

**D-10: Development of a DNA probe to detect *Rhizoctonia solani***

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*Rhizoctonia solani* is the causative organism of sheath blight disease of rice. It is also pathogenic on several other plant species throughout the world. The most common method of identification of the pathogen is based on morphology which is time consuming and laborious. To overcome these disadvantages, repetitive DNA sequences from the genome of *R. solani* were cloned for the development of DNA based diagnostic assays.

*R. solani* was grown in modified Czapek Dox nutrient broth. Total genomic DNA was extracted from freeze dried fungal mat. *R. solani* genomic DNA was partially cleaved with *Sau* 3AI and the resulting fragments were used to construct a genomic library in the vector  $\lambda$ ZAP. The amplified library was screened for repetitive clone with <sup>32</sup>P-labelled total genomic DNA of *R. solani*. Following several cycles of plaque purification, a clone designated RZ10 was selected for further studies. The clone RZ10 was *in vivo* excised to produce pRZ10 phagemid.

Southern blot analysis of genomic DNA showed strongly hybridizing bands in all lanes and based on the restriction pattern, the cloned fragment appears to be an interspersed repeat. Dot blot analysis revealed that the clone pRZ10 was capable of detecting as little as 100pg of genomic DNA.

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