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Characterizing bean pod rot in Arkansas and Missouri

Jeremey H. Taylor and Craig S. Rothrock†*

ABSTRACT

Green beans are an important crop grown for processing in both Arkansas and Missouri. Green beans are harvested mechanically using non-selective picking fingers. Harvested beans are then transported in bulk to processing plants that are located at various locations throughout the mid-South. Thus, the crop is managed for high quality, avoiding pod blemishes caused by insects and diseases. One of the consistent quality problems that affect Arkansas and Missouri green bean crops is pod rot. Two of the causal agents of pod rot that have been reported by researchers and vegetable companies alike are *Pythium aphanidermatum* and an unidentified *Phytophthora* sp. In this study, 15 growers' fields were selected and soil samples (at planting), pod samples (at harvest), and environmental data were taken from each field. Disease incidence for field sites ranged from 0 to 7.3%. Pathogens associated with pod rot were *Sclerotinia sclerotiorum*, *Rhizoctonia solani*, a *Phytophthora* sp., and *Pythium* spp. The two suspected causal agents for pod rot, *Pythium* and *Phytophthora* spp., were found in all but one of the 12 field sites assessed for pod rot. *Pythium* inoculum potential, as determined by a baiting technique, was not a good indicator of pod rot incidence. In addition, soil temperature and water were not associated with pod rot. Pods collected at harvest having symptoms of pod rot were either in direct contact with the soil, senescing leaf tissue, or other diseased pods.

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† Craig S. Rothrock, faculty mentor, is a professor in the Department of Plant Pathology.

MEET THE STUDENT-AUTHOR

Jeremey H. Taylor

I graduated high school in spring 1999 from Cassville High School in Cassville, Missouri. I began my undergraduate work at the University of Arkansas in the fall of 1999 as a music education major and devoted three years of my college career to the razorback band. I was also active in Kappa Kappa Psi, a national honorary service fraternity for college bands and held several offices both local and on a district level. In the fall of 2001, I started in the pest management program and left music due to medical reasons. The switch was very difficult for me but one I am very glad I made. I finished the pest management program in December 2003.

I began this research as part of the Adair Internship program offered by the department of Plant Pathology in summer 2003, under the direction of Dr. Craig Rothrock. Through this program, I was able to conduct my own research, from methodology to analyzing results, and finally present my research. I am now a graduate student in plant pathology at the University of Arkansas under the direction of Dr. Craig Rothrock. I am very happy to say that if it had not been for the Adair internship, I probably would have never thought myself capable of pursuing a master's degree in plant pathology. I would like to express my appreciation to Dr. Rothrock for all of his cooperation and support on this project.

INTRODUCTION

In the central United States about 8900 ha of green beans, *Phaseolus vulgaris*, are grown each year. The processors of green beans require a large volume of high-quality product annually. In order to achieve this volume, crops must be harvested in a timely and economic manner that maintains high quality. Thus, crops are grown with little tolerance to insect damage or diseases. Bean crops are mechanically harvested with machines that use non-selective picking fingers which allow for no separation of diseased pods from healthy pods prior to transport. An increase in pod rot has been reported in the last 5 years in several of Allen Canning Company growers' fields (William Russell, personal communication). Some cultural practices that have been associated with this increase of pod rot are closer row spacings, consecutive plantings of snap bean, and increased fertilizer rates.

Pod rot is caused by several soilborne pathogens. Dr. John Damicone, Professor/Extension Plant Pathologist at Oklahoma State University, reported that pod rot was caused by at least two main casual agents, *Pythium aphanidermatum* and a *Phytophthora* sp. *Phytophthora nicotianae* var. *parasitica* is the cause of downy mildew pod rot on snap bean, which has been reported in Florida and North Carolina (Farr et al., 1995). However, the *Phytophthora* sp. that is associated with pod rot in the mid-South does not resemble *P. nicotianae* var. *parasitica* (John Damicone, personal communication). Diseases that result from these pathogens on bean also include seed rot and preemergence or postemergence damping-off of seedlings (Hall, 1991). Other pathogens associated with symptoms of pod rot are *Sclerotinia sclerotiorum* and *Rhizoctonia solani* (Hall, 1991).

Pythium and *Phytophthora* spp. are often called water molds, being favored by wet conditions in soil or on foliage (Erwin and Ribeiro, 1996). They grow as filamentous hyphae that are similar in appearance to fungi but these organisms are in the kingdom *Stramenopila*. These two genera are cosmopolitan and species may have a wide host range (Erwin and Ribeiro, 1996). Both pathogens produce long-lived sexual spores, oospores, and asexual structures and spores, sporangia and zoospores, respectively. The sporangia may germinate and infect directly similar to oospores or may produce

zoospores, which are the motile form of the pathogen. The zoospores, once released from the sporangium, swim to the host, encyst, and form a germ tube to infect the host (Ribeiro, 1978).

The study was designed to examine pod rot incidence and causal agents on green beans in Arkansas and Missouri. In addition, association of pod rot with field history, cultural practices, soil inoculum potential, and environment was examined.

MATERIALS AND METHODS

Fifteen field sites were selected that represented different cultivars and cultural practices used by growers for the Allen Canning Company in Missouri and Arkansas (Table 1). Histories of each field were documented based on fertility management, consecutive years planted in snap bean, row spacing, irrigation, and cultivars. In 12 of the 15 sites, environmental monitoring systems were placed to take soil temperature and water readings every 30 minutes.

A baiting technique was developed to examine *Pythium* inoculum potential. Approximately 30 arbitrary soil cores (2.5 cm in diameter by 15.0 cm deep) were taken along two diagonals from adjacent corners of each field at planting. For the baiting technique, 10 grams of soil from a single field were placed in the bottom of a plastic container (13.9 cm by 10.1 cm by 3.8 cm). A total of 150 mL of sterilized water was added to the container. Finally, 'Roma II' bean pods were washed in a 0.5 % NaOCl solution for 1.5 min, blotted dry on a paper towel, cut into 1-cm sections, and 12 pieces placed on a screen for 24 h in the dark. The screen was placed over the soil to separate the soil from the pod sections. Three plastic containers, serving as replicates, were run per field. After 24 h, the 12 pod pieces from each container were transferred to water agar plates and monitored. Developing colonies were transferred from the water agar plates to potato-dextrose agar (PDA) plates. Once growth was established on PDA plates, cultural characteristics were recorded. To determine which of the cultures were virulent, a pathogenicity assay was conducted. One flat, 'Roma II' bean pod was used for every isolate that was suspected to be a *Pythium* or *Phytophthora* spp. A 1-cm wound was made using a sterilized scalpel at the bottom of the pod and inoculated by placing a portion of a culture in the wound. The inoculated flat beans were placed on wet paper towels in a sealed container to maintain high humidity. The containers were placed in the dark at room temperature (approximately 21°C) for 3 days at which time pods were examined and measured for lesion length from the point of inoculation.

Prior to harvest, each field was scouted and sampled. In each field, 15 sample sites, arbitrarily selected but representing the geographical layout of the field, were inspected for diseased pods. At each site, 40 pods were examined for a total of 600 pods per field. Diseased pods were placed in individual bags to avoid contamination and labeled by site. The diseased pods were taken back to the Plant Pathology Department laboratory, rinsed with deionized water, blotted dry with a paper towel, and diseased pod tissue placed on P5ARP, a selective medium for *Pythium* and *Phytophthora* (Jeffers and Martin, 1986), and water agar plates. Once growth had occurred, cultures were transferred to PDA. From the PDA plates, cultures were identified or a plug was transferred to corn meal agar (CMA) for identification of *Pythium* and *Phytophthora* isolates. In order to identify *Pythium* and *Phytophthora* isolates, three strips were cut from a CMA plate and placed in a Petri dish that contained sterilized water and wheat (*Triticum aestivum*) leaves. Wheat leaves were sterilized by autoclaving twice for 20 min at 24 h intervals. The plates were placed in the dark for 2 days. On the third day, slides were made from the wheat leaves and isolates identified based on asexual and sexual fruiting structures (Erwin and Ribeiro, 1996; Van Der Plaats-Niterink, 1981). All *Pythium* and *Phytophthora* isolates were tested for pathogenicity.

RESULTS AND DISCUSSION

Incidence of pod rot averaged 1.4 %, ranging from 0 to 7.3 %, across the 12 locations (Table 1). Three out of the 15 fields were harvested prior to disease assessment. In field sites JT5, JT9, and JT12, no diseased pods were found (Table 1). A *Phytophthora* sp. was recovered from pods in only one of the field sites whereas *Pythium* spp. were isolated in seven of the 12 fields (Table 2). All but one of the isolates from diseased pods, site JT4, were virulent (Table 2). *Rhizoctonia solani* was recovered from diseased pods in four fields. In three of these fields, *R. solani* was recovered from 25% or more of diseased pods (Table 2). *Sclerotinia sclerotiorum* was recovered from three fields, with this pathogen being isolated from 39 to 67% of diseased pods (Table 2). Speciation of *Pythium* isolates resulted in at least two *Pythium* species, *P. ultimum*, and *P. aphanidermatum*. All *Phytophthora* isolates had similar appearance, including sporangium size, in this study.

The results for the soil assays showed *Pythium* spp. were detected in soil from all 15 fields (Table 2). Pathogenicity tests resulted in virulent isolates from each of the soil samples. Although inoculum was present in each field, no association was found between inoculum potential and disease incidence for the fields. No associ-

ations between the environmental data collected (soil temperature and soil moisture) and incidence of pod rot were observed.

While conducting this study, several observations were made regarding the occurrence of pod rot. Pods that had symptoms of pod rot were either in direct contact with soil, dead or senescing leaf tissue, or other diseased pods. 'Roma II' had greater pod rot and plants tended to be more prostrate than with the cultivar 'Nelson.' Cultural practices, including row spacing, fertilizer use, and canopy cover, did not consistently influence disease incidence. In fields that used irrigation, disease tended to be present more consistently.

In conclusion, the pathogens *Pythium aphanidermatum* and *Phytophthora* sp. were found associated with pod rot. Future research should be directed toward examining cultural practices such as irrigation, row spacing, tillage (conventional versus no-tillage), and fertility to clarify their effect on disease. Furthermore, plant architecture, including pod location, should be investigated for the possibility of selecting a more erect plant as opposed to having a plant with a prostrate growth habit, as found for 'Roma II'. Lastly, environmental conditions that favor pod rot need to be determined. In this study, data were taken on soil moisture and temperature only. Environmental data should include air temperature, leaf wetness, and rain fall. The clarification of factors important to pod rot development will aid in the management of this disease through plant genetics and changes in cultural practices.

ACKNOWLEDGMENTS

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Table 1. Field information and pod rot incidence.

Field designation	Location	Field history[y]	Cultivar	Row spacing (m)	Fertility[z]	Canopy cover (%)	Pod rot incidence(%)
JT1	Washington Co., Ark.	2	Roma II	0.91	0	86	0.7
JT2	Benton Co., Ark.	>3	Roma II	0.91	1	89	3.0
JT3	Benton Co., Ark.	>3	Roma II	0.91	1	88	0.7
JT4	Benton Co., Ark.	2	Nelson	1.02	1	82	2.0
JT5	Newton Co., Mo.	1	Roma II	0.91	2	93	0
JT6	Newton Co., Mo.	1	Nelson	1.02	1	61	1.3
JT7	Newton Co., Mo.	>3	Roma II	0.91	1	100	7.3
JT8	Newton Co., Mo.	>3	Roma II	0.91	1	79	2.7
JT9	Newton Co., Mo.	>3	Nelson	0.91	1	100	0
JT10	Newton Co., Mo.	>3	Nelson	0.91	2	86	1.3
JT11	Newton Co., Mo.	1	Roma II	0.91	2	96	1.7
JT12	Newton Co., Mo.	>3	Roma II	1.02	1	81	0
JT13	Benton Co., Ark.	>3	Nelson	0.91	2	0	-
JT14	Benton Co., Ark.	>3	Roma II	0.91	1	0	-
JT15	Newton Co., Mo.	>3	Nelson	0.91	2	0	-

[y] Number of consecutive years of bean production

[z] Level of N-fertility; 0 = below average, 1 = average, 2 = above average

Table 2. Pathogen inoculum potential and pod rot pathogen recovery

Field designation	Soil baiting of *Pythium* isolates (%)[y]		Pathogen recovery (%)			
	Total	Virulent	*Pythium* spp.(virulence)[z]	*Phytophthora* sp.	*Rhizoctonia solani*	*Sclerotina sclerotiorum*
JT1	72	36	100 (1.0)	0	0	0
JT2	86	34	50 (4.2)	0	0	39
JT3	69	33	100 (4.0)	0	0	0
JT4	53	25	1 (0)	0	1	67
JT5	28	14	0	0	0	0
JT6	53	17	75 (2.0)	0	25	0
JT7	31	17	0	16 (1.2)	55	0
JT8	33	11	94 (2.0)	0	1	0
JT9	39	11	0	0	0	0
JT10	28	17	100 (2.1)	0	0	0
JT11	67	1	0	0	50	40
JT12	19	3	0	0	0	0
JT13	47	25	-	-	-	-
JT14	42	22	-	-	-	-
JT15	31	14	-	-	-	-

[y] Isolation from three replicates of 12 pod sections each per soil sample; isolates were tested by a bean pod pathogenicity test for virulence.

[z] Mean expansion of lesion from the point of inoculation after 3 days