathogen is useful in controlling the diseases. Presently the most common method of identification of the pathogen is based on morphology which is time consuming and laborious. To overcome these difficulties a repetitive sequence from the genome of *R. solani* was cloned with a view to develop DNA based diagnostic assays.

Different isolates of *R. solani* were collected from low country wet zone of Sri Lanka and isolates were compared by using their external morphology, virulence, growth rates and RAPD polymorphisms. Most of these isolates did not show considerable variation in external morphology (except the isolate RZ-10). Growth rates of the fungus were compared by average linkage cluster analysis (in SAS statistical analysis system) using the data obtained by growing the fungus in different solid and liquid culture media. Comparison of the isolates was also made by RAPD polymorphism using Nei and Li's pair wise distance calculations.

The results indicated that the isolates grouped in a similar manner both in RAPD polymorphism studies and growth rate comparisons on Czapek Dox medium. Some isolates of *R. solani* appeared to be virulent on the rice variety BW 328-1 where as other isolates were non-virulent.

A repetitive DNA fragment from the genome of *R. solani* was cloned and sequenced. The cloned sequence was 1550 bp long and appeared to be interspersed throughout the genome. The repeats were present in about 500 copies per haploid genome and constitute approximately 1.3% of the fungal genome. The cloned repetitive sequence hybridized only to *R. solani* DNA and was sensitive enough to detect 100 pg of *R. solani* DNA.

PCR primers were designed from the cloned sequence and it was possible to develop a PCR assay for the specific detection of the fungal DNA with 10 pg sensitivity.