Geographic distribution of *Rhizoctonia* and *Pythium* species in soils from dryland cereal cropping systems in eastern Washington

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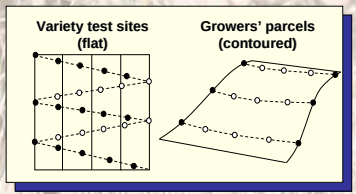
ABSTRACT

Rhizoctonia and *Pythium* species cause substantial reductions in yield in eastern Washington. Both pathogens are common in agricultural soils; however, the specific species or anastomosis group (AG) present can vary from site to site. Due to a wide range in virulence among these different groups, the impact of these pathogens could vary greatly depending on the species composition at a particular site. Soils were collected from 21 wheat variety testing sites and 29 grower fields throughout eastern Washington. DNA was extracted from these soils using a commercial kit in combination with pressure cycling treatment. Specific primers were used with real-time PCR to detect and quantify *R. solani* AG-8 and AG-2-1, *R. oryzae*, *P. irregulare* groups I and IV, and *P. ultimum*. *R. solani* AG-8 was found most frequently in areas of low rainfall (<350 mm/year) while *R. oryzae* and *P. irregulare* groups I and IV were prevalent in higher rainfall zones (>350 mm/year). The presence of *P. ultimum* was sporadic and occurred only in the higher rainfall zone or irrigated plots, while *R. solani* AG-2-1 was primarily found in fields with a history of mustard or canola cultivation. The use of these robust tools for quantification of soilborne pathogens can greatly improve our understanding of the diversity in the soil and potential correlations with plant damage and yield.

INTRODUCTION

Soilborne pathogens of wheat and barley such as *Pythium* and *Rhizoctonia* spp. are virtually ubiquitous in dryland cereal production areas of Washington. Both are chronic problems, causing substantial yield reductions each year. Unlike for foliar diseases, estimating plant damage and quantifying the populations of soilborne pathogens is challenging. Traditional quantification techniques are labor intensive and may result in underestimating slower growing species. In addition, identification of the specific species causing the problem requires time-consuming baiting procedures and can take months to complete. Both *Rhizoctonia* and *Pythium* have multiple pathogenic species, each with different degrees of virulence on host crops. Use of species-specific primers with real-time PCR allows for precise identification of species present as well as estimating the pathogen populations in the soil.

Soils were collected from Washington State University variety testing sites and grower parcels in early June, 2006. Samples were collected using a soil corer that removed a 2.5 cm diameter by 15 cm deep core. For the variety testing sites, a "W" pattern was used to collect soils and for grower parcels, three transects were taken, following the contour of the field. Each sample consisted of five pooled cores removed from each transect. DNA was extracted from the soils using the MO-BIO soil DNA kit and a Barocycler™ NEP3229 (Pressure Biosciences, Inc., West Bridgewater, MA). Real-time PCR was used to amplify DNA from the soil extracts. Signal was detected by use of SYBR Green I fluorescent dye in combination with the Roche LightCycler. DNA extracts were screened for the presence of the more virulent species of *Rhizoctonia* and *Pythium* which included *R. solani* AG-8 and AG-2-1, *R. oryzae*, *P. irregulare* groups I and IV, and *P. ultimum* by use of species-specific primers.

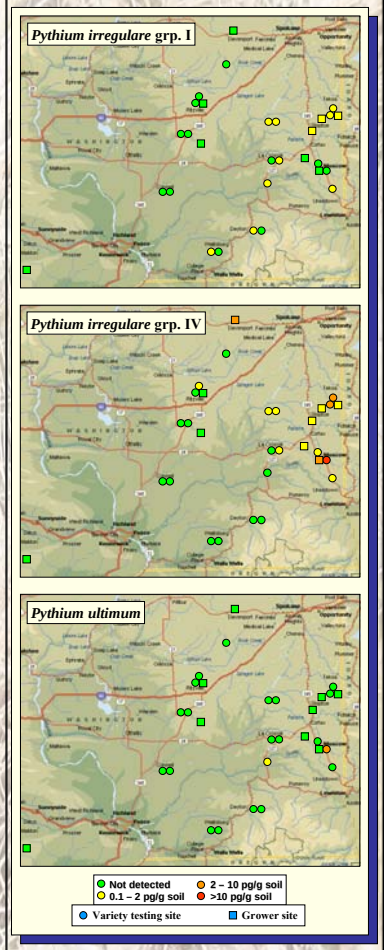


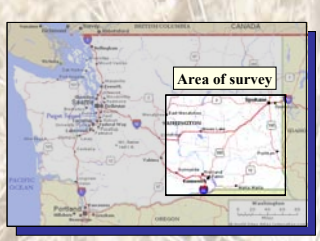
***RHIZOCTONIA* SPP.**

DNA quantities were mapped and categorized as not detected, <3 pg/g soil, or >3 pg/g soil for *Rhizoctonia* spp. *R. solani* AG-8 was ubiquitous, being detected at all sample locations. However, higher DNA quantities were only observed in Connell, Dayton, Ritzville and Walla Walla. *R. solani* AG-2-1, which is particularly virulent on brassicas, was observed at 6 of 7 sites that were planted to or had a recent history (2005 or 2006 growing season) of a brassica or legume in the rotation. Sites that did not have a recent history of brassica or legume production were not assayed, so the incidence of *R. solani* AG-2-1 needs further exploration. Like *R. solani* AG-8, *R. oryzae* also had a wide distribution, being detected in most regions that were sampled. However, a clear trend in distribution was not evident. *R. oryzae* was found at greater than 3 pg/g of soil in higher rainfall sites (Steptoe, Farmington – south of Tekoa, Garfield – northeast of Steptoe), an intermediate rainfall site (Harrington – south of Davenport) and at lower rainfall sites (Ritzville).

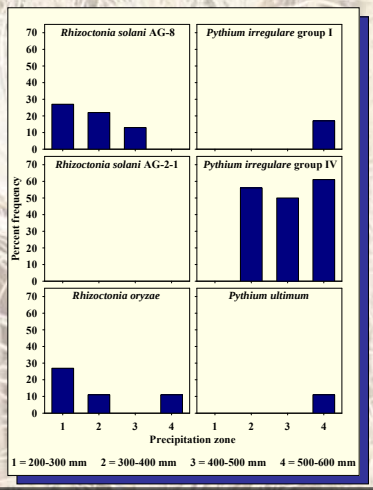
***PYTHIUM* SPP.**

DNA quantities were mapped and categorized as 0, 0 to 2, 2 to 10, or >10 pg/g of soil for *Pythium* spp. *P. irregulare* group I was observed at 12 of 30 locations. In all cases, the quantity of DNA detected was below 2 pg/g of soil. *P. irregulare* group I was found primarily in the higher rainfall regions toward the eastern side of Washington. The range of *P. irregulare* group IV was similar to that of group I, although higher DNA quantities were detected, with more than 10 pg/g of soil at one location in Pullman (west of Moscow). *P. ultimum* was seldom detected, being recovered from only two locations. One location was in Pullman (higher rainfall), while the second was an irrigated plot in Central Ferry (the only irrigated site in the study, south of La Crosse).





Annual precipitation across the dryland production areas of Washington ranges from 200 to 600 mm. Data presented in the graphs to the left were separated into precipitation zones shown in the figure below. Only sites that had a quantifiable level of the pathogen were included (>3 pg/g soil for *Rhizoctonia* spp. and >0.23 pg/g for *Pythium* spp.). *R. solani* AG-8 was most prevalent in the 200 to 300 mm zone (27% of sites). With increasing rainfall, the occurrence of *R. solani* AG-8 decreased, with no quantifiable detection in zones with greater than 500 mm precipitation. *R. solani* AG-2-1 was not detected at DNA quantities greater than 3 pg/g of soil, whereas *R. oryzae* was observed in most precipitation zones, with the highest incidence in zone 1 (200 to 300 mm). *P. irregulare* group I and *P. ultimum* were found only in the high rainfall zone (500 to 600 mm), in 17% and 10% of the sites respectively. *P. irregulare* group IV was absent in zone 1, but was present in zones with annual precipitation between 300 to 600 mm. In these zones, the frequency was greater than 50%.



CONCLUSIONS

- Highly virulent species of *Rhizoctonia* and/or *Pythium* were detected at all locations sampled.
- *Rhizoctonia solani* AG-8 (the pathogen causing bare patch of wheat and barley) is most prevalent in regions with less than 300 mm of precipitation.
- All *Pythium* spp. tested were primarily restricted to the 500 to 600 mm annual rainfall zone, although *P. irregulare* group IV had a greater range and was detected at a much higher frequency.