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Evaluation of culture media to select the most suitable medium for the growth of leaf spot of pigeonpea [*Cajanus cajan* (L.) Millsp.] Caused by *Cercospora cajani*

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Abstract

Cercospora leaf spot caused by *Cercospora cajani* Henningsis one of the most important fungal diseases of Pigeonpea [*Cajanus cajan* (L.) Millsp.]. Out of six culture media maximum fungal growth was recorded in Potato dextrose agar medium followed by V-8 agar juice, seed extract arhar agar, Carrot dextrose agar, Old leaves extract arhar agar and new leaf extract arhar agar at seven days. Out of six culture media maximum (83.00 mm) fungal growth was recorded in Potato dextrose agar medium.

Keywords: Media, suitable, pigeonpea, *Cercospora cajani*, *Cajanus cajan* L.

Introduction

Pigeonpea [*Cajanus cajan* (L.) Millsp.] is an important grain legume crop of rainfed agriculture in the semi-arid tropics. Besides Indian sub-continent, it is widely grown in Eastern Africa and Central America. It is not only an important source of protein, but also plays an important role in atmospheric nitrogen fixation into soil (Reddy *et al.*, 2012) [5]. Globally pigeonpea is cultivated in about on 4.7 million ha area with 3.69 million tonnes annual production. India accounts 78% of the global output with current production of 2.78 million tonnes from 3.5 million ha. In Madhya Pradesh Pigeonpea is grown in about 0.57 million ha with an annual production of 0.57 million tonnes. The average yield of Pigeonpea in M.P. is 848 kg/ha which is much larger than the potential yield of crop (1500-2000 kg/ha). Several biotic and abiotic factors are responsible for reducing the yield (Anno. 2018) [1]. *Cercospora* leaf spot inflicts heavy yield losses ranging from 23 to 96 per cent under natural epiphytotic conditions. (Kasno, 1990; Iqbal *et al.*, 1995; Kaur, 2007) [3, 2, 4]. The yield losses vary depending upon how early the crop is infected in the season, crop variety and prevailing weather. The leaf spot disease is caused by fungus *Cercospora cajani*. It is present in parts of Uttar Pradesh, Bihar and several places of south India (Reddy *et al.*, 2012) [5].

Materials and Methods

Evaluation of culture media to select the most suitable medium for the growth of *Cercospora cajani*

The different media were evaluated for obtaining maximum mycelial growth of the *Cercospora cajani*. The experiment was laid out in complete randomized design with replicated in four times. Six solid culture media viz., Seed extract arhar, Old leaves extract, New leaves extract arhar, V-8 agar juice Carrot agar and Potato Dextrose agar used to compare the growth rate of *Cercospora cajani* (Table-2). The Culture Medium were prepared by the standardized method and autoclaved at 121.6 °C, 15 psi pressure for twenty minutes. The quantities (20 ml) of each medium were poured in 90 mm Petri dish. Each Petri plate was inoculated separately with uniform mycelia culture bits (7 mm) cut with the help of cork borer from young (5 days) vigorously growing culture were placed on the middle of the each pre poured medium and incubated at 25±1 °C (Dela Paz *et al.*, 2006). Each treatment was replicated three times. The diameter of the growth of the fungus was measured after inoculation 3, 5 and 7 days on radial growth of mycelium were recorded.

Experiment details

Design: CRD

Replication: 4

Treatments: 6

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Table 1: Composition in culture medium

S. No.	Name of Media	Ingredients	Quantity
1	V-8 juice Agar	v-8 juice	44.3g
		Distilled water	1000 ml
2	Potato Dextrose Agar	Peeled and sliced potato	200.0 g
		Dextrose	20.0g
		Agar-agar	20.0g
		Distilled water	1000 ml
3	New leaves arhar agar	New leaves extract	20.0g
		Agar-agar	20.0g
		Dextrose	20.0g
		Distilled Water	1000.0 ml
4	Old leaves arhar Agar	Old leaves extract	200.0g
		Dextrose	20.0g
		Agar-agar	20.0g
		Distilled water	1000 ml
5	Carrot Dextrose Agar	Carrot extract	200.0g
		Dextrose	20.0g
		Agar-agar	20.0g
		Distilled water	1000 ml
6	Seed arhar Agar	Seed extract	200.0 g
		Dextrose	20.00 g
		Agar-agar	20.0g
		Distilled water	1000 ml

Results and Discussion

A total of six culture media were evaluated for the growth of *Cercospora cajani* and the mycelium growth was measured at 3, 5 and 7 DAI. It is evident from the (Table-2, Fig-1&Plate-1) that the significant difference in the fungal growth was observed in the tested media at 3, 5 and 7 DAI.

In 3 DAI the maximum mycelium growth (22.75 mm) was recorded in Potato dextrose agar medium followed by V-8 agar juice (20.75 mm), seed extract arhar agar medium (16.75 mm), Old leaves extract arhar agar medium (16.25 mm), New leaf extract arhar agar medium (10.75 mm), where as its minimum mycelium growth (13.25 mm) was recorded in

Carrot dextrose agar.

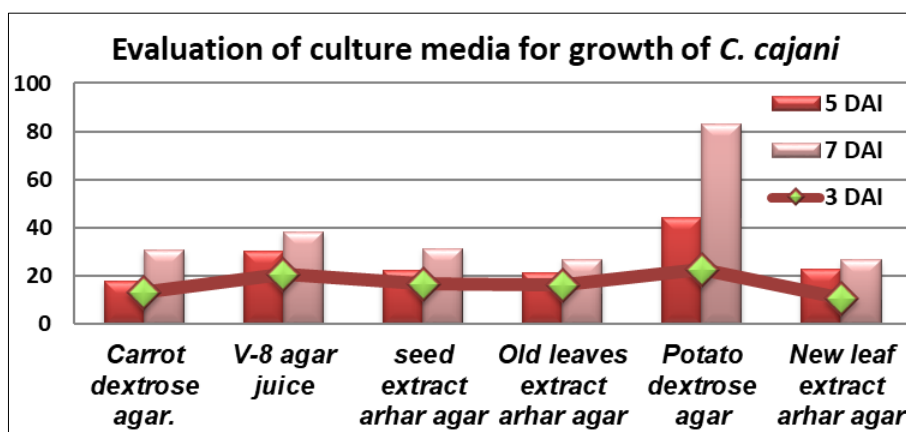
Potato dextrose agar medium is significantly superior to seed extract arhar agar medium, Old leaves extract arhar agar medium, New leaf extract arhar agar medium and Carrot dextrose agar and statistically at par with V-8 agar juice. Carrot dextrose agar is also statistically at par with new leaf extract arhar agar and significantly superior to other remaining media.

In 5 DAI the maximum mycelium growth (44.25 mm) was recorded in Potato dextrose agar medium (44.25 mm), followed by V-8 agar juice (30.25 mm), seed extract arhar agar medium (22.50 mm), New leaf extract arhar agar medium (22.75 mm), Old leaves extract arhar agar medium (21.50 mm), where as its minimum mycelium growth (18.00 mm) was recorded in Carrot dextrose agar. New leaf extract arhar agar statistically at par with old leaves extract arhar media and significantly superior over with remaining media.

In 7 DAI the maximum fungal growth was recorded in Potato dextrose agar medium (83.00mm), followed by V-8 agar juice (38.25 mm), seed extract arhar agar (31.25 mm), Carrot dextrose agar (30.75 mm), Old leaves extract arhar agar (26.75 mm) and New leaf extract arhar agar (26.50 mm). New and old extract arhar extract media both are statistically at par with Carrot dextrose agar media.

Table 2: In-vitro evaluation of culture media for growth of *C. cajani*

S. No	Culture medium	Radial growth (mm / day)		
		3 DAI	5 DAI	7DAI
1	Carrot dextrose agar.	13.25	18.00	30.75
2	V-8 agar juice	20.75	30.25	38.25
3	seed extract arhar agar	16.75	22.50	31.25
4	Old leaves extract arhar agar	16.25	21.50	26.75
5	Potato dextrose agar	22.75	44.25	83.00
6	New leaf extract arhar agar	10.75	22.75	26.50
S.Em±		1.017	1.192	1.659
CD at 5%		3.046	3.568	4.968

**Fig 1:** In-vitro evaluation of culture media for growth of *C. cajani*.

Colour of mycelia

Among all the tested media, the colour of mycelia did differ from each other. The colour of mycelia in all the used media were found white to off white in colour. In V-8 agar media and seed extract arhar agar media the mycelia colour are off white whereas Carrot dextrose agar, old leaves extract agar, potato dextrose agar, and new leaf extract agar mycelia colour was white.

Texture

In mycelia texture V-8 media and Potato dextrose agar media mycelia were smooth and regular patterns of growth whereas seed extract arhar, new leaves extract arhar, old leaves extract arhar and carrot agar media the mycelia pattern are scattered and irregular growth.

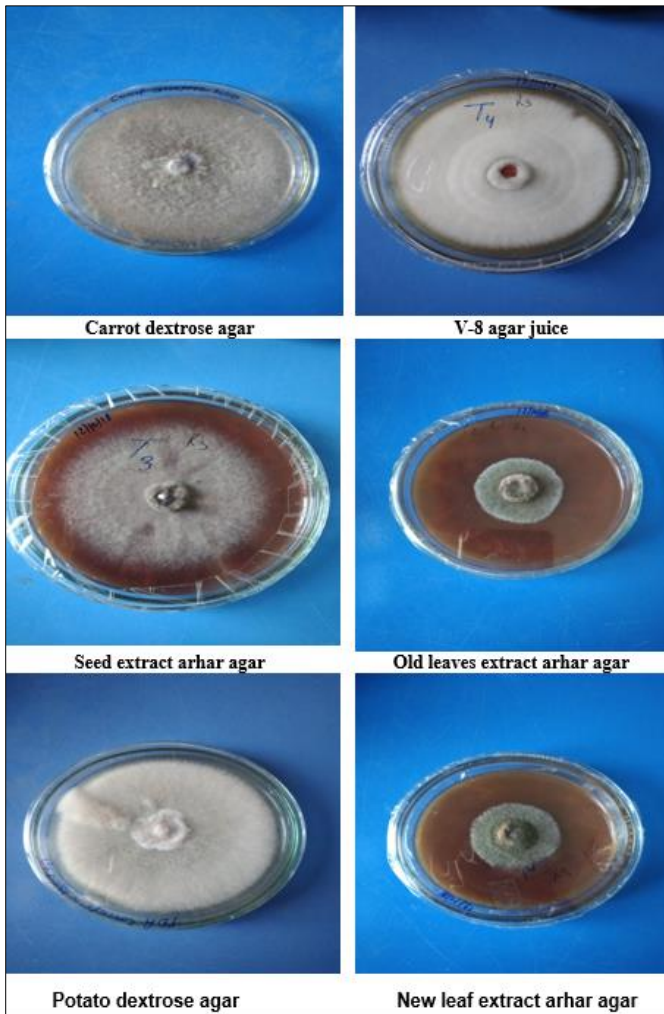


Plate 1: *In-vitro* evaluation of culture media against *Cercospora cajani*

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