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Morphological and Pathogenic Variability among *Rhizoctonia bataticola* Isolates Associated with Soybean (*Glycine max* L.) from India

R.M. Gade*, Y.K. Belkar and Y.V. Ingle

Department of Plant Pathology, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola (M.S) 444 104, India

**Corresponding author*

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A B S T R A C T

Rhizoctonia bataticola is a serious pathogen of many crops. In the present studies, 40 isolates of *Rhizoctonia bataticola* from different agro climatic regions of India were analyzed for morphological and pathogenic variability. Regardless of their geographic origins, significant differences were detected among 40 isolates in their radial growth, sclerotial size, and shape as well as in pathogenicity. Thirty two isolates were rated as fast growing and the rest of the isolates as medium growing. Sixteen isolates were classified as large sized, 8 as small sized, and the remaining 16 isolates as medium sized on the basis of their sclerotial size. On the basis of disease expression, isolate Rb-29 (Kurundwad, Sangli) showed broad virulence range as it infected all the genotypes followed by isolate Rb-38 (Udaipur, Rajasthan) which infected eleven genotypes. No relationship was found among the morphological characters and pathogenicity of the isolates. These morphological and pathogenic variations in various isolates of *R. bataticola* may be considered important in disease management systems and will be useful in breeding programmes of soybean cultivars resistant to root rot.

Introduction

Soybean [Glycine max (L.) Merill] is the major oilseed crop in the world and is cultivated on an area of 108.83 Lakh ha with a total production of 114.90 Lakh MT. The crop is grown in a wide range of agro climatic zones. The low yield of soybean in India can be attributed to legions of biotic and abiotic constraints. Among biotic factors, diseases are the most dominant. Soybean crop can be attacked by more than 100 pathogens (Sinclair and Shurlleff, 1975). In India, losses due to various diseases are estimated as 12 per cent of total production. The diseases include rust, wilts, leaf spot, rots and powdery mildew, bacterial and viral diseases. Among these, root rot caused by *Rhizoctonia bataticola* is of prime importance in reducing crop yield.

Yield losses 30-50 per cent due to *Macrophomina phaseolina* in soybean crop has been reported (Yang and Navi, 2005). *Rhizoctonia bataticola* reduces the yield of soybean by 2-21 per cent (Wrather and Koenning, 2006). The pathogen is distributed in diverse climatic conditions from arid to tropical regions and has a broad host range.

There are more than 500 hosts of the fungus including legume and cereal plants (Dhingra and Chagas, 1981; Sinclair, 1982). *M. phaseolina* is a soil- and seed-borne pathogenic fungus and produces cushion shaped black sclerotia (Wheeler, 1975). Its prevalence can be enhanced by different physiological and ecological factors such as low moisture contents, high temperature, and heat (Papavizas, 1977; Dhingra and Sinclair, 1978). Disease severity is correlated with viable sclerotia present in the soil. Charcoal rot infects plants at almost all growth stages. Dark lesions appear on the epicotyls and hypocotyls followed by seedling death due to obstruction of xylem vessels. In plants, the pathogen causes red to brown lesions on roots and stems with production of dark mycelia and black microsclerotia. Ultimately the plant becomes defoliated and wilted (Abawi and Pastor-Corrales, 1990) and perishes.

Among the main management strategies, use of cultivars resistant to *M. phaseolina* has gained wide popularity and acceptance amongst farmers as application of fungicides is often intertwined with potential hazards to humans and the environment. Furthermore, resistant cultivars outstrip fungicides in various respects and emphasis is being laid on the development of new resistant germplasm. However, it has been observed that control measures against pathogens become complicated and even ineffective due to the variability among populations of the same pathogen in different agro ecological zones.

There are reports in other parts of the world that populations of *M. phaseolina* showed significant variations morphologically, physiologically, pathogenically and genetically. These variations aid the pathogen to adapt and survive in diverse environments. A thorough knowledge of pathogenic variability of *R. bataticola* is essential to design disease management strategies for different agro climatic zones of the country by breeding resistant cultivars. Hence, we investigated morphological and pathogenic variability among 40 isolates of *R. bataticola* infecting soybean, collected from five different major soybean growing states of India. It has also been determined whether morphological variations among *M. phaseolina* isolates have any relationship with the pathogenic variability.

Materials and Methods

Collection of fungal isolates

A total of 40 isolates of *Rhizoctonia bataticola* were collected from 5 major soybean growing states *viz*. Madhya Pradesh, Maharashtra, Karnataka, Rajasthan and Andhra Pradesh of the India representing different agro climatic zones of country delineated mainly on the basis of physiographic and climatic characteristics, soil type, and agricultural land use (Table 1). Samples of stems bearing microsclerotia of the fungus and characteristic symptoms of root rot were collected from the infected soybean plants from farmers' fields and designated. The diseased samples were first packed in paper bags and then in 15 x 20 cm polyethylene bags, labeled, brought to the lab, and stored at 4°C until processed for identification.

Isolation, purification, and identification of *R. bataticola*

The fungus was isolated from stem bark tissues of soybean bearing fungal sclerotia and showing characteristic root rot symptoms. The samples were cut into small pieces (5-10 mm long) and surface sterilized with 1:1000 mercuric chloride for 30 seconds and then rinsed thrice in sterilized distilled water. The pieces were placed on potato dextrose agar medium in Petri dishes and incubated in dark at 27 ± 2°C for 7days. A small portion of the

fastest growing colony of *R. bataticola* was taken from the periphery of a 90 mm diameter Petri dish, spread onto Petri dishes containing potato dextrose agar medium and incubated in the dark at 27 ± 2°C for 7 days. A small portion of the colony having sclerotia was taken up into a drop of sterilized water and agitated with a sterilized needle to separate the sclerotia from the mycelia. Sclerotia were then transferred to 90 mm diameter Petri dishes containing potato dextrose agar medium. Colonies appearing from single sclerotium were again transferred to potato dextrose agar medium in 90 mm Petri plates, incubated as mentioned above and identified as described.

Storage of pure cultures of *R. bataticola*

The purified culture (5 mm disc) from each isolate growing on PDA was transferred to10 ml culture tubes and incubated in the dark at 27 ± 2°C for 6 days, until the surface of PDA was covered with a dense sclerotial layer of the fungal culture. The culture tubes were labeled and stored at 4°C.

Multiplication of *R. bataticola*

Ground sorghum seeds were water-soaked overnight, air-dried under room temperature, and placed in conical flasks. The mouth of each flask was plugged with cotton wool, wrapped in aluminum foil, and autoclaved at 15 psi (121° C) for 20 minutes. After cooling, the seeds in flasks were inoculated with 4 mm mycelial plugs from a 7-day old culture of *R. bataticola* and incubated at 27 ± 2°C for 15 days. The flasks were shaken at alternate days for uniform colonization of the grains. The inoculum thus produced was used in pot assay.

Determination of morphological variability

Morphological variability among 40 isolates of *R. bataticola* was studied on the basis of the following parameters.

Radial growth

For studying variability in radial growth, the isolates were grown on Potato Dextrose Agar. Fifteen milliliters of autoclaved PDA was poured in 90 mm diameter Petri plates, allowed to solidify, and inoculated in the center with a 5 mm plug from the actively growing culture of each isolate of the fungus. The plates were incubated at 27 ± 2°C for 7 days. Each isolate was replicated three times. After the stipulated period the growth of each isolate was measured in terms of colony diameter and their means were computed. On the basis of radial growth, the isolates were categorized as fast (>80 mm), and medium (70-80 mm) growing.

Sclerotial size

For measuring sclerotial size, slides from 7-day-old pure cultures of *R. bataticola* isolates were prepared and examined under a microscope ocular micrometer. Sizes of fifty randomly selected sclerotia were measured and their means were calculated. The isolates were classified as large (>80 µm), medium (60-80 µm), and small (<60 µm) sized.

Determination of pathogenic variability

The pathogenicity of 40 isolates was studied on susceptible soybean cultivar TAMS-38 in the greenhouse with six replications for each isolate. Seeds were disinfected by immersing in 0.1 per cent mercuric chloride for 1 min, rinsed in sterilized water, and air-dried. Ten seeds of cultivar TAMS-38 of soybean were sown in pots containing 2 kg soil infested with each isolate of *R. bataticola* in 1:9 proportions (inoculum + soil). Pots without inoculum served as controls. The pots were placed in a greenhouse at 30 ± 2°C. The pots were watered as and when required and observations on occurrence of root rot were recorded. On the basis of occurrence and

symptoms, the isolates were identified as pathogenic.

Grouping of isolates

Forty isolates of *R. bataticola* were tested by sick soil method for their virulence against susceptible variety TAMS-38. The percent root rot was recorded on the basis of healthy and root rot infected plants. The isolates of R. bataticola were tentatively divided into five groups on the basis of virulence as follows (Pawar, 2010).

Category	Per cent mortality due to root rot
Non-pathogenic	0 %
Weakly pathogenic	1-20 %
Moderately pathogenic	21-50 %
Strongly pathogenic	51-70 %
Highly pathogenic	>70%

Host differential reaction (Pot culture)

Virulence analysis of *R. bataticola* isolates was carried out on set of twelve host differentials showing absolutely resistant, moderately resistant, moderately susceptible, susceptible and highly susceptible reactions *viz*., GBIC-18758, AMS-595, AMS-MB-5-18, AMS-MB-5-19, AMS-1003, AMS-475, AMS-3923, AMS-9933, Bragg, Punjab-1, TAMS-38 and TAMS-9821 in green house. Seven isolates of *R. bataticola* collected from different locations showing different pathogenic reactions were selected and inoculated separately to each host differential. Ten seeds of each cultivar were sown in triplicate in surface sterilized pots filled with 2 kg sterilized soil, inoculated with 14 days old culture @ 100 g kg $^{-1}$ of the pathogen multiplied on sand sorghum grain medium seven days before sowing. The pots without inoculum were served as control. The incidence of root rot was recorded at 15 days interval up to crop maturity. Reactions were graded according to the uniform method of disease rating as absolutely resistant (No mortality), highly resistant (0.01-11.11% mortality), moderately resistant (12.22-33.33 % mortality), moderately susceptible (34.44-55.55 % mortality), susceptible (56.77-77.77 % mortality) and highly susceptible (78.88-100.00 % mortality).

Results and Discussion

Morphological variability among Rhizoctonia bataticola isolates

Significant variations were observed in the morphological parameters among 40 isolates of *R. bataticola* collected from different major soybean producing states of India.

Radial growth

Significant differences among 40 isolates of *R. bataticola* collected from different states were observed on the basis of radial growth. The individual average radial growths of 40 isolates of *R. bataticola* ranged from 75.00 to 90.00 mm. Maximum colony diameters of 90.00 and 85.66 mm were observed in case of isolate Rb-1, Rb-6, Rb-11, Rb-12, Rb-15, Rb-22 Rb-23, Rb-29, Rb-37 and Rb-38, and proving to be the fast growing, while isolates Rb-5 and Rb-26 showed the minimum radial growths and were rated as medium growing. The individual radial growths of all the isolates are shown in Table 2. Thirty two isolates showed radial growths above 80 mm and were rated as fast growing while rest of the isolates showed growth between 70 and 80 mm and hence were classified as medium growing (Table 3).

Sclerotial size

Significant variations were also observed among these isolates regarding the size of their sclerotia. Maximum sclerotial size was

observed in case of isolate Rb-33 (120.11 µm) while the isolate Rb-40 produced the smallest size sclerotia (42.03 µm). The individual average sclerotial sizes of isolates ranged from 42.03-120.11 µm which are given in Table 2.

The size of sclerotia of 16 isolates was above 80 µm and was classified as large sized while 16 isolates with sclerotial size less than 60 µm were rated as small sized. The remaining 8 isolates ranged between 60 and 80 µm sclerotial size and were categorized as medium sized (Table 4).

Pathogenic variability among *R. bataticola* isolates

Pathogenic ability of isolates indicated that out of 40 isolates of *R. bataticola*, all isolates proved pathogenic with susceptible cultivar TAMS-38 (Table 5). Among different isolates tested, two *viz*., Rb-3 and Rb-17 were highly pathogenic with 71-100 per cent root rot incidence whereas eight *viz.*, Rb-10, Rb-13, Rb-18, Rb-22, Rb-29, Rb-34, Rb-39 and Rb-40 were strongly pathogenic with average of 51-70 per cent root rot. Isolates, Rb-4, Rb-5, Rb-8, Rb-9, Rb-11, Rb-12, Rb-14, Rb-15, Rb-16, Rb-20, Rb-21, Rb-23, Rb-24, Rb-25, Rb-26, Rb-27, Rb-28, Rb-30, Rb-31, Rb-33, Rb-35, Rb-36, Rb-37 and Rb-38 were moderately pathogenic showing 21 to 50 per cent root rot and remaining six viz.,Rb-1, Rb-2, Rb-6, Rb-7, Rb-19 and Rb-32 were weakly pathogenic with average root rot incidence of 1 to 20 per cent. However, none of the isolate proved to be non-pathogenic to cultivar TAMS-38.

Grouping of isolates

Forty isolates of R. bataticola were tested by sick soil method for their virulence against susceptible cultivar TAMS-38. The per cent root rot was recorded on the basis of healthy and root rot infected plants. The data on pathogenic variability in different isolates of R. bataticola are given in Table 5. The isolates were tentatively divided in five groups based on their pathogen ability (Table 6).

Host differential reaction (Pot culture)

The pathogenic reactions of seven isolates of *R. bataticola* against a set of twelve genotypes of soybean *viz.*, GBIC-18758, AMS-595, AMS-MB-5-18, AMS-MB-5-19, AMS-1003, AMS-475, AMS-3923, AMS-9933, Bragg, Punjab-1, TAMS-38 and TAMS-98-21 were assessed under sick soil conditions.

Significant differences among isolates, varieties, and their interactions in which the disease response on various cultivars varied between 3-9 grades shown in Table 7.

Genotype GBIC-18758 was found highly resistant against isolate Rb-17 (9.52 %) and moderately susceptible against Rb-3 (46.15 %) and Rb-10 (35.29 %).

Genotype AMS-595 was moderately susceptible to Rb-17 (46.15 %) and highly susceptible to isolates Rb-29 (100 %), Rb-3 (83.33 %) and Rb-10 (80 %) while isolates Rb-39 (75.00 %), Rb-38 (76.47 %) and Rb-22 (66.67 %) exhibited susceptible reaction.

AMS-MB-5-18 showed moderate resistance against Rb-17 (21.74 %) and Rb-22 (31.25 %) whereas moderately susceptible reaction against Rb-38 (53.85 %) and Rb-29 (50 %). Isolates, Rb-38 (50 %), Rb-10 (50 %), Rb-29 (46.15 %), Rb-3 (47.37 %) and Rb-17 (45.00 %) exhibited moderately susceptible reaction towards genotype AMS-MB-5-19 while moderately resistant reaction against Rb-39 (16.67 %).

Isolates Rb-38, (18.18 %) and Rb-10 (21.43 %) were least virulent as moderately resistant reaction was developed against genotype AMS-1003.

Table.1 Isolates of *Rhizoctonia bataticola* collected from soybean plants from different agro climatic regions

Sr No	State	Agro Climatic Regions	Soil Type	Rainfall (mm)	Districts	Isolates
1	Madhya Pradesh	Malwa Plateau	Medium Black (Medium)	800-1200	Indore	Rb-1, Rb-2, Rb-3
					Dewas	Rb-4, Rb-5,Rb-6, Rb-7, Rb-8
					Ujjain	Rb-9, Rb-10, Rb-11
		Nimar Plains	Medium Black (Medium)	800-1000	Harda	Rb-12, Rb-13, Rb-14
		Vindhya Plateau	Medium Black and Deep Black	1200-1400	Sehore	Rb-15,Rb-16, Rb-17
		Kymore Plateau and Satpura Hills	Mixed Red and Black soils (Medium)	1000-1400	Jabalpur	Rb-18, Rb-19, Rb-20
2	Maharashtra	Central Maharashtra Plateau	Black to Red	700-900	Akola	Rb-21
					Amaravati	Rb-22
					Buldhana	Rb-24
		Central Vidharbha Region	Black soils, medium to heavy texture	1130	Wardha	Rb-23
					Nagpur	Rb-25
		Western Maharashtra Plain Zone	Greyish Black	700-1200	Satara	Rb-26 and Rb-27
					Sangli	Rb-28 and Rb-29
					Kolhapur	Rb-30 and Rb-31
3	Karnataka	Northern Dry Zone	Black Clay Medium	464-785	Belgaun	Rb-32, Rb-33, Rb-34
					Dharwad	Rb-35 and Rb-36
4	Rajasthan	Sub Humid Southern Plains	Alluvial	500-900	Bhilwara	Rb-38
					Udaipur	Rb-39
		Humid South Eastern Plains	Black of Alluvial origin	650-1000	Kota	Rb-37
5	Telangana	North Telangana Zone	Red Sandy soils	900-1500	Adilabad	Rb-40

Table.2 Morphological variations among different isolates of *Rhizoctonia bataticola*

Sr. No	Isolates	Radial growth (mm)*	Mean sclerotial size (µm)**	Shape of sclerotia
1	Rb-1	90	47.26±5.44	Round
2	Rb-2	81.33	51.56±3.68	Oblong
3	Rb-3	82.33	108.16±6.42	Round
4	Rb-4	80	86.49±6.88	Round
5	Rb-5	75	93.03±15.37	Round
6	Rb-6	85.66	49.69±4.83	Round
7	Rb-7	78	50.06±4.95	Oblong
8	Rb-8	84.33	53.42±4.48	Round
9	Rb-9	83.66	63.14±7.64	Round
10	Rb-10	82	65.19±8.14	Round
11	Rb-11	90	46.14±6.91	Round
12	Rb-12	85.66	57.72±9.13	Round
13	Rb-13	76.33	58.66±9.34	Round
14	Rb-14	80	61.08±10.94	Round
15	Rb-15	90	68.74±5.64	Oblong
16	Rb-16	82.66	60.52±3.92	Round
17	Rb-17	80	61.64±10.78	Oblong
18	Rb-18	86	118.24±7.34	Round
19	Rb-19	83.33	46.33±6.59	Round
20	Rb-20	82	103.11±9.61	Round
21	Rb-21	76.33	44.27±5.57	Oblong
22	Rb-22	85.66	112.27±9.48	Round
23	Rb-23	90	61.27±9.07	Round
24	Rb-24	84.33	63.89±6.42	Round
25	Rb-25	78	56.23±6.80	Round
26	Rb-26	75	61.46±5.33	Round
27	Rb-27	82.33	117.87±13.08	Oblong
28	Rb-28	81.33	100.69±9.82	Round
29	Rb-29	90	93.03±15.37	Round
30	Rb-30	82	84.99±6.02	Round
31	Rb-31	79.33	86.49±6.88	Round
32	Rb-32	84.33	47.82±5.91	Round
33	Rb-33	82	120.11±7.68	Round
34	Rb-34	78.33	65.57±6.24	Round
35	Rb-35	85	54.92±6.21	Round
36	Rb-36	80	50.44±6.46	Round
37	Rb-37	85.66	116.19±13.36	Oblong
38	Rb-38	90	55.48±5.77	Round
39	Rb-39	82	80.88±4.61	Round
40	Rb-40	81.33	42.03±4.86	Oblong

* Average of three replications ** Mean of 50 observations for each isolate

Table.3 Categorization of *R. bataticola* isolates on the basis of radial growth

Sr. No	Category	Number	Isolates
1	Fast growing (>80 mm)	32	Rb-1,Rb-2,Rb-3,Rb-4,Rb-6,Rb-8,Rb-9,Rb-10,Rb-11,Rb-12,Rb-14,Rb-15,Rb-16,Rb-17,Rb-18,Rb-19, Rb-20, Rb-22, Rb-23, Rb-24, Rb-27, Rb-28, Rb-29, Rb-30, Rb-32, Rb-33, Rb-35, Rb-36, Rb-37,Rb-38, Rb-39, Rb-40
2	Medium growing (70-80 mm)	8	Rb-5, Rb-7, Rb-13, Rb-21, Rb-25, Rb-26, Rb-31, Rb-34

Table.4 Categorization of *R. bataticola* isolates on the basis of size of sclerotia

Sr. No	Category	Number	Isolates
1	Large sized (>80 µm)	16	Rb-3,Rb-4, Rb-5, Rb-18, Rb-20, Rb-22, Rb-23, Rb-24, Rb-27, Rb-28, Rb-29, Rb-30, Rb-31, Rb-33,Rb-37, Rb-39
2	Medium sized (60-80 µm)	8	Rb-9, Rb-10, Rb-14, Rb-15, Rb-16, Rb-17, Rb-26, Rb-34
3	Small sized (<60 µm)	16	Rb-1, Rb-2, Rb-6, Rb-7, Rb-8, Rb-11, Rb-12, Rb-13, Rb-19, Rb-21, Rb-25, Rb-32, Rb-35, Rb-36, Rb-38 and Rb-40

Table.5 Pathogenic variability among *Rhizoctonia bataticola* isolates against soybean cultivaTAMS-38

Sr. No	Isolates	Total no. of seeds sown	Total no. of plants germinated	No. of plants infected	Days to initiate symptoms	Per cent mortality due to pathogen (30 DAS)
1.	Rb-1	60	47	9	24	19.15 (26.02)*
2.	Rb-2	60	51	10	24	19.61 (26.31)
3.	Rb-3	60	48	39	14	81.25 (64.55)
4.	Rb-4	60	46	12	22	26.09 (30.90)
5.	Rb-5	60	43	12	22	27.91 (32.00)
6.	Rb-6	60	45	7	28	15.56 (23.12)
7.	Rb-7	60	49	9	25	18.37 (25.55)
8.	Rb-8	60	43	20	18	46.51 (43.17)
9.	Rb-9	60	48	15	20	31.25 (34.01)
10.	Rb-10	60	53	30	17	56.60 (48.77)
11.	Rb-11	60	44	17	20	38.64 (38.44)
12.	Rb-12	60	46	13	22	28.26 (32.10)
13.	Rb-13	60	42	23	17	54.76 (47.86)
14.	Rb-14	60	46	14	22	30.43 (33.56)
15.	Rb-15	60	46	18	20	39.13 (38.86)
16.	Rb-16	60	47	21	18	44.68 (42.11)
17.	Rb-17	60	45	36	14	80.00 (63.74)
18.	Rb-18	60	44	23	17	52.27 (46.57)
19.	Rb-19	60	46	9	24	19.57 (26.57)
20.	Rb-20	60	46	13	22	28.26 (32.57)
21.	Rb-21	60	50	22	18	44.00 (41.39)
22.	Rb-22	60	53	32	16	60.38 (51.05)

23.	Rb-23	60	48	22	18	45.83 (42.60)
24.	Rb-24	60	48	18	20	37.50 (38.17)
25.	Rb-25	60	47	23	18	48.94 (44.37)
26.	Rb-26	60	54	26	18	48.15 (44.05)
27.	Rb-27	60	48	23	18	47.92 (43.80)
28.	Rb-28	60	44	19	18	43.18 (41.21)
29.	Rb-29	60	45	23	17	51.11 (46.04)
30.	Rb-30	60	46	18	20	39.13 (38.62)
31.	Rb-31	60	51	19	20	37.25 (37.40)
32.	Rb-32	60	49	9	26	18.37 (25.50)
33.	Rb-33	60	49	21	18	42.86 (41.03)
34.	Rb-34	60	48	28	17	58.33 (49.71)
35.	Rb-35	60	51	26	17	50.98 (45.63)
36.	Rb-36	60	49	12	22	24.49 (29.74)
37.	Rb-37	60	44	17	20	38.64 (38.45)
38.	Rb-38	60	47	18	20	38.30 (38.37)
39.	Rb-39	60	51	30	17	58.82 (50.03)
40.	Rb-40	60	49	34	16	69.39 (56.24)
41.	Control	60	56	0	0	0 (0)
F Test						Sig
S.E (M)±						1.71
C.D (p=0.05)						4.81

*Figures in parenthesis are arc sin value transformed

Table.6 Grouping of *Rhizoctonia bataticola* isolates based on their pathogenic ability

Sr. No	Category	Per cent (%) mortality due to Root Rot	No. of Isolates	Isolates
1.	Non-pathogenic (NPI)	0	Nil	Nil
2	Weakly pathogenic (WPI)	1-20	6	Rb-1, Rb-2, Rb-6, Rb-7, Rb-19, Rb-32
3	Moderately pathogenic (MPI)	21-50	24	Rb-4, Rb-5, Rb-8, Rb-9, Rb-11, Rb-12, Rb-14, Rb-15, Rb-16, Rb-20, Rb-21, Rb-23, Rb-24, Rb-25, Rb-26, Rb-27, Rb-28, Rb-30, Rb-31, Rb-33, Rb-35, Rb-36, Rb-37, Rb-38
4	Strongly pathogenic (SPI)	51-70	8	Rb-10, Rb-13, Rb-18, Rb-22, Rb-29, Rb-34, Rb-39, Rb-40
5	Highly pathogenic (HPI)	>70	2	Rb-3, Rb-17

Table.7 Reaction of host differentials to *Rhizoctonia bataticola* under sick soil condition

Cultivar	*Rhizoctonia bataticola* isolates						
	Rb-39	**Rb-38**	**Rb-29**	**Rb-22**	**Rb-10**	**Rb-17**	**Rb-3**
GBIC-18758	85.71*	82.35	64.29	70.00	35.29	9.52	46.15
	HS	HS	S	S	MS	HR	MS
AMS-595	75.00	76.47	100.00	66.67	80.00	46.15	83.33
	S	S	HS	S	HS	MS	HS
AMS-MB-5-18	100.00	53.85	50.00	31.25	100.00	21.74	75.00
	HS	MS	MS	MR	HS	MR	S
AMS-MB-5-19	16.67	50.00	46.15	22.22	50.00	45.00	47.37
	MR	MS	MS	MR	MS	MS	MS
AMS-1003	81.82	18.18	88.89	75.00	21.43	42.86	66.67
	HS	MR	HS	S	MR	MS	MS
AMS-475	72.22	57.14	100.00	75.00	66.67	66.67	100.00
	S	S	HS	S	S	S	HS
AMS-3923	60.71	57.14	85.71	21.43	57.14	43.75	64.00
	S	S	HS	MR	S	MS	S
AMS-9933	22.73	60.00	90.91	71.43	75.00	65.00	42.11
	MR	S	HS	S	S	S	MS
Bragg	61.54	61.11	78.57	42.86	69.23	64.71	50.00
	S	S	HS	MS	S	S	MS
Punjab-1	68.75	46.67	80.00	40.00	41.67	42.86	63.64
	S	MS	HS	MS	MS	MS	S
TAMS-38	71.43	53.85	63.64	81.82	50.00	75.00	78.57
	S	MS	S	HS	MS	S	HS
TAMS-98-21	71.43	66.67	100.00	83.33	81.82	76.47	76.47
	S	S	HS	HS	HS	S	S

* Per cent disease incidence

MR- Moderately Resistant (12.22-33.33 % mortality), MS- Moderately Susceptible (34.44-55.55 % mortality), S- Susceptible (56.66-77.77 % mortality), HS- Highly Susceptible (78.88-100.00 % mortality)

AMS-475 genotype was highly susceptible to isolates Rb-29 (100.00 %) and Rb-3 (100.00 %) indicating highest virulence followed by isolates Rb-39 (72.22 %), Rb-22 (75.00 %), Rb-38 (57.14 %), Rb-10 (66.67 %) and Rb-17 (66.67 %). AMS-3923 showed moderate resistance against Rb-22 (21.43 %) while moderately susceptible reaction against Rb-17 (43.75 %). Isolates Rb-38 (60.00 %), Rb-22 (71.43 %), Rb-10 (75.00 %) and Rb-17 (65.00 %) exhibited more virulence against AMS-9933. At the same time, same genotype was moderately resistant to isolate Rb-39 (22.73 %). Soybean Bragg showed susceptible to highly susceptible reaction against all isolates. Three isolates *viz*., Rb-38 (46.67 %), Rb-22 (40.00 %), Rb-10 (41.67 %) and Rb-17 (42.86 %) exhibited moderately susceptible reaction against Panjab-1. Soybean TAMS-38 was highly susceptible to isolate Rb-22 (81.82 %) and Rb-3 (78.57 %) indicating highest virulence followed by isolate Rb-39 (71.43 %), Rb-29 (63.64 %) and Rb-17 (75.00 %) which showed susceptible reaction. TAMS-98-21 showed highest susceptible reaction against Rb-29 (100.00 %), Rb-22 (83.33 %) and Rb-10 (81.82 %) followed by Rb-39 (71.43 %), Rb-38 (66.67 %), Rb-17 (76.47 %) and Rb-3 (76.47 %) which showed susceptible reaction.

Rb-29 isolate showed broad virulence range as it infected all the genotypes followed by isolate Rb-38 which infected eleven genotypes. AMS-MB-5-18 and AMS-MB-5-19 were moderately resistance to three isolates Rb-22, Rb-17 and Rb-39 which were not able to cause severe infection. In present study, isolate from Kurundwad, Sangli (Rb-29) has exhibited constant severe pathogenic reaction in most of the tested entries. So this isolate will be useful for evaluation of resistance in soybean germplasm and breeding lines and in resistance breeding programmed against root rot of soybean. Seven isolates of *R. bataticola* isolated from diversed locations exhibit the different pathogenic reaction against tested. entries. It indicated considerable variability in tested isolates. This showed physiological specialization within *R. bataticola*.

In the present studies, 40 isolates of *R. bataticola* belongs to different soybean growing states of India showed variations in different morphological traits *viz.,* radial growth, sclerotial size as well as in pathogenicity. The variations in morphology might be due to differences in temperature, moisture, soil types, and other edaphic factors of various places (Iqbal and Mukhtar, 2014). Morphological variability has also been reported by many workers in terms of growth, color, pycnidium production, and chlorate sensitivity among different isolates of *M. phaseolina* on different hosts (Dhingra and Sinclair, 1973; Pearson *et al.,* 1986; Atiq *et al.,* 2001 and Riaz *et al.,* 2007) which corroborated our findings. However, in the present studies, no relationship was found among the morphological characters and pathogenicity of the isolates.

Thirty two isolates showed radial growths above 80 mm and were rated as fast growing while rest of the isolates showed growth between 70 and 80 mm and hence were classified as medium growing. The size of sclerotia of 16 isolates was above 80 µm and was classified as large sized while 16 isolates with sclerotial size less than 60 µm were rated as small sized. The remaining 8 isolates ranged between 60 and 80 µm sclerotial size and were categorized as medium sized. Earlier workers while working on *R. bataticola* associated with different crop plants also grouped the isolates into different categories based on colony characters on medium (Raut and Ingle, 1989; Meena Shekhar *et al.,* 2006; Aghakhani and Dubey, 2009; Sharma *et al.,* 2012 and Datta *et al.,* 2013).

The pathogenic fungus, *M. phaseolina*, has a broad host range and exits in two asexual forms which maintain its survival better (Dhingra and Sinclair, 1978; Cloud and Rupe, 1988). Some workers also related variability to the phenomena of host specialization in *M. phaseolina.* Su *et al.*, (2001) found host specialization in maize on the basis of pathogenic, genetic, and physiological differences. Similarly, Cloud and Rupe (1988) analyzed host specialization in soybean.

This mechanism takes long time to establish within a specific host. Mihail and Taylor (1995) suggested that, due to heterogenic nature of *M. phaseolina*, categorization into distinct subgroups based upon pathogenicity and morphology could not take place. Pathogenesis along with genetic diversity plays a specific role in host-plant resistance. Isolates having morphological similarity are not necessarily identical genetically, they might have some differences. The variable genetic pattern contributes to variation in morphology and pathogenesis, which has been confirmed by using different molecular tools (Mayek-P´erez *et al.*, 2001; Reyes-Franco *et al.*, 2006; Almeida *et al.*, 2003 and Jana *et al.*, 2003).As the pathogen has no sexual phase, genetic diversity is produced either by fusion of vegetative cells or by parasexual recombination between nuclear genes (Carlile, 1986). In nature genetic variability improves survival of a fungus (Bashasab, and Kuruvinashetti, 2007).

It is quite evident that variability in morphology, physiology, genetics, pathogenicity, and so forth is imperative for the fungus to have better adaptation in response to diversified environmental behavior. It also leads to host-plant resistance, development of resistant varieties of different crops against disease, and implementation of new disease controlling strategies (Purkayastha *et al.*, 2006).

The determination of variability among *R. bataticola* isolates is fundamental to guide the development of appropriate strategies for disease management according to different agro ecological zones. The present studies provide information on the variability of *R. bataticola* in major soybean growing states of India. These results will be useful in developing integrated strategies for the management of soybean root rot and breeding programs for pulses and other crops.

References

Abawi, G.S. and M. A. Pastor-Corrales. 1990. Root Rots of Beans in Latin America and Africa: Diagnosis, Research Methodologies and Management Strategies, CIAT, Cali, Colombia.

Aghakhani Maryam and S.C. Dubey. 2009. Morphological and pathogenic variation among isolates of *Rhizoctonia bataticola* causing dry root rot of chickpea. Indian Phytopath. 62 (2): 183-189.

Almeida, A. M. R., R. V. Abdelnoor, C. A. A. Arias. 2003. Genotypic diversity among Brazilian isolates of *Macrophomina phaseolina* revealed by RAPD. Fitopatologia Brasileira. 28 (3): 279–285.

Atiq, M., A. Shabeer and I. Ahmed. 2001. Pathogenic and cultural variation in *Macrophomina phaseolina*, the cause of charcoal rot in sunflower. Sarhad Journal of Agriculture. 2: 253–255.

Carlile, M. J. 1986. Genetic exchange and gene flow: their promotion and prevention. p. 203–214. In: A. D. M. Rayner, C. M. Brasier, and D. Moore, (ed). Evolutionary Biological of the Fungi. Cambridge University Press, Cambridge, UK.

Cloud G. L. and J. C. Rupe. 1988. Preferential host selection of isolates of *Macrophomina phaseolina*.

Phytopathology. 78 (12): 1563.

Datta Upma, Sachin Gupta, C. S. Kalha and V. K. Razdan. 2013. Morpho cultural and pathogenic variability isolates *Rhizoctonia solani* causing sheath blight of rice in Jammu. J. Mycol. Pl. Pathol.43 (2): 210-215.

Dhingra, O. D. and D. Chagas. 1981. Effect of soil temperature, moisture and nitrogen on competitive saprophytic ability of *Macrophomina phaseolina*. Transactions of British Mycological Society. 77 (1): 15-20.

Dhingra, O. D. and J. B. Sinclair. 1973. Location of *Macrophomina phaseolina* on soybean plants related to cultural characteristics and virulence. Phytopathology. 63 (7): 934-936.

Dhingra, O. D. and J. B. Sinclair. 1978. Biology and Pathology of *Macrophomina phaseolina*. Universidade Federal de Vicosa, Minas Gerais, Brazil.

Jana, T., T. R. Sharma, R.D. Prasad and D. K. Arora. 2003. Molecular characterization of *Macrophomina phaseolina* and *Fusarium* species by a single primer RAPD technique. Microbiological Research. 158 (3): 249–257.

Mayek-P´erez, N., C. L´opez-Casta˜neda, M. Gonz´alez-Chavira. 2001. Variability of Mexican isolates of *Macrophomina phaseolina* on basis of pathogenesis and AFLP genotype. Physiological and Molecular Plant Pathology. 59 (5): 257–264.

Meena Shekhar, R. C. Sharma, Sujay Rakshit, Poonam Yadav, Lokendra Singh and Ram Dutta, 2006. Genetic variability in *Macrophomina phaseolina* (Tassi.) Goid. incitant of charcoal rot of maize in India. Indian Phytopath. 59 (4): 453-459.

Mihail, J. D. and S. J. Taylor. 1995. Interpreting variability among isolates of *Macrophomina phaseolina* in pathogenicity, pycnidium production, and chlorate utilization. Canadian Journal of Botany.73 (10): 1596–1603.

Papavizas, G. C. 1977. Some factors affecting survival of sclerotia of *Macrophomina phaseolina* in soil. Soil Biology and Biochemistry. 9 (5): 337-341.

Pawar, N. B. 2010. Pathogenic variability among the isolates of chickpea wilt complex. Ph. D diss., Dept. Plant Path., Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola, Maharashtra, India.

Pearson, C. A. S., J. F. Leslie and F. W. Schwenk. 1986. Variable chlorate resistance in *Macrophomina phaseolina* from corn, soybean, and soil. Phytopathology. 76 (6) 646-649.

Purkayastha, S., B. Kaur, N. Dilbaghi and A. Chaudhury. 2006. Characterization of *Macrophomina phaseolina*, the charcoal rot pathogen of cluster bean, using conventional techniques and PCR-based molecular markers. Plant Pathol. 55: 106- 116.

Rajkumar, F. Bashasab, and M. S. Kuruvinashetti, 2007. Genetic variability of sorghum charcoal rot pathogen (*Macrophomina phaseolina*) assessed by random DNA markers. Plant Pathology. 23: 45–50.

Raut, J.G. and R.W. Ingle. 1989. Variations in isolates of *Rhizoctonia bataticola*. Indian Phytopath. 42 (4): 506-508.

Reyes-Franco, M. C., S. Hern´andez-Delgado, R. Beas- Fern´andez, M. Medina-Fern´andez, J. Simpson, and N. Mayek-P´erez. 2006. Pathogenic and genetic variability within *Macrophomina phaseolina* from Mexico and other countries. Journal of Phytopathology. 154(7-8): 447–453.

Riaz A., S. H. Khan, S.M.Iqbal and M. Shoaib. 2007. Pathogenic variability among *Macrophomina phaseolina* (Tassi) Goid, isolates and identification of sources of resistance in mash against

charcoal rot. Pakistan Journal of Phytopathology. 19 (1): 44-46.

Sharma Mamta, Raju Ghosh, Ramesh R. Krishnan, Upala N. Nagamangala, Shiva Chamarthi, Rajeev Varshney and Suresh Pande, 2012. Molecular and morphological diversity in *Rhizoctonia bataticola* isolates causing dry root rot of chickpea (*Cicer arietinum* L.) in India. African Journal of Biotechnology. 11 (37): 8948-8959.

Sinclair J. B. 1982. Compendium of Soybean Diseases. 2nd ed. American Phytopathological Society, Saint Paul, Minn, USA.

Sinclair, J. B. and M. C. Shurlleff. 1975. Compendium of soybean diseases. Amer. Phytopath. Soc. Inc. Minnesota, USA: 8-10.

Su, G., S. O. Suh, R.W. Schneider and J. S. Russin. 2001. Host specialization in the charcoal rot fungus, *Macrophomina phaseolina*. Phytopathology. 91 (2): 120–126.

Umer Iqbal and Tariq Mukhtar. 2014. Morphological and pathogenic variability among *Macrophomina phaseolina* isolates associated with Mungbean (*Vigna radiata* L.) Wilczek from Pakistan. Scientific World Journal. Article ID: 950175. pages 9.

Wheeler, H. 1975. Plant Pathogenesis, Academic press, London, UK.

Wrather, J. Allen and Steve R. Koenning, 2006. Estimates of disease effects on soybean yields in the United States 2003-2005. J. of Nematology. 38 (2):173-180.

Yang, X. B. and S. S. Navi. 2005. First report of charcoal rot epidemics caused by *Macrophomina phaseolina* in soybean, in IOWA. Pl. Dis. 89(5): 26.

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