Title: Specific detection of *Rhizoctonia* pathogens using genome fingerprinting and hybridization based analyses

Amaradasa, B.S[1]. and Lakshman, D.K[2].

[1] Department of Plant Pathology, University of Nebraska-Lincoln, Lincoln, NE 68583, and [2]Floral & Nursery Plants Research Unit and Sustainable Agricultural Systems Lab, USDA-ARS, Beltsville, MD.

Abstract: Several species and hyphal anastomosis groups (AG) of *Rhizoctonia solani* (sensu lato) cause brown patch diseases of turfgrasses. Conventional methods of identification of *Rhizoctonia* pathogens are time consuming and often inaccurate. To achieve more reliable pathogen identification, sequence characterized amplified region (SCAR) markers from Universally primed PCR (UP-PCR) products were developed to identify turf pathogenic isolates of *R. solani* AG 1-IB and AG 2-2IIIB. The developed SCAR markers could distinguish isolates of AG 1-IB or AG 2-2IIIB groups and did not amplify any product from genomic DNA of non-target isolates of *Rhizoctonia*. The specific primers were sensitive and unique enough to produce a PCR band from total DNA of diseased turfgrasses infected with either AG 1-IB or AG 2-2IIIB. Moreover, a rapid identification assay for *Waitea circinata* (anamorph: *Rhizoctonia* spp.) varieties *zeae* and *circinata* which cause patch diseases, was developed based on the UP-PCR products cross-blot hybridization. Isolates within a *W. circinata* variety cross-hybridized strongly while non-homologous isolates did not cross-hybridize or did so weakly. Closely related *W. circinata* varieties *zeae* and *circinata* were clearly distinguished using this assay. For phylogenetic differentiation, seventy-nine previously characterized *R. solani* (n = 55) and *W. circinata* (n = 24) isolates were analyzed with AFLP markers generated by four primer pairs. Separate analysis of *R. solani* and *W. circinata* isolates based on unweighted pair group method with arithmetic mean (UPGMA) correctly grouped them according to their AG, AG subgroup, or *W. circinata* variety. Principle component analysis (PCA) corroborated UPGMA clusters. This is the first time AFLP analysis has been tested as a method to decipher the AG, AG subgroup, or *W. circinata* variety across a wide range of *Rhizoctonia* isolates.