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Survey, incidence and integrated disease management of cotton root rot caused by *Rhizoctonia solani* (Kuhn.)

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Abstract
Field survey of six major cotton growing districts of Rajasthan was carried out during *Kahrif* 2017 to access the incidence and collection of root rot samples. The average plant mortality due to the fungus *R. solani;* was recorded in the range of 12.29 to 28.36 percent at Borwat (Banswara) and Bhadwalo villages of Sri Ganganagar, respectively. Systemic fungicide Carbendazim and bio control agent namely *Trichoderma viride* strain (T-5) found promising under *in vitro* condition among all tested fungicides and bio-control agents; thus both were tested under *in planta* condition with integration of neem cake and found that maximum germination percent (73.0 percent) with minimum percent mortality (5.5 percent) were recorded in treatment combination of seed treatment with Carbendazim and soil application of *T. Viride* + neem cake as compared to other treatments.

Keywords: *Rhizoctonia solani*, integrated disease management, Carbendazim, Trichoderma viride, disease incidence

Introduction
Cotton (*Gossypium* spp.) is one of the most important fibers and cash crop belongs to genus *Gossypium* of the family *Malvaceae.* It is originated as a tropical and subtropical perennial plant, but is produced as an annual crop in many temperate regions around the world. Cotton is cultivated in about 80 countries in the world in which top five producers are India, China, Pakistan, USA and Brazil (Anonymous, 2016) [1]. Cotton cropping provides 60 percent of the fiber used in our textile industries, supplies more than 1 million metric ton of cooking oil, and another million metric ton of quality animal feed and 40 million metric tons of biomass in the form of cotton stalks (FICCI, 2012) [4].

Most common fungal diseases in cotton are leaf spot and leaf blight caused by (*Alternaria macrospora*, *Alternaria alternata*, *Cercospora gossypina*, *Cochliobolus spicifera*, *Myrothecium roridum* (Kamal and Moghal, 1968; Jagirdar and Jagirdar, 1980; Jiskani, 1992, 2001) [9, 10, 12]. Anthracnose (*Colletotrichum gossypii*), Areolate mildew (*Cercosporella gossypii*), Ascochyta blight (*Ascochyta gossypii*) and black root rot (*Thielaviopsis basicola*), boll rot is caused by several pathogen including (*Ascochyta gossypii*, *Colletotrichum gossypii*, *Fusarium spp*., *Lasiodiplodia theobromae*, *Rhizoctonia solani*). Under favorable environmental condition Charcoal rot (*Macrophomina phaseolina*), Fusarium wilt *Fusarium oxysporum* f. sp. *vasinfectum)*, powdery mildew (*Leveillula taurica*), cotton rust (*Puccinia schedonnardii*, *Puccinia cacabata*), Sclerotium stem rot (*Sclerotium rolfsii*) and Damping-off and root rot (*Rhizoctonia solani* (Silva *et al*., 1995) [24], (Belot and Zambiasi, 2007) [2]. Among the seedling diseases, damping-off and root rot of cotton caused by *Rhizoctonia solani* showed heavy losses particularly during the early stage of crop growth (Nawar, 2008) [19]. Plant stand and significant losses in cotton production throughout the cotton growing countries of the world (Michael *et al*., 2007) [15]; (Koenning, 2008) [13]. *Rhizoctonia* is a widespread, destructive and versatile plant pathogen, are distributed worldwide in both agriculture and forest soils and are known to cause root diseases of several crop plants (Garcia *et al*., 2006) [5]. The severity of this disease varies from locality to locality and attends maximum intensity (up to 90%) during rainy season in cotton (Singh and Verma, 1988) [25]. The soil moisture of 20 percent and temperature of 35 °C have been reported most suitable for infection (Haq *et al*., 1999) [7]; (Monga and Sheo Raj, 1994) [16].

Seed treatment with fungicides is the appropriate method for control the seed/soil borne diseases but, the fungus *R. solani* is non-spore forming and soil borne, it's propagules and sclerotia are evenly distributed in the soil and fungicides are not able to reach at the target point for manage the disease up to longer period.

On other hand overzealous and indiscriminate use of most of the synthetic fungicides has created environmental pollution and toxicological problems. Attention has been paid towards exploitation of non-hazardous bio control agents or botanical amendment in plant protection. Therefore, the ultimate aim of research has been the development of integrated control strategies, as such; use of alternative methods like eco-friendly botanicals and biological control with integration of fungicides to manage this disease.

Materials and Methods

The investigations on "Integrated Management of Root Rot of Cotton Caused by *Rhizoctonia solani* (Kuhn)" were carried out at the Department of Plant Pathology, Rajasthan College of Agriculture (RCA), Udaipur during 2016-17. The details of experimental techniques followed and the methods and materials used during the course of studies are being described in this chapter.

Survey for incidence and collection of disease samples

The diseased samples of cotton showing typical root rot symptoms were collected in *Kharif* 2017 from farmer's fields of different cotton growing areas of Rajasthan *viz*., Udaipur, Dungarpur, Banswara and Chittorgarh (Southern Rajasthan) and Shri Ganganagar, Hanumangarh districts (Northern Rajasthan) all from local land races. The main aim was to explore possibility of existence of different species and/or variables of root rot pathogen and incidence caused by them. The infected plants were carefully uprooted and placed in polythene bags, properly tagged and brought to the laboratory and subjected to microscopic examination and tissue isolation.

Isolation, purification and identification of pathogen

The pathogens were isolated on potato dextrose agar (PDA) medium. Small pieces (1-2 mm) of diseased roots were cut, washed with sterilized water, surface sterilized with 0.1 percent sodium hypochlorite (NaOCl) solution for 1 minutes followed by three to four washings with sterilized distilled water and were transferred aseptically to 2 percent PDA (Potato Dextrose Agar) poured Petri-plates. The plates were incubated in an incubator at 28 ± 1 ^{0}C for 7 days. Hyphae coming out from the bits were sub-cultured on the fresh PDA in Petri dishes. From these bits mostly cultures of *Rhizoctonia solani*, *Sclerotium* spp. and *Fusarium oxysporum* were recovered. The culture of *Rhizoctonia* was purified by single hyphal-tip method and that of *Fusarium* by single spore method using a dummy objective. The cultures were identified by comparing the morphological and cultural characters described in standard references (Mordue, 1988) [17] for *Rhizoctonia* and (Booth, 1971) [3] for *Fusarium*, and were identified as *Rhizoctonia solani* and *Fusarium oxysporum.*

Evaluation of bio-control agents, neem cake and fungicides against *R. solani* under *in planta* condition

Multiplication of *R. solani* for soil application

For the micro plot experiment, virulent isolate of *R. solani* were multiplied on autoclaved sorghum grains for 3 weeks. This inoculum soil mixture was mixed with field soil in micro plots @ 10 g/plot (1.8 x 1.8 m^2). For each treatment three plots as three replications were maintained. All the plots were lightly irrigated immediately after inoculation. The plots were kept free for 10 days, to allow establishment of the pathogen before sowing.

Seed and soil application of bio-control agents, neem cake and fungicides

Seed treatment with bio-control agent

For seed treatment, cultures of the bio-control agents were individually grown on 2 percent malt extract agar (MEA), The spores colonies so developed were harvested by suspending in 20 ml water in each Petri dish and mixed with sterilized fine clay (talc powder) 10 gm to make a slurry. This formulation of the BCA was used for seed treatments @ 8 g/kg seed. The coated seeds were kept overnight in moist chamber so as to enable the antagonists to establish on seeds.

Soil application of bio-control agent

Hundred kilograms well decomposed farm yard manure inoculated with 2 kg of *Trichoderma viride* (T-5) powder and incubated for 15 days and thereafter mixed in soil @ 50 g/plot.

Fungicidal seed treatment

Since as only small quantity of fungicide to be used for seed treatment, according to compatibility of bio-control with fungicide, the seeds were soaked in Carbendazim 50 WP @ 0.1% solution for 30 minutes. The treated seeds were air dried in shade and then sown.

Sowing and inter-culture operations

Treated seeds of cotton cultivar "Jai BG-II" were sown in micro plots (60 x 60 cm spacing) keeping three replications for each treatment. Agronomic practices or inter-culture operations (*viz*., recommended dose of NPK, weeding, hoeing) as follows as prescribed.

Experiment details

Design: RBD
Replication: 3
Spacing: 60 X 60 cm
Treatments: 8

Treatment details

T_1 Carbendazim (ST)
T_2 *T. viride* (ST)
T_3 Neem cake (SA)
T_4 *T. viride* (ST) + Neem cake (SA)
T_5 Carbendazim (ST) + Neem cake (SA)
T_6 Carbendazim (ST) + *T. viride* (SA)
T_7 Carbendazim (ST) + *T. viride* (SA) + Neem cake (SA)
T_8 Control

Observations

The numbers of total germinated seedlings were recorded 10 days after sowing. The numbers of root rot infected plants (post-emergence mortality) were recorded from germination to 90 days after sowing (*viz*., 45, 60 and 90 DAS, respectively).

$$\text{Percent mortality} = \frac{\text{Number of infected plants}}{\text{Total number of plant observed}} \times 100$$

Results and Discussion

Survey: Field survey was carried out during *kharif* 2017 to assess the incidence of root rot of cotton and observed that average percent plant mortality vary from 12.29 (minimum) to 28.36 (maximum) in Borwat village of Banswara and Bhadwalo of Sri Ganganagar, respectively, which indicates wide distribution and occurrence in large area.

Table 1: Percent incidence of cotton root rot caused by *R. solani* in different cotton growing districts of Rajasthan.

Name of districts	Name of villages	Percent root rot incidence*	Isolated pathogen	Isolate code
1. Udaipur	1. Mawali	19.43 (26.14)	*R. solani*	UDP Rs-01
	2. Intali	21.55 (27.64)		
	3. Nawania	23.68 (29.10)		
2. Hanumangarh	1. Sherda	17.68 (24.86)	*R. solani*	HMG Rs-02
	2. Dobhi	19.37 (26.10)		
	3. Suratpura	22.41 (28.24)		
3. Sri- Ganganagar	1. Thandi	25.34 (30.21)	*R. solani*	SGN Rs-03
	2. Thakri	26.52 (30.98)		
	3. Bhadwalo	28.36 (32.16)		
4. Chhitorgarh	1. Kapasan	21.42 (27.56)	*R. solani*	CHT Rs-04
	2. Heeraji Khera	23.30 (28.85)		
	3. Ajan Khera	27.31 (31.49)		
5. Banswara	1. Borwat	12.29 (20.51)	*R. solani*	BNS Rs-05
	2. Dahod	14.27 (22.18)		
	3. Salia	17.42 (24.65)		
6. Dungurpur	1. Amajara	15.44 (23.12)	*R. solani*	DNG Rs-06
	2. Balwara	17.22 (24.51)		
	3. Deval Khas	19.23 (26.00)		
S.Em ±		0.133		
CD (P = 0.05)		0.38		

*Average of three fields surveyed in each village.

The results depicted in (Table-1) showed that maximum percent plant mortality 28.36 was recorded at Bhadwalo village of Sri Ganganagar district followed by Ajan Khera (27.31) village of Chittorgarh and 26.52 percent at Thakri village of Sri Ganganagar district, whereas 25.34 percent plant mortality was found at Thandi (Sri Ganganagar), 23.68 percent in Nawania village of Udaipur district, Heeraji Khera (23.30), Suratpura (22.41), Intali (21.55), Kapasan (21.42), Mawali (19.43), Dhobi (19.37), Deval Khas (19.23), Sherda (17.68), Salia (17.42), Balwara (17.22), Amajara (15.44), Dahod (14.27) percent of Banswara. Various workers have given variable reports (Gazaway, 1998) [6]; (Mathur and Gurjar, 1995) [14]; (Rani *et al*., 2013) [20] and (Rothrock, 1996) [23].

Integrated management of root rot of cotton

Percent germination

The fungicide and bio-control agent which was found most effective under *in vitro* study, were further tested in the field for the suppression of root rot of cotton in alone as well as in various combinations. The results depicted in (Table-2) where all treatments found superior over control (55.5 percent). Maximum 73.0 percent germination was recorded in treatment combination of [Carbendazim (ST) + (*T. viride* + neem cake) (SA)], followed by [Carbendazim (ST) + *T. viride* (SA), whereas no significant difference found among the treatment where application of [Carbendazim (ST) + neem cake (SA)] and [*T. viride* (ST) + neem cake (SA)]. While; alone application of Carbendazim (ST) showed 64 percent seed germination, followed by soil application of neem cake 60.0 and *T. viride* (ST) 57.1 percent.

Percent mortality

The data revealed in (Table-2) that the minimum plant mortality 5.5 percent till 90 DAS was recorded in treatment combination where Carbendazim was applied as seed treatment (ST) and *T. viride* + neem cake were applied in the soil (SA), followed by Carbendazim (ST) + *T. viride* (SA) with percent mortality of 10.5 at 45 and 11.4 percent at 60 and 90 DAS, respectively, whereas; Carbendazim (ST) + neem cake (SA) was found least effective among all treatment combinations with 15.0 percent plant mortality at 45 DAS, 17.2 percent constantly at 60 and 90 days each, respectively. Among individual application of different treatments; Carbendazim (ST) was showed 20.0, 28.3 and 32.7 percent plant mortality at 45, 60 and 90 DAS, respectively, whereas; *T. viride* (ST) showed 23.5 percent at 45 and 29.3 at 60 and 90 DAS, respectively. Highest percent plant mortality i.e. 24.5, 32.8 and 35.5 were recorded in soil application of neem cake only at 45, 60 and 90 DAS, respectively. Similar results were also reported by (Howell, 2007); (Nagamani *et al*., 2011) [18]; (Rawal *et al*., 2013) [21] and Vyas, (1994) [26].

Table 2: Effect of seed treatment and soil application of fungicide, bio-control agent and neem cake on germination and plant mortality of cotton in micro-plots.

Treatments	Germination percent	Mortality percent		
		Upto 45 days	Upto 60 days	Upto 90 days
Carbendazim (ST)	64.0 (53.1)	20.0 (26.6)	28.3 (32.1)	32.7 (34.8)
T. viride (ST)	57.1 (49.1)	23.5 (29.0)	29.3 (32.8)	29.0 (32.6)
neem cake (SA)	60.0 (50.8)	24.5 (29.7)	32.8 (34.9)	35.5 (36.6)
T. viride (ST) + Neem cake (SA)	67.0 (54.9)	14.0 (22.0)	22.3 (28.2)	28.7 (32.4)
carbendazim (ST) +Neem cake (SA)	67.0 (54.9)	15.0 (22.8)	17.2 (24.5)	17.2 (24.5)
carbendazim (ST) + *T. viride* (SA)	70.0 (56.8)	10.5 (18.9)	11.4 (19.7)	11.4 (19.8)
carbendazim (ST) + *T. viride* (SA) + Neem cake (SA)	73.0 (58.7)	5.5 (13.6)	5.5 (13.6)	5.5 (13.6)
Control	55.5 (48.2)	33.3 (35.3)	63.9 (53.0)	68.0 (55.5)
S.Em ±	0.3831	4.185	0.220	0.167
CD (P= 0.05)	1.16	0.69	0.67	0.51

*Average of three replications. Figures in parentheses are arcsine √ percent angular transformed values. (ST- seed treatment; SA- soil application).

Conclusion

Incidence of root rot of cotton caused by *Rhizoctonia solani* in different agroclimatic zones of Rajasthan were varied from 12.29 to 28.36 percent during Kharif 2017. Among different treatments the minimum plant mortality 5.5 percent till 90 days after sowing was recorded where Carbendazim was applied as seed treatment and *T. viride* + neem cake were applied in the soil before sowing.

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