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Effect of Different Culture Media on Growth and Establishment of Phomopsis vexans Inciting Fruit Rot of Brinjal (Solanum melongena L.)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The fruit rot caused by *Phomopsis vexans* (Sacc. & Syd.) Harter is a destructive disease of brinjal and is considered to be a major constraint for its production. The present study was carried out at the Centre of Excellence for Sanitary and Phytosanitary, Department of Plant Pathology, Sardar Vallabhbhai Patel University of Agriculture and Technology, Meerut, for the evaluation of different culture media on growth and development of *Phomopsis vexans*. The study showed the maximum radial growth on OMA (90.00 mm), followed by PDA (88.20 mm) and Sabouraud Dextrose Agar (80.16 mm). The pycnidial densities were recorded as excellent on OMA and TFA, good on PDA, SDA, BFA, and MEA, and poor on YMA and CMA media with brown to black colour, while on RSA



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and WA they were absent. The colony colour were observed on several cultural media with circular and irregular shapes. The maximum pycnidial size was recorded on TFA (454.35x316.18 µm), followed by SDA (415.60x 296.26 µm) and BFA (324.72x368.84 µm), whereas the minimum was recorded on CMA (289.48x128.52 µm), followed by MEA (308.66x140.25 µm). The α -conidia were noticed to be sub-cylindrical and elliptical in shape and largest conidia observed on TMA (10.6x2.6µm), followed by BFA (9.8x2.6µm), and the smallest on RSA (6.1x1.6µm). The number of conidia was recorded highest on OMA (29.) medium and lowest on RSA (12.63). The β -conidia were not observed in this study. The number of guttullae were recorded 1-2 in all tested culture media except TFA and SDA, which had 2-3.

Keywords: Brinjal; culture media; fruit rot; pathogen.

1. INTRODUCTION

India is considered to be native land of the brinjal (Solanum melongena L.), a member of the Solanaceae family [1]. Brinjal is a popular vegetable cultivated in India and other countries. After China, India is the world's second largest producer of brinjal. It's a significant and native vegetable crop in India that's frequently referred to the farmers as cash crop. In India, major growing state of brinjal are West Bengal, Orissa, Bihar, Gujarat, Maharashtra, Andhra Pradesh and Karnataka. For every 100 g of fruit pulp, brinjal fruits provide 0.3 g, 1.4 g, and 0.3 g of lipids, proteins, and minerals, respectively [2].

different phases of its growth At and development, the crop is vulnerable to a variety of biotic and abiotic challenges: the most important ones are Phomopsis blight and fruit rot caused by Phomopsis vexans (Sacc. and Syd.) Harter is most destructive one. The diseaseaffected fruits and plants have an unpleasant, unattractive appearance that renders them unmarketable and inedible. Though leaves and stems may also show signs, fruits are most frequently impacted, with small, circular lesions showing on them. Sunken, discolored, and soft fruit lesions are surrounded by black fruiting bodies. In dry conditions, infected fruits turn shrivelled, dry and form black mummies [3]. Phomopsis vexans produce two types of conidia i.e., α and β conidia. Both conidia have been exposed to various carbon sources, plant components, plant leachates, and water grades Theα-conidia [4]. are released from pycnidia and disseminated by insects, rain, and contaminated equipment, are the main cause of the disease. The conidia are quickly germinate due to presence of free moisture on plant surface [5]. This disease typically causes a 15-25% reduction in crop yield [6,7].

2.MATERIALS AND METHODS

The experiment was conducted at the Centre of Excellence for Sanitary and Phytosanitary (SPS) Laboratory, Department of Plant Pathology, Vallabhbhai Patel Sardar University of Agriculture and Technology, Meerut, U.P. The pathogen was isolated from the diseased fruits of brinjal. The diseased fruits were first washed thoroughly under tap water and dried with the help of blotting paper. After that, cut small bits of infected tissue along with healthy tissues from diseased fruits. These bits were surface sterilized in a 1 percent sodium hypochlorite solution for about 40-60 seconds and two to three times rinsed with distilled water to remove the chemical. Then these bits were placed between the sterilized blotting paper to remove the water from the surface. These bits were placed aseptically in Petri plates containing potato dextrose agar medium and incubated in a BOD incubator at 26 ±1 °C for 2-3 days then pathogen was purified. Phomopsis vexans was used from cultures and five mm discs pure of pathogen was inoculated at the centre of sterilized Petri plates containing ten different culture media viz., Oat meal agar, Potato dextrose agar, Yeast malt agar, Water agar, Malt extract agar, Brinjal fruit agar, Tomato fruit agar, Corn meal Sabouraud agar, Richards dextrose agar, and synthetic Petri plates media. The were agar incubated for 7 days at 26 ±1°C in BOD incubator after that cultural and and morphological characteristics of the pathogen were observed.

3. RESULTS AND DISCUSSION

Isolated pathogen was grown on different culture media at 25 ± 1 °C temperature with pH 6.5. Several cultural characteristics were studied on different culture media such as colony colour, zonation, growth rate, shape, radial growth and

pycnidial density. The maximum radial growth of Phomopsis vexans were recorded on Oat meal agar (90.00 mm) followed by potato dextrose agar (88.20) and sabouraud dextrose agar (80.16 mm) after seven days of incubation whereas minimum radial growth was recorded on Richard's synthetic agar (16.66 mm) followed by Water agar (32.33 mm) and Corn meal agar (48.00 mm). The fast growth rate of pathogen was observed on Oat meal agar, Potato dextrose agar, Sabouraud dextrose agar, Brinjal fruit agar, Tomato fruit agar, Yeast malt agar, medium growth rate on Corn meal agar, Malt Extract Agar and slow growth rate on Water agar, Richard's synthetic agar medium. In case of colony colour, white mycelial growth was showed on Oat meal agar, Potato dextrose agar, Sabouraud dextrose agar, Richard's synthetic agar, and Yeast malt agar, grevish white colony exhibited on Brinjal fruit agar. Tomato fruit agar and Malt extract agar while creamy white colony appeared on Corn meal agar and grey on Water agar media. The colony shape was recorded irregular on Potato dextrose agar, Sabouraud dextrose agar, and Brinjal fruit agar while circular shape on Oat meal agar, Richard's synthetic agar, Yeast malt agar, Tomato fruit agar, Malt extract agar, Corn meal agar, and Water agar. The distinct zonation was found on Potato dextrose agar, Sabouraud dextrose agar, Yeast malt agar, Malt extract agar and Corn meal agar and Indistinct zonation on Tomato fruit agar whereas no zonation was found on Brinjal fruit agar, Oat meal agar, Richard's synthetic agar, Malt extract agar, water agar. The excellent pycnidial density was recorded on Oat meal agar and Tomato fruit agar while on Richard's synthetic agar and Water agar were absent and rest five culture media showed the good pycnidial density except Yeast malt agar had poor depicted in Table 1. Whereas for conformity of result compares with Jamir, [8] who reported the pathogen was differed in various aspect viz. colony colour, shape and zonation etc on different culture media and recorded radial PDA maximum growth on media and minimum radial growth on CDA media. Verma, [9] also observed similar result and recorded maximum radial growth of pathogen on PDA media. Several workers reported Potato dextrose agar medium to be good for mycelium growth of pathogen [10,11,12].

Several morphological characteristics were observed on different culture media viz: colour, shape, size, density pycnidia, occurrence, concentration, size as well as presence of

guttulae in conidia. Pycnidial colour was noticed black and brown on different culture media as well as flask and spherical shape was observed. The size of pycnidia was recorded maximum on Tomato fruit agar (454.35×316.18 µm) followed by Sabouraud dextrose agar (415.60x 296.26 μm) and Brinjal fruit agar (324.72×368.84 μm). The smallest size of pycnidia was recorded on Corn meal agar (289.48×128.52 µm) followed by Malt extract agar (308.66×140.25 µm) and Oat meal agar (235.42×188.65 µm) whereas no pycnidial formation on Richard's synthetic agar and Water agar media were observed. Only aconidia were occurred on all culture media and a-conidia shape was observed elliptical or sub cylindrical. The α -conidia were varied in size. Smallest α-conidia were found on Richard's synthetic agar (6.1×1.6 µm) followed by (6.8×1.6 µm) on Corn meal agar, (8.6×1.7 µm) on Yeast malt agar and (7.8×2.1µm) on Malt extract agar while as largest found on Tomato fruit agar (10.6×2.6 µm) followed by Brinjal fruit agar (9.8×2.6µm) and Oat meal agar (8.8×2.7 µm). Minimum spore count of a-conidia was recorded on Richard's synthetic agar (12.63×10⁴ per mL) followed by Corn meal agar (14.97×104 per mL), Yeast malt agar (16.33×10⁴ per mL) whereas maximum on Oat meal agar (29.24×10⁴ per mL) followed by Tomato fruit agar (25.86×10⁴ per mL). Majority showed two guttulae in α-conidia of pathogen. One to two guttulae were recorded in all culture media except Sabouraud dextrose agar and Tomato fruit agar exhibited two to three guttullae presented in Table 2. The presented work confirms by Jamir, [8] who observed that variation in the presence of guttulae (oil drop) in α-conidia was evident. Mostly 2 guttulae was recorded in PDA media. The similar results were showed by Devi, [13] who observed several colony characters including mycelial growth. colony colour, zonation and sporulation of Phomopsis vexans were differ on various culture media. The best performance of pathogen was observed on potato dextrose agar media and heavy sporulation on oat meal agar medium while thin mycelial growth with no zonation in case of synthetic media. Zhao and Simon [14] noticed that different culture media were significantly influenced the mycelium growth rate and conidial production of Phoma exigua and observation the α -conidia were subcylindrical to elliptical with size as (10.0 µm x 2.5 µm) size of pycnidia on PDA media was (58.49-381.49 µm x 79.3- 439 μm) whileβ-conidia were not observed throughout the study [15].Patil, [16] showed the results *i.e.*, growth behaviour on five different media (PDA, TDA, MEA, PCA and host leaf extract agar) showed significant difference in d colour, morphology with sporulation in potato

dextrose agar media.

Table 1. Cultural characteristics of Phomopsis vexans on different culture media

Culture Media	Colour	Shape	Growth rate	Radial growth	Zonation	Pycnidial density
Potato Dextrose Agar	White	Irregular	Fast	88.20	Distinct	+++
Sabouraud Dextrose Agar	White	Irregular	Fast	80.16	Distinct	+++
Richard's synthetic agar	White	Circular	Slow	16.66	Absent	_
Tomato Fruit Agar	Greyish White	Circular	Fast	68.83	Indistinct	++++
Yeast Malt Agar	White	Circular	Fast	65.25	Distinct	+
Oat Meal Agar	white	Circular	Fast	90.00	Absent	++++
Brinjal Fruit Agar	Greyish White	Irregular	Fast	69.66	Absent	+++
Malt Extract Agar	Greyish white	Circular	Medium	51.25	Absent	+++
Water Agar	Grey	Circular	Slow	32.33	Absent	_
Corn Meal Agar	Creamy White	Circular	Medium	48.00	Distinct	+

- Absent, + poor, +++ Good, ++++ Excellent



Fig. 1. Showing Radial growth on different culture media, a) Yeast malt agar b) Brinjal fruit agar c) Malt extract agar d) Sabouroud dextrose Agare) Corn meal agar f) Tomato fruit agar g) Oat meal agar h) Potato dextrose agar i) Richard's synthetic agar j) Water agar

Pycnidial Character									
Culture Media	Colour	Shape	Size (µm)	Occurrence Alpha (α)	e of conidia Beta (β)	Shape	Size (µm)	No. of Guttulae	Concentration of α- conidia (×10 ⁴) /
Potato Dextrose Agar	Black	Flask	278.96×192.38	+		Sub cylindrical	9.4×2.3	1-2	21.42
SabouroudDextrose	Black	Spherical	415.60× 296.26	+	_	Sub cylindrical	8.4×2.4	2-3	20.00
Richards Agar				+		Sub cylindrical	6.1×1.6	1-2	12.63
Tomato Fruit Agar	Brown	Spherical		+	_	Elliptical	10.6×2.6	2-3	25.86
Yeast Malt Agar	Brown	Flask	319.88× 153.71	+	_	Elliptical	8.6×1.7	1-2	16.33
Oat Meal Agar	Black	Flask	235.42×188.65	+	_	Sub cylindrical	8.8×2.7	1-2	29.24
Brinjal Fruit Agar	Black	Spherical	324.72×368.84	+	_	Sub cylindrical	9.8×2.6	1-2	21.13
Malt Extract Agar Water Agar	Brown	Spherical	308.66×140.25	+	_	Elliptical	7.8×2.1	1-2	18.51
Corn Meal Agar	Black	Spherical		+	_	_ Sub cylindrical			_ 14.97

Table 2. Morphological characteristics of *Phomopsis vexans* on different culture media

4. CONCLUSION

In this finding, several cultural media were examined that significantly affected the cultural and morphological characters of Phomopsis vexans. The maximum radial growth and excellent pycnidial formation were recorded on oat meal agar media with heavy sporulation, whereas the minimum radial growth was observed on Richards synthetic agar, followed by water agar and corn meal agar. The observation of pycnidial formation was poor on corn meal agar while absent on Richards synthetic agar and water agar media. In cases of the occurrence of conidia, the α -conidia were recorded with a subcylindrical or elliptical shape. while the β-conidia were absent throughout the investigation. Mostly 1-2 guttulae were noticed in all culture media except tomato fruit agar, and sabouroud dextrose agar had 2-3 guttulae. This study may be useful for future researchers to develop a management strategy for the disease.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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