The Journal of Rural and Agricultural Research Vol. 14 No. 1, 12-14 (2014)
Received February 2013; Acceptance Januuary 2014

Pathogenicity and Characterization of *Rhizoctonia solani* Kühn Inciting Wet Root Rot in Chickpea

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Abstract

Eight isolates of Rhizoctonia solani Kühn, the incitant of Wet root rot of chickpea were assessed for pathogenicity. Pathogenicity of these isolates varied both in sterilized and unsterilized soils when tested on chickpea variety C-235. All the isolates were pathogenic and isolate Bikaner was highly virulent. The mortality of plant in sterilized soil was higher. There was no correlation between the cultural characters and virulence in any of the isolates.

Key words: Chickpea, Rhizoctonia solani, Variation, Pathogenicity

Introduction

Wet root rot of chickpea caused by *Rhizoctonia solani* Kühn is one of the serious disease in chickpea growing region of Rajasthan and adjoing states of India. The disease symptoms are characterized by sudden and complete wilting of the plants. The infected plants can easily be pulled out. No information is available with regards to pathogenicity in *R. solani* isolates in terms of variability. In India, use of molecular markers in characterization of plant pathogens have been recently reviewed (Monga *et al*., 2004). The present investigations is aimed at differentiation of isolates of *Rhizoctonia solani* collected from various regions of Rajasthan, Delhi and Haryana by using pathogenicity studies.

Materials and Methods

Collection of isolates

Out of eight isolates of *R. solani*, Four isolates were collected from chickpea infested field of different districts of Rajasthan *viz.* Bikaner, Hanumangarh, Nagaur, Sriganganagar and one each from Haryana *viz.* Choudhary Charan Singh, Haryana Agricultural University, Hisar, Krishi Vigyan Kendra, Karnal, Agricultural Research Station, Gurgaon and one from Indian Agricultural Research Institute, New Delhi. Pure culture of these isolates obtained through single hyphal tip method, were maintained on potato dextrose agar slants for further investigation.

Isolation, Purification and Identification

The samples collected from diseased plants were used for isolation. The roots were thoroughly washed with tap water to remove soil. Small pieces of about 0.5 cm length were surface sterilized with 0.1 per cent mercuric chloride solution for 2 minutes, three washings with sterilized distilled water were given, placed on PDA slant under a laminar flow and incubated at 28 ± 1^0C temperature for seven days.

To maintain the pure culture of *R. solani* single hyphal tip isolation technique was adopted. One ml of suspension having 5-6 pieces of hypha per 10 x microscopic field were spread over 2 per cent plain agar in Petri dishes evenly by tilting the Petri dishes clockwise as well as anticlockwise. The excess amount of suspension was decanted and Petri dishes were incubated at 28 ± 1^0C for 24 hours. The single piece of hypha was demarcated under low power of microscope (10 X) and cut with the help of mechanical cutter. Individual piece of hypha was transformed on PDA slants with the help of an inoculating needle. The inoculated slants were kept in B.O.D. incubator for growth at 28 ± 1^0C for 7 days. Thus, the purified cultures were maintained by periodical transfers on PDA slants and used for further studies.

Pathogenicity test

Pathogenicity of these isolates was tested both in sterilized and unsterilized soil on *Cicer arietinum* L. variety C-235. Inoculum of each isolate was multiplied on sterilized sand maize flour medium (partially broken maize grains 10g, sand 10g, and 20ml distilled water in each 250 ml Erlenmeyer's flask). The flask containing the sterilized media was inoculated with mycelial disc of *R. solani* (5 mm diameter) and inoculated at 28±1^0C for 15 days. These inocula were used for soil inoculation in ratio of 1: 200 w/w basis and was added in disinfected burnt earthen pots (25 cm) maintained in triplicates. Seed samples were surface sterilized with 0.1 per cent mercuric chloride for 30 second. Five seeds of susceptible variety C-235 were sown in each pot. Observations were

recorded after 40 days of sowing.

Results and Discussion

Wet root rot of chickpea, caused by *R. solani* is an important disease mainly in Bikaner and Sriganganagar districts of Rajasthan. The affected seedlings showed gradual yellowing and the petiole leaflets droop without collapse of seedlings. The stems and roots near the lesions show rotting frequently with pinkish mycelial growth. Sclerotia were usually seen.

Pathogenicity of isolates

All eight isolates tested for their pathogenicity towards C-235 variety of chickpea were found virulent in sterilized and in unsterilized soil. Koch's postulates were proven for each isolate. Symptoms of disease showed gradual yellowing and the petiole leaflets droop without collapse of seedlings (Plate.1). The stems and roots near the lesions show rotting frequently with pinkish mycelial growth. Sclerotia were usually seen. Symptoms of wet root rot appeared after 40 days of germination in sterilized soil while it was delayed upto 52 days in unsterilized soil. The root rot incidence (%) in sterilized and unsterilized soil varied from 40.00 to 86.67 and 46.67 to 66.67, respectively (Table.1). Isolate Bikaner was highly virulent, followed by isolates Delhi, Hisar, Karnal, Sriganganagar, Hanumangarh, Gurgaon and Nagaur. In unsterilized soil the reaction of isolates was less and delayed as compared to sterilized except isolate Gurgaon and Nagaur was at par in both type of soil.

Root rot was found as a major disease of chickpea in different districts of Rajasthan state, as reported by Singh and Sirohi (2003) in Madhya Pradesh in India. Aghakhani and Dubey (2009) differentiated 23 isolates of *Rhizoctonia bataticola* incitant of root rot of chickpea and observed variability in their morphological, cultural characteristics and pathogenicity as found in present investigations. In unsterilized soil the reaction of isolates was less and delayed as compared to sterilized one. The mortality of plants in unsterilized soil was reduced except isolates Gurgaon and Nagaur where mortality found increased over sterilized soil. There was practically no direct correlation between the cultural characters and virulence in any of the isolates. Prameela Devi and Singh (1998) categorized 56 isolates of *M. phaseolina* obtained from blackgram and greengram crops collected from 11 different location of North, South, North-East and central India. They observed higher incidence of root rot in sterilized soil than in unsterilized soil in both the crops as investigated in the present studies.

Table 1: Pathogenicity of eight isolates of *Rhizoctonia solani* on C-235 variety of chickpea in sterilized and unsterilized soil.

Isolate	Disease incidence in per cent		
	Sterilized	Unsterilized	Mean
Bikaner	86.67	66.67	76.67
	(72.29)	(54.99)	(63.64)
Sriganganagar	60.00	46.67	53.33
	(50.77)	(43.08)	(46.92)
Hanumangarh	53.33	46.67	50.00
	(46.92)	(43.08)	(45.00)
Nagaur	40.00	53.33	46.67
	(38.86)	(47.30)	(43.08)
Hisar	60.00	46.67	53.33
	(51.14)	(43.08)	(47.11)
Karnal	60.00	46.67	53.33
	(50.77)	(43.08)	(46.92)
Gurgaon	40.00	60.00	50.00
	(39.23)	(50.77)	(45.00)
Delhi	73.33	46.67	60.00
	(59.21)	(43.08)	(51.14)
Mean	59.17	51.67	55.42
	(51.15)	(46.06)	(48.60)
	S.Em±	CD (P=0.05)	CD (P=0.01)
Isolates	3.38	9.58	12.75
Soil types	1.69	4.79	6.37
Isolates x s oil types	4.78	13.54	18.03
CV (%)	3.38	9.58	12.75

Figure in parenthesis are mean angles corresponding to percentage

Plate 1. Pathogenicity of isolates of *Rhizoctonia solani*

A - Wet root rot infected plants in sterilized soil
B - Wet root rot infected plants in unsterilized soil
C - Control

Plate 2. Variability in isolates of *Rhizoctonia solani*

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