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Goskula Kiran

Ph.D. Scholar, Department of
Plant Pathology, Bidhan Chandra
Krishi Viswavidyalaya, Nadia,
West Bengal, India

Raghunath Mandal

Assistant Professor, Department of
Plant Pathology, Bidhan Chandra
Krishi Viswavidyalaya, Nadia,
West Bengal, India

Characterization of cultural and morphological variability of *Rhizoctonia solani* isolates from different regions of West Bengal

Goskula Kiran and Raghunath Mandal

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Abstract

Cultural and physiological characters of different *R. solani* isolates was studied on different solid media, pathogen growth, colony colour, morphology, branching type, width of mycelium, varied from medium to medium. Majority of isolates exhibiting in all three media viz., PDA, OMA and MEA respectively. The mycelium of the majority of the isolates was whitish brown in colour after three days of growth. Majority of the isolates abundant in growth. Right-angled branching was observed in case of all isolates tested in said media.

Keywords: *R. solani*, cultural, right-angled branching

Introduction

Rice (*Oryza sativa* L.) is a major staple crop, providing nearly 20% of global dietary energy and sustaining more than half of the world's population. To meet future food demands, global rice production must increase substantially in the coming decades. Sheath blight, caused by the soil-borne pathogen *Rhizoctonia solani*, is a major constraint to rice productivity, causing yield losses ranging from 5.9 to 69% depending on crop stage and disease severity (Venkat Rao *et al.*, 1990; Naidu, 1992)^[9, 14]. In India, rice is cultivated over 43.5 M ha, with West Bengal being one of the leading rice-producing states (Indiastat, 2018)^[7].

In West Bengal, particularly in the new alluvial agro-climatic zone of Nadia district, high humidity, heavy rainfall, fertile soils, and intensive cropping systems create favourable conditions for sheath blight development. The disease initially appears as water-soaked lesions on the leaf sheath near the water level, which later enlarge into greyish-white lesions with brown margins, leading to drying of leaves and tillers under severe infection.

Management of sheath blight is difficult due to the collective species nature of *R. solani*, which exhibits wide cultural, morphological, and pathogenic variability (Anderson, 1982)^[1]. Chemical control is often ineffective and unsustainable because of fungicide resistance, soil heterogeneity, and environmental concerns (Grasela *et al.*, 1990)^[6]. Hence, integrated and eco-friendly disease management strategies are increasingly emphasized (Deepak Reddy *et al.*, 2017)^[4]. Understanding the cultural and morphological variability of *R. solani* populations is essential for devising effective management strategies; therefore, the present study focuses on characterizing *R. solani* isolates from different regions of West Bengal.

Materials & Methods

Isolation and purification of the pathogen

Cultural Variability

Cultural variability of *Rhizoctonia solani* isolates was studied on Potato Dextrose Agar (PDA), Oat Meal Agar (OMA), and Malt Extract Agar (MEA) along with their respective broths. Standard media compositions were used. Isolates were incubated at 28±2°C to record colony colour, texture, growth rate, and colony diameter.

Colony colour was assessed at 10 days using Munsell's colour chart following Burpee *et al.* (1980)^[2]. Colony growth pattern (round, elliptical, radiated) was recorded from 3-day-old cultures as described by Singh *et al.* (2014)^[11].

Corresponding Author:

Goskula Kiran

Ph.D. Scholar, Department of
Plant Pathology, Bidhan Chandra
Krishi Viswavidyalaya, Nadia,
West Bengal, India

Colony diameter and growth rate were measured at 12-hour intervals for 3 days using 5-mm mycelial discs with four replications.

Sclerotial Variability

Sclerotial characters including time of initiation, pattern, size, texture, location, and colour were recorded on PDA after 15 days at 28±2°C. Isolates were categorized as fast, moderate, or slow based on sclerotial initiation. Sclerotial colour was determined using Munsell soil colour chart and grouped as light brown, brown, or dark brown (Burpee *et al.*, 1980)^[2].

Morphological Variability: Morphological traits such as hyphal width, septation, and angle of branching were studied using lactophenol cotton blue staining under an Olympus CH 20i microscope. Observations were recorded from 3-day-old mycelia with five replications per isolate. Pathogenic Variability Pathogenic variability was assessed using the detached leaf method. Leaf blades (5 cm) were placed on Benzimidazole (400 ppm) and inoculated with mycelium or sclerotia. Plates were incubated at 26-28°C, and lesion development was recorded within 24-72 hours.

Results & Discussion

4.1 Cultural and physiological studies

4.1.1 Cultural characteristics and morphological characteristic of *R. solani* on different solid media

The cultural and morphological characteristics of ten isolates of *Rhizoctonia solani* were studied based on average radial growth, colony colour, and growth pattern on different solid media, namely Potato Dextrose Agar (PDA), Oat Meal Agar (OMA), and Malt Extract Agar (MEA), after three days of incubation at 28±2 °C. The results are presented in Tables 4.1.1, 4.1.2 and 4.1.3.

Data presented in Table 4.1.1 indicated that among the ten isolates, seven isolates (RS-2, RS-5, RS-6, RS-7, RS-8, RS-9 and RS-10) exhibited maximum radial growth on PDA after three days of incubation, whereas the least growth was recorded in isolate RS-1. The mycelium of most isolates appeared

whitish-brown in colour after three days. However, isolates RS-1 and RS-5 produced dark white colonies, while RS-3 showed sand-yellow pigmentation. The growth pattern was abundant in the majority of isolates, except RS-1, RS-3 and RS-4, which exhibited comparatively sparse growth (Plates 4.2 and 4.3). Similar variability in colony colour and growth behaviour of *R. solani* on PDA has been reported by Ghosh *et al.* (2019), Yadav *et al.* (2021), and Singh *et al.* (2022)^[5, 12, 14].

In the case of Oat Meal Agar (OMA) (Table 4.1.2), seven isolates (RS-2, RS-5, RS-6, RS-7, RS-8, RS-9 and RS-10) again showed maximum growth after three days, while RS-1 recorded minimum growth. The majority of isolates produced whitish-brown mycelium, whereas RS-1 and RS-5 showed dark white colonies and RS-3 exhibited sand-yellow colouration. An abundant growth pattern was observed in most isolates, except a few showing moderate to sparse growth (Plate 4.3). Comparable results on enhanced mycelial growth of *R. solani* on OMA have been documented by Sarma *et al.* (2020)^[10].

On Malt Extract Agar (MEA) (Table 4.1.3), significant differences in radial growth were observed among all the isolates of *R. solani*. The mycelium of most isolates was whitish-brown, while RS-1 and RS-5 produced dark white colonies, and RS-3 showed sand-yellow pigmentation. The growth pattern of the majority of isolates was abundant on MEA, indicating favourable utilization of nutrients present in the medium (Plate 4.3). Similar observations on the influence of MEA on colony morphology and growth rate of *R. solani* have been reported by Yadav *et al.* (2021), Singh *et al.* (2022)^[12, 14]. Across all the media tested, the mycelium of *R. solani* isolates consistently exhibited characteristic right-angled branching, which is a diagnostic feature of the pathogen (Plate 4.4). These observations are in agreement with earlier findings by Dasgupta (1992), Upadhyaya *et al.* (2014), Singh *et al.* (2014)^[11], and Lal *et al.* (2014). Recent studies by Ghosh *et al.* (2019), Sarma *et al.* (2020), Yadav *et al.* (2021)^[3, 5, 8, 10, 14], further confirm that *R. solani* exhibits considerable cultural and morphological variability depending on nutritional media, supporting its classification as a highly adaptable and phenotypically diverse pathogen.

Table 4.1.1: Culture characteristic of different isolates of *R. solani* on PDA medium

Isolate	Variety	Place of collection	Colony colour	Growth pattern	Branching pattern	Mycelial width(μm)	Colony growth dia(mm) 3DAI
RS-1	Swarna (MTU7029)	Chakdah	Dark white	Scarce	Right angle	6.9	50
RS-2	Ajit	Kalyani	Whitish brown	Abundant	Right angle	7.2	90
RS-3	Swarna (MTU7029)	Gayeshpur	Sand yellow	Moderate	Right angle	8.5	85
RS-4	Pratiksha	Gayeshpur	Whitish brown	Moderate	Right angle	7.1	85
RS-5	Swarna (MTU7029)	Gopalpur, Bardhaman	Dark white	Abundant	Right angle	7.3	90
RS-6	Swarna (MTU7029)	Jaguli	Whitish brown	Abundant	Right angle	7.5	90
RS-7	Swarna (MTU7029)	Balindi	Whitish brown	Abundant	Right angle	9.1	90
RS-8	Pratiksha	Balindi	Whitish brown	Abundant	Right angle	6.7	90
RS-9	Swarna (MTU7029)	Galsi, Bardhaman	Whitish brown	Abundant	Right angle	8.8	90
RS-10	Satabdi	Jaguli	Dark white	Abundant	Right angle	6.6	90

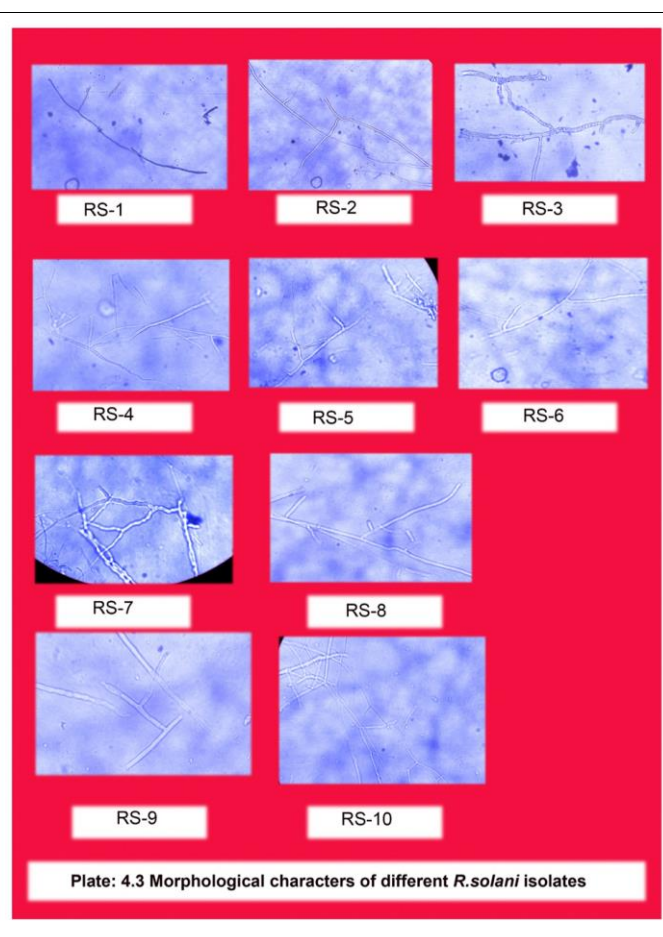
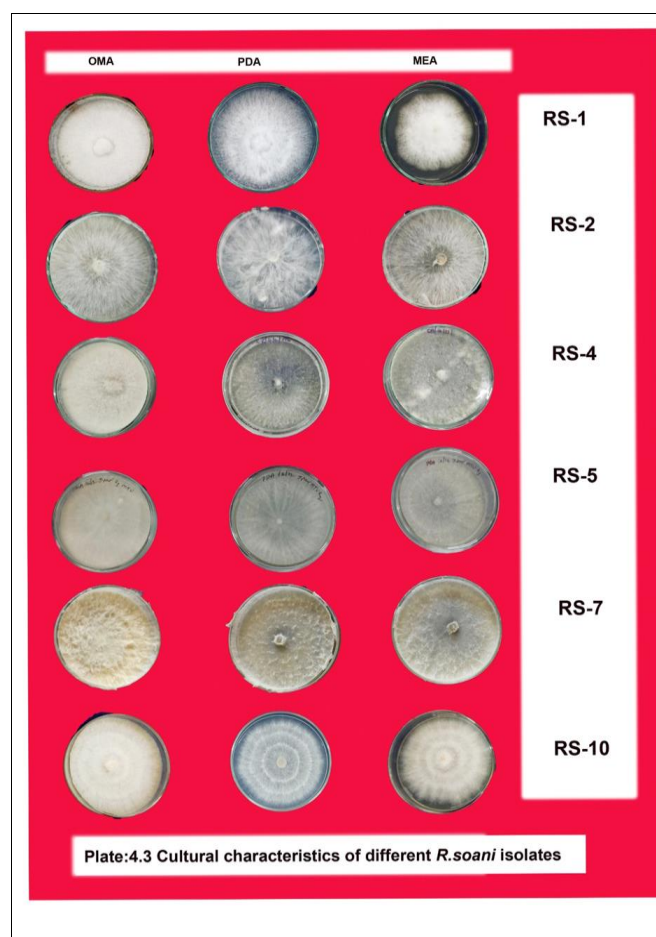
Table 4.1.2: Clutural characteristic of different isolates of *R.solani* on OMA medium

Isolate	Variety	Place of collection	Colony colour	Growth pattern	Branching pattern	Mycelial width(μm)	Colony growth dia(mm) 3DAI
RS-1	Swarna (MTU7029)	Chakdah	Dark white	Scarce	Right angle	6.8	50
RS-2	Ajit	Kalyani	Whitish brown	Abundant	Right angle	6.9	90
RS-3	Swarna (MTU7029)	Gayeshpur	Sand yellow	Moderate	Right angle	8.3	80
RS-4	Pratiksha	Gayeshpur	Whitish brown	Moderate	Right angle	7.1	85
RS-5	Swarna (MTU7029)	Gopalpur, Bardhaman	Dark white	Abundant	Right angle	7.1	90
RS-6	Swarna (MTU7029)	Jaguli	Whitish brown	Abundant	Right angle	7.3	90
RS-7	Swarna (MTU7029)	Balindi	Whitish brown	Abundant	Right angle	8.5	90

RS-8	Pratiksha	Balindi	Whitish brown	Abundant	Right angle	6.5	90
RS-9	Swarna (MTU7029)	Galsi, Bardhaman	Whitish brown	Abundant	Right angle	8.2	90
RS-10	Satabdi	Jaguli	Dark white	Abundant	Right angle	6.6	90

Table 4.1.3: Culture characteristic of different isolates of *R. solani* on MEA medium

Isolate	Variety	Place of collection	Colony colour	Growth pattern	Branching pattern	Mycelial width(μ m)	Colony growth dia(mm)
							3DAI
RS-1	Swarna (MTU7029)	Chakdah	Dark white	Scarce	Right angle	6.7	50
RS-2	Ajit	Kalyani	Whitish brown	Abundant	Right angle	6.8	90
RS-3	Swarna (MTU7029)	Gayeshpur	Sand yellow	Moderate	Right angle	8.3	85
RS-4	Pratiksha	Gayeshpur	Whitish brown	Moderate	Right angle	6.9	85
RS-5	Swarna (MTU7029)	Gopalpur, Bardhaman	Dark white	Abundant	Right angle	7.0	90
RS-6	Swarna (MTU7029)	Jaguli	Whitish brown	Abundant	Right angle	7.1	90
RS-7	Swarna (MTU7029)	Balindi	Whitish brown	Abundant	Right angle	8.2	90
RS-8	Pratiksha	Balindi	Whitish brown	Abundant	Right angle	6.4	90
RS-9	Swarna (MTU7029)	Galsi, Bardhaman	Whitish brown	Abundant	Right angle	8.0	90
RS-10	Satabdi	Jaguli	Dark white	Abundant	Right angle	6.6	90



Conclusions: The study revealed considerable cultural and morphological variability among *Rhizoctonia solani* isolates collected from different regions of West Bengal. Significant differences in radial growth, colony colour, and growth patterns were observed across PDA, OMA, and MEA media, while all isolates exhibited characteristic right-angled hyphal branching. This variability highlights the adaptive nature of the pathogen and explains inconsistencies in disease severity and management. The findings emphasize the need for region-specific and integrated strategies for effective sheath blight management in rice.

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