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INCIDENCE OF DISEASES IN GERMPLASMS OF *CORCHORUS OLITORIUS* L. AND CONTROL OF FUNGAL PATHOGENS

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Abstract

Screening of 40 germplasms and two released varieties of *Corchorus olitorius* L. was made to determine the source of resistance in jute germplasm against various diseases under condition of natural infection in the field. The germplasm lines were classified on the basis of their per cent disease incidence according to the scale of Mayee and Datar with some modification. There were 12 germplasm lines of Acc. Numbers 1045, 1050, 1060, 1062, 1065, 1143, 1261, 1338, 3711, 3724, 4178, 5009 and variety O-72, found resistant to jute diseases which can be utilized for developing disease resistant variety. The effects of Bavistin DF, Capvit 50 WP, Dithane M-45, Hayvit 80 WP, Ridomil Gold MZ 68 WG and Tilt 250 EC were evaluated against *Botryodiplodia theobromae, Colletotrichum* sp., *Fusarium* sp. 1, *Fusarium* sp. 2, *Rhizoctonia solani* and *Sclerotium rolfsii* at different concentrations following poisoned food technique. Out of six fungicides, Bavistin, Dithane and Tilt completely inhibited the growth of test pathogens except *Sclerotium rolfsii* at 500 ppm. Bavistin, Dithane and Tilt also showed promising results against the selected test pathogens at 100, 200 and 400 ppm. The findings of this investigation will be helpful for the plant breeders to select varieties for breeding purposes as well as to control jute pathogens.

Introduction

Corchorus capsularis L. and *C. olitorius* L. are commonly known as jute in Bangladesh. Jute (*Corchorus* spp.) is the second most important fiber after cotton, in terms of usage, global consumption, production and availability. It is a very important cash crop of Bangladesh. Among the jute growing countries of the world, Bangladesh ranks second in respect of production. In 2014-15, 7.5 million bells of jute were produced in the country from 1.66 million acres of land(1). Major constraints of jute production are due to various diseases in which fungal diseases are most important one. It has been estimated that about 60% yield of jute fiber are lost due to the incidence of fungal diseases alone every year. Major diseases of jute plants in Bangladesh are stem rot (*Macrophomina phaseolina*), anthracnose (*Colletotrichum corchori* and *C. gloeosporioides*), soft rot (*Sclerotium rolfsii*), black band (*Botryodiplodia theobromae*), die-back (*Gloeosporium* sp.), root rot (*Rhizoctonia* sp.) and powdery mildew (*Oidium* sp.). They are not only responsible for yield loss but also deteriorate the quality of fiber and seeds(2).

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Germplasm is the major source of genetic resources of any crop. At present there are 6060 germplasms including 4084 of *Corchorus* (15 species), 1512 of *Hibiscus* (22 species), 345 of allied genera (15 species) and 119 of interspecific hybrid preserved in Genetic Resources and Seed Division of Bangladesh Jute Research Institute. Plant breeders are always searching disease resistant traits by screening germplasm time to time as their routine work. Very few reports are available on the screening of germplasm against various diseases of jute. So, it is very important to screen plant genetic resources of jute, so that disease resistant germplasm line can be recommended for further crop improvement program or directly released as disease resistant variety.

Nowadays, many inorganic and organic fungicides are used to control plant diseases(3). Various workers in different countries of the world evaluated the efficacy of various fungicides against *Colletotrichum* spp., *Macrophomina phaseolina*, *Fusarium* spp., *Botryodiplodia theobromae*, *Colletotrichum gloeosporioides*, *Sclerotium rolfsii* and *Alternaria* spp., under laboratory and field conditions(4,5,6). Very few works have been done for the control of fungal pathogens associated with various diseases of *C. olitorius* by fungicides. Hence, the present work was undertaken to find out disease resistant germplasm by screening plant genetic resources and *in vitro* fungitoxicity of fungicides of fungal pathogens of *C. olitorius*.

Material and Methods

Seeds of 40 different germplasm lines and two released varieties of *Corchorus olitorius* were collected from Genetic Resources and Seed Division of Bangladesh Jute Research Institute (BJRI), Manik Mia Avenue, Dhaka. Experiment was conducted at BJRI Agricultural Experimental Station, Manikganj and Mycology and Plant Pathology Laboratory, Department of Botany, University of Dhaka during April, 2013 to June, 2014. Jute seeds were sown in the 2nd April, 2013 in line at 3 cm depth, maintaining 20 cm seed to seed and 30 cm line to line distances. The block to block distances were 1 m from each other. Per cent disease incidence of plant in each individual line was calculated by using following formula:

$$\% \text{ disease incidence} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 100$$

After counting the per cent disease incidence of plants were categorized as immune, resistant, moderately resistant, moderately susceptible, susceptible and highly susceptible according to the scale given by Mayee and Datar(7) with some modifications (Table 1).

Disease samples were collected in separate sterile polyethylene bags, labeled properly and then brought to the laboratory for isolation of fungi following “Tissue planting method" on PDA medium[8]. Identification of the isolates was determined following standard literatures[9-13].

Six fungicides *viz.,* Bavistin DF (50% Carbendazim), Capvit 50 WP (Cupper oxychloride), Dithane M-45 (80% Mancozeb), Hayvit 80 WP, Ridomil Gold MZ 68 WG (4% Metalaxyl and 64% Mancozeb) and Tilt 250 EC (Propiconazole) were selected to evaluate their *in vitro* efficacy against the test pathogens following poisoned food technique[14]. Firstly, *in vitro* fungitoxicity of these six fungicides at 500 ppm was evaluated against the six test pathogens to screen out the effective fungicides. The fungicides which were effective at 500 ppm were further tested in 100, 200 and 400 ppm to find out their minimal inhibitory concentration.

Table 1. Categorization of plants on the basis of percentage of disease incidence (After Mayee and Datar with some modification).

Scale	Per cent disease incidence	Categories
0	No symptom of disease	Immune/highly resistant
1	1 - 10% plants affected	Resistant
3	11 - 20% plants affected	Moderately resistant
5	21 - 50% plants affected	Moderately susceptible
7	51 - 70% plants affected	Susceptible
9	71% and above	Highly susceptible

For each fungicide, a stock solution having the concentration of 10000 ppm was prepared. The calculated amount of stock solution of fungicide as supplemented with sterilized PDA medium to get the concentrations of 100, 200, 400 and 500 ppm, respectively. In control set, required amount of water was used instead of a fungicide. Then 15 ml of medium was poured in each Petri plate and allowed them to solidify. Then test pathogen was inoculated at the center of the plate with a 5 mm mycelial agar disk cut from the margin of actively growing culture of it. Three replications were maintained in each case. The inoculated plates were incubated at 25 ± 2ºC. The radial growth of the colonies was measured after 7 days of incubation. The per cent growth inhibition of each test fungus was calculated by using the formula used by Hossain and Bashar[5].

Results and Discussion

Total number of plants, diseased plants and percentage of disease incidence in 40 germplasm lines and 2 released varieties of *Corchorus olitorius* are presented in Table 2. In case of 40 germplasm lines and two released varieties, six fungal diseases (stem rot, die back, soft rot, root rot, black band and anthracnose), two viral diseases (leaf mosaic and leaf curl) and one nematode (root knot) disease were recorded.

Table 2. Number of total plants, diseased plants and percentage of disease incidence in different germplasms of *Corchorus olitorius*.

Accession No.	Total no. of plants	No. of plants affected by different diseases										
		Stem-rot	Die-back	Soft-rot	Root-rot	Black-band	Anthracnose	Leaf mosaic	Leaf curl	Root knot	No. of infected plants	% disease incidence
1041	72	1	1	-	-	-	-	4	-	2	8	11.11
1045	39	-	1	-	-	-	-	1	-	1	3	07.69
1050	59	1	1	-	-	-	-	1	1	-	4	06.77
1056	57	2	2	1	1	-	-	1	-	-	7	12.28
1060	60	3	1	-	-	-	-	-	-	1	5	08.33
1062	61	1	1	-	1	-	-	-	-	1	4	06.55
1065	57	1	1	-	-	-	-	-	-	-	2	03.50
1093	58	3	2	-	-	-	-	1	1	1	8	13.79
1095	54	6	3	1	-	-	1	2	-	1	14	25.92
1143	53	2	2	-	2	-	-	-	1	-	5	09.43
1151	58	1	1	-	-	3	-	3	2	5	15	25.86
1155	60	2	1	1	-	1	-	-	1	-	7	11.66
1156	59	5	3	1	1	-	-	5	-	-	15	25.42
1157	50	2	2	1	1	-	-	1	2	-	9	18 .00
1158	60	3	1	-	1	-	-	2	1	1	9	15.00
1261	54	1	2	-	-	-	-	2	1	1	4	07.40
1312	52	1	1	-	-	-	-	1	1	-	7	13.46
1313	54	3	1	-	2	-	-	3	1	-	9	16.66
1338	55	-	2	1	1	-	-	-	-	-	6	10.90
1346	52	2	2	1	1	-	-	1	-	1	8	15.38
1357	55	-	1	-	-	-	-	5	-	-	7	12.72
1442	54	-	3	1	1	-	-	2	-	1	8	14.81
2407	62	-	1	-	1	-	-	6	-	1	9	14.52

(contd.)

(Contd.)

Accession No.	Total no. of plants	No. of plants affected by different diseases										
		Stem rot	Die back	Soft rot	Root rot	Black band	Anthracnose	Leaf mosaic	Leaf curl	Root knot	No. of infected plants	% disease incidence
3491	43	3	2	-	-	-	-	2	-	1	8	18.60
3708	56	-	2	-	1	-	-	2	-	-	7	12.50
3711	64	3	1	-	-	-	-	3	1	1	7	10.94
3716	38	2	1	-	1	-	1	-	2	2	9	23.68
3724	32	-	-	-	1	-	-	-	-	-	1	03.13
3755	78	2	2	-	2	1	-	6	2	3	18	23.08
3756	47	1	1	-	-	-	-	1	-	2	6	12.77
3801	64	2	1	-	1	-	-	6	-	2	12	18.75
3802	15	-	-	-	1	-	-	1	-	2	4	26.66
3809	60	-	1	1	1	-	-	1	1	3	7	11.67
3812	53	3	1	-	-	-	-	1	-	2	7	13.21
4159	57	2	2	-	-	1	1	2	-	1	9	15.79
4178	72	-	1	-	-	-	-	4	-	-	5	06.94
4229	75	-	-	-	-	-	-	-	-	-	0	0.00
4305	9	-	1	-	-	-	-	2	1	-	4	44.40
4852	45	1	-	1	2	-	-	-	-	1	5	11.11
5009	63	-	1	-	-	-	-	4	-	-	5	07.93
O-9897	94	6	5	-	1	-	1	-	-	1	14	14.89
O-72	97	1	1	-	-	2	-	5	-	-	9	09.27
Total		66	59	10	25	12	4	51	20	41		

- = Absence of affected plant.

Maximum number of jute plants was infected by stem rot (66) disease as followed by die back (59), leaf mosaic (51), root knot (41), root rot (25), leaf curl (20), black band (12), soft rot (10) and anthracnose (4) (Table 2).

In case of individual disease evaluation highest number of stem rot was observed in Acc. number 1095 and variety O-9897. There was no stem rot in Acc. numbers 1045. 1338, 1357, 1442, 2407, 3708, 3724, 3802, 3809, 4178, 4229, 4305 and 5009. The highest number of die back was observed in variety O-9897 and it was absent in Acc. numbers 3724, 3802, 4229, 4305 and 4852. The highest number of root rot was observed in Acc. numbers 1143 ,1313, 3755, 4852, and was absent in Acc. numbers 1041, 1050, 1060, 1065, 1093, 1095, 1151, 1155, 1261, 1312, 1357, 3491, 3711, 3756, 3812, 4159, 4178, 4229, 5009 and variety O-72 (Table 2). Less number of soft rot was observed in Acc. numbers 1056 1095, 1155, 1156, 1157, 1338, 1346, 1442, 3809 and 4852. Accession numbers 1151, 1155, 3755, 4159 and variety O-72 showed less number of black band disease. Anthracnose was rarely observed in Acc. numbers 1095, 3716, 4159 and variety O-9897. Leaf mosaic was observed in young stage in highest number in Acc. number 3755 and absent in Acc. numbers 1060, 1062, 1065, 1143, 1155, 1338, 3716, 3724, 3809, 4229, 4852 and variety O-9897. Accession numbers 1050, 1093, 1143, 1151, 1155, 1157, 1158, 1312, 1313, 37113716, 3755, 3756, 3809 and 4305 showed less number of leaf curl disease. Highest number of root knot was observed in Acc. number 1151 and it was absent in Acc. numbers 1050 , 1056, 1065, 1143, 1155, 1156, 1157, 1261, 1313, 1338, 3708, 3724, 4178, 4229, 4305, 5009 and variety O-9897 (Table 2).

According to the scale of Mayee and Datar(7) with some modification only Accession number 4229 behaved as highly resistant or immune. Twelve germplasm lines *viz.*, Accession numbers 1045, 1050, 1060, 1062, 1065, 1143, 1261, 1338, 3711, 3724, 4178, 5009 and released variety O-72 were found resistant. Twenty germplasm lines *viz.*, Accession numbers 1041, 1056, 1093, 1155, 1157, 1158, 1312, 1313, 1346, 1357, 1442, 2407, 3491, 3708, 3756, 3801, 3809, 3812, 4159, 4852 and released variety O-9897 were moderately resistant. Seven germplasm lines of Acc. numbers 1095, 1151, 1156, 3716, 3755, 3802 and 4305 responded as moderately susceptible. None of the varieties and germplasm line behaved as susceptible and highly susceptible according to Mayee and Datar(7).

A total of six pathogens *viz.*, *Botryodiplodia theobromae, Colletotrichum* sp., *Fusarium* sp.1, *Fusarium* sp. 2, *Rhizoctonia solani* and *Sclerotium rolfsii* were selected as test pathogens owing to their previous report as jute pathogens(15-17) (Fig. 1).

Results of six fungicides on the radial growth of test pathogens at 500 ppm are presented in Table 3. Out of six fungicides Bavistin, Dithane and Tilt showed the complete growth inhibition of the test pathogens except *Sclerotium rolfsii* in case of Bavistin. These three fungicides were again tested in 100, 200 and 400 ppm concentrations to find out their minimal requirement of concentration.

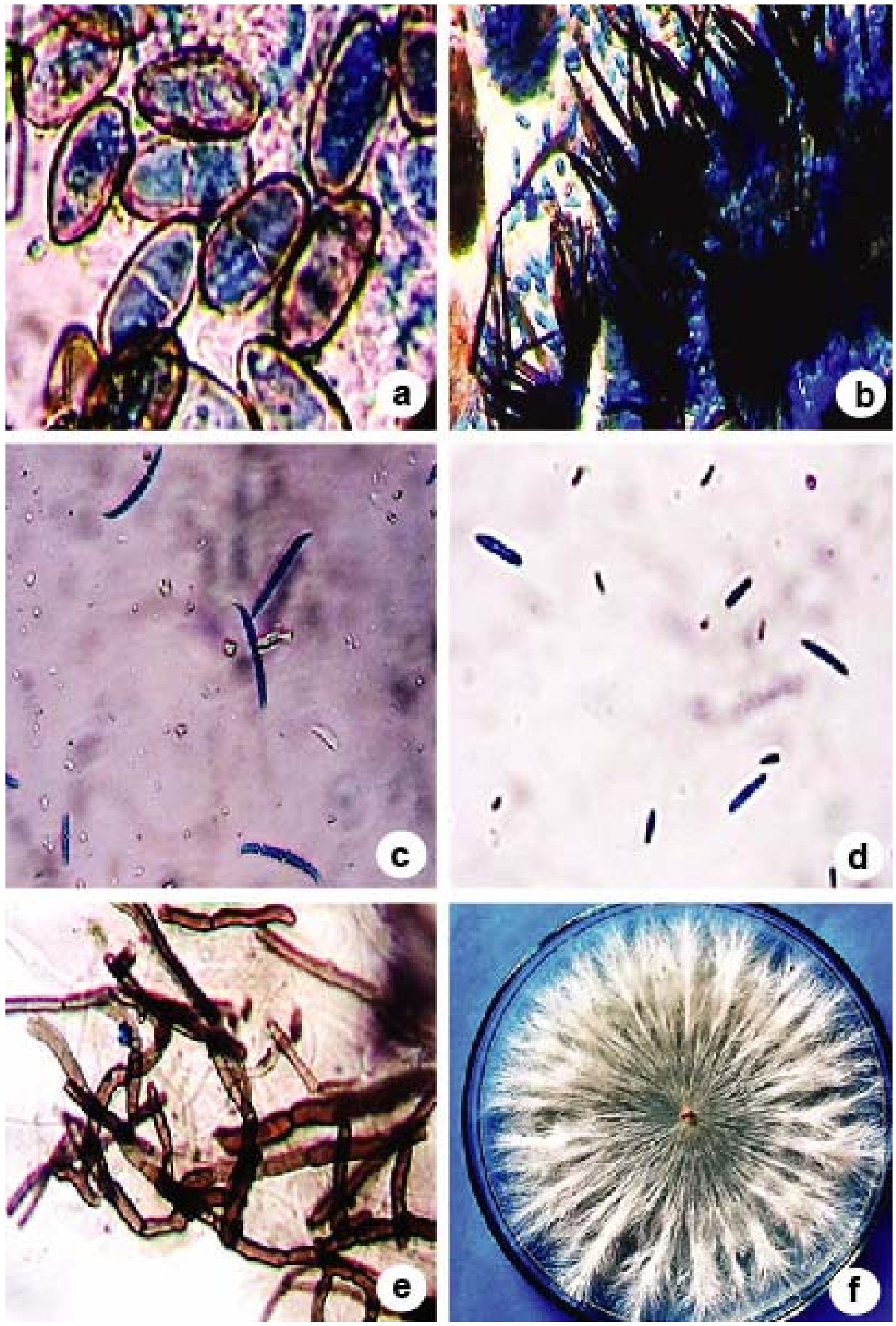

Fig. 1. Different pathogens of *Corchorus olitorius*. a. Conidia of *Botryodiplodia theobromae*, b. Conidia and setae of *Colletotrichum*sp., c-d. Macro and micro conidia of *Fusarium* sp. 1 and *Fusarium* sp. 2, e. Mycelium of *Rhizoctonia solani* and f. Mycelium of *Sclerotium rolfsii.*

Effects of Bavistin, Dithane and Tilt on the radial growth of the test pathogens at 100, 200 and 400 ppm are presented in Table 4. Bavistin showed complete growth inhibition of all the test pathogens at 400 ppm except *S. rolfsii*. Bavistin also showed complete growth inhibition of all the test pathogen sat 100 and 200 ppm except *Colletotrichum* sp. and *S. rolfsii.* Bashar[18] reported that bavistin checked the complete growth of *F. oxysporum* f. sp. *ciceri,* causal agent of chickpea wilt at 100 ppm. Chakraborty *et al.*[19] reported that, at 0.5% dose, bavistin happened to be the most efficient one contributing the highest inhibition (83.7%) of growth of *Fusarium solani* causing wilt of brinjal under *in vitro* condition.

Table 3. Fungitoxicity of fungicides against the test pathogens at 500 ppm concentration.

Fungicides	% inhibition of radial growth of test pathogens					
	Botryodiplodia theobromae	*Colletotrichum* sp.	*Fusarium* sp.1	*Fusarium* sp.2	*Rhizoctonia solani*	*Sclerotium rolfsii*
Bavistin DF	100	100	100	100	100	100
Capvit 50 WP	100	100	100	100	100	100
Dithane M 45	100	100	100	100	100	5.36
Hayvit 80 WP	100	66.66	56.09	45.83	100	100
Ridomil Gold MZ 68 WG	62.22	25.00	46.67	37.06	100	4.34
Tilt 250 EC	73.88	56.41	46.34	25.00	26.11	33.33

The complete growth inhibition of *R. solani, S. rolfsii* and *Fusarium* sp.1 was observed at 400 ppm except *B. theobromae* (88.90%), *Colletotrichum* sp. (83.63%) and *Fusarium* sp.2 (64.06%). The highest growth inhibition of *Fusarium* sp.1 (100%) was noticed at 100 ppm which was followed by *B. theobromae* (78.90%), *S. rolfsii* (77.78%), *Colletotrichum* sp. (70.90%), *R. solani* (66.67%) and *Fusarium* sp. 2 (50%). The highest growth inhibition of *Fusarium* sp.1 (100%) was found at 200 ppm concentration which was followed by *S. rolfsii* (91.11%), *B. theobromae* (82.22%), *R. solani* (80%), *Colletotrichum* sp. (78.18%), and *Fusarium* sp. 2 (56.25%) (Table 4). Javaid *et al.*[20] reported that out of four fungicides such as Acrobat MZ, Dithane M-45, Aliette and Ridomil Gold, Dithane was found to be the most effective against *Colletotrichum gloeosporioides*. There were 60 - 66% reduction in fungal biomass owing to application of recommended and various lower doses of this fungicide. Dithane was also effective in controlling yam (*Discorea rotundata*) rot fungi, namely *Aspergillus, Botryodiplodia, Fusarium, Penicillium* and *Rhizopus*[21].

The maximum growth inhibition at 100 ppm was observed in *R. solani* (90%) followed by *Fusarium* sp. 1 (86.27%), *S. rolfsii* (85.56%), *Colletotrichum* sp. (82.14%), *B. theobromae* (69.44%) and *Fusarium* sp. 2 (61.29%). The complete growth inhibition of *Colletotrichum* sp., *Fusarium* sp.1 and *R. solani* was observed at 200 and 400 ppm (Table 4). The same fungicides also showed different effects on different pathogens in the present investigation due to the selection of different test pathogens. In contrast to the present study, Singh *et al*.(22) reported that Tilt 25 EC was found most effective to inhibit *Colletotrichum gloeosporioide* 100 ppm followed by hexaconazole and carbendazim. Handiso and Alemu(23) reported that Tilt 250 EC showed 99.33, 99.33 and 68.33% growth inhibition of *Colletotrichum capsici* at 150, 250 and 300 ppm, respectively.

Table 4. Per cent inhibition of radial growth of test pathogens with different fungicides at various concentrations.

Fungicides	Conc. (ppm)	% inhibition of radial growth of test pathogens					
		Botryodiplodia theobromae	*Colletotri-chum* sp.	*Fusarium* sp. 1	*Fusarium* sp. 2	*Rhizoctonia solani*	*Sclerotium rolfsii*
Bavistin DF	100	100	89.09	100	100	100	0.00
	200	100	89.09	100	100	100	0.00
	400	100	100	100	100	100	0.00
Dithane M 45	100	78.9	70.90	100	50.00	66.67	77.78
	200	82.22	78.18	100	56.25	80.00	91.11
	400	88.90	83.63	100	64.06	100	100
Tilt 250 EC	100	69.44	82.14	86.27	61.29	90	85.56
	200	88.89	100	100	70.96	100	91.11
	400	94.44	100	100	74.20	100	100

In the present study, 12 germplasm lines of Acc. numbers 1045, 1050, 1060, 1062, 1065, 1143, 1261, 1338, 3711, 3724, 4178, 5009 and one released variety O-72 found to be resistant to jute diseases which can be utilized for developing disease resistant variety. Efficiency gradients observed in the present investigation expressed that Bavistin DF, Dithane M-45, and Tilt 250 EC were the best inhibiting agent against the *in vitro* growth of the test pathogens associated with *Corchorus olitorius*.

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