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In-vitro Evaluation of Some Plant Leaf Extract against Coconut Leaf Spot Caused by *Pestalotia palmarum* (Cooke) in Bastar Plateau of Chhattisgarh

Vandana Chadar*, R. R. Bhanwar, Rajendra Kashyap, Shraddha Karcho and Luchika Rana

Department of Plant Pathology, S. G. College of Agriculture and Research Station, Jagdalpur, IGKV, Chhattisgarh, 494001, India

**Corresponding author*

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A B S T R A C T

In the present investigation 30 Plant leaf extract were evaluated in *in vitro* condition against *P. palmarum* adopting poisoned food technique. The per cent inhibition of pathogen was 100 per cent by Dhatura, Anjwain and Tobacco at 10 per cent concentration followed by the turmeric (95.84), safed musli (90.62), garlic (87.5), hathjodh (86.5), kalmegh (86.46), jetropha (83.34), neem bark (79.18), satavar (78.12), lemongrass (69.81), laung (67.71), ashoka (65.62), aadusa (63.68), karanj bark (63.06), bhringraj (62.5), karanj (62.5), dalchini (61.15), beshram (60.43), brijdanti (57.46), arandi (56.25), neem (54.18), nilgiri (54.18), pattharchatta (52.09), tulsi (46.87), marigold (40.62) and aloevera (40.62) whereas the lowest inhibition was recorded in amari (Gangura) with 36.46 per cent.

Introduction

Coconut is one of the major plantation crop in india. Coconut tree (*Cocos nucifera*) is a member of the palm tree family (Arecaceae). In India, coconut farming is inseparably embedded in the socio-historical culture as well as the ethnic identity. Considering the versatile nature of the crop and the multi-uses of its products, the coconut palm is eulogized as Kalpavriksha (Tree of Heaven). In India with a total cultivated area of 1975.81 thousand hectares with a production of 21,665 million nuts which makes India stand 3rd in the world.

India occupies the premier position in the world with an annual production of 13 billion nuts, overtaking Indonesia and the Philippines, the other two prominent coconut-growing

countries (Raghavi et al., 2019). Yield of the coconut also reduces day by day due to the causes of various diseases. Such as, sooty mould, stem bleeding, leaf spot, white thread blight, root rot, brown root rot and bud rot disease which are caused by different fungus. Among the diseases every year grey leaf spot disease caused by *Pestalotia palmarum* (Cooke.) attacks the gardens and decreases the growth and development of the tree as well as the yield of the fruit. The symptom is only developed in the mature leaves in the form of grayish white spots surrounded by brown margin. Several of the spots coalesce together and form irregular grey necrotic patches and show burnt or blighted appearances. The upper surface of the affected leaves reveals dark grey eruptions like pin heads. This disease is a serious problem all over the coconut growing regions of Bangladesh (Rahman et al., 2013).

Materials and Methods

Collection of sample

Diseased leaves of Coconut with typical leaf spot symptoms were collected from AICRP on Palms Research field of SGCARS, Jagdalpur (C.G.) (Fig. 2).

Preparation of potato dextrose agar (PDA) medium

The basic medium, PDA was prepared following the standard procedure (Anon. 1968). At first 200 g peeled potato is cut into slice and then boiled in 1000 ml water. After that it was sieved and 15 gm agar were mixed with it in a water bath, after few minutes 20 g dextrose were mixed with it and stirred properly so that it cannot be coagulated. The pH was adjusted to 6.5 of the media by using pH meter with the help of 1N HCL and sterilized in autoclave at 121°C temperature for 20 minutes.

Isolation of the fungus

The fungus was isolated from the infected leaf of coconut following tissue planting technique (Tuite, 1969). The infected diseased samples along with healthy tissues were cut into small pieces and surface sterilized by dipping in 0.1% sodium hypochloride (NaOCl) solution for two minutes. NaOCl on the surface of the leaf pieces was decanted by soaking with sterilized blotting paper. The cut pieces were then placed onto sterilized potato dextrose agar (PDA) in glass petridishes (20 ml/ petridish) and incubated in an incubator at 27 ± 1°C until mycelium formation. The hyphal tips were transferred onto PDA plate after growing the mycelium.

Identification of fungus

The fungus was then identified on the basis the morphological of characteristics with the help of identifying key book (Barnett and Hunter, 1972).

Purification

To obtain pure culture of the pathogen, the hyphal tips were transferred aseptically onto PDA plate by using the flame sterilized tip of an inoculation needle. The plate was incubated at room temperature for seven days.

Multiplication of *P. palmarum*

PDA was poured in sterilized petridishes, 25 ml in each. After solidification, the plates were inoculated by placing 5 mm discs of three days old PDA culture of *P. palmarum*. The discs were cut with flame sterilized cork borer (5 mm diameter). The inoculated petridishes were kept in the growth chamber at a temperature of 28 ± 1ºC for few days. All the works were undertaken under the laminar air flow cabinet.

Evaluation of different plant extract used in this experiment

These plant extracts were tested initially under in-vitro condition by using poison food technique (Schmitz, 1930). The fresh leaves were grounded in a blender with distilled water. The extract was filtered through double layered muslin cloth. The extracts were tried at concentration of 10 per cent for seed treatment, prepared by diluting the extract in distilled water (Table 1).

Different plant extract were evaluated in *in vitro* condition against *P. palmarum* following poison food technique (Dhingra and Sinclair, 1985). All the plant extract were tested at recommended by adopting poisoned food technique. The test pathogen was grown on PDA medium in Petri plates for seven days prior to setting up of experiment. The required plant extract was added to the melted PDA medium to obtain the desired concentration.

20 ml of poisoned medium was poured in each Petri plate. Suitable checks were maintained without addition of fungicides. A mycelial disc of five mm diameter was taken from the periphery of 7 days old colony and placed in the centre and incubated at 28 ± 2℃ for full growth of the fungus.

Three replications were maintained for each treatment. The radial growth of the colony was measured in two directions and average was recorded. Per cent inhibition was recorded by using the formula given by Vincent (1947) as under:

$$PI = \left[\frac{(C - T)}{C}\right] X\ 100$$

Where,

PI = Per cent inhibition, C = Growth in control and T = Growth in treatment.

Results and Discussion

Among the 30 plant leaf extract were evaluated against coconut leaf spot (*P. palmerum*) adopting poisoned food technique. Observations of the radial growth of the pathogen were recorded after 7 day after inoculation. The percent inhibition of the pathogen over control was calculated and presented in Table 2, Fig. 3 and Chart 1. The superiority in controlling the inhibition of pathogen was managed by Dhatura, Anjwain and Tobacco inhibited the growth of *P. palmarum* was 100 per cent.

No growth was found at given concentration. the turmeric (95.84), safed musli (90.62), garlic (87.5), hathjodh (86.5), kalmegh(86.46), jetropha (83.34), neem bark (79.18), satavar (78.12), lemongrass (69.81), laung (67.71), ashoka (65.62), aadusa (63.68), karanj bark (63.06), bhringraj (62.5), karanj (62.5), dalchini (61.15), beshram (60.43), brijdanti (57.46), arandi (56.25), neem (54.18), nilgiri (54.18), pattharchatta (52.09), tulsi (46.87), marigold (40.62) and aloevera (40.62) whereas the lowest inhibition was recorded in amari (Gangura) with 36.46 per cent.

Islam *et al.*, (2004) revealed that the two doses (4 and 5 %) of garlic extract were found most effective in inhibiting the redial growth of the fungus i. e. 88.76 per cent which favors the present study.

Fig.1 Pure culture of *P. palmarum*

Table.1 List of botanicals used in the experiment

S.N.	Treatment	Name of Botanicals	Botanical name
1.	T_1	Tulsi	*Ocimum tenuiflorum*
2.	T_2	Turmeric	*Curcuma longa*
3.	T_3	Marigold	*Tagetes spp.*
4.	T_4	Garlic	*Allium sativam*
5.	T_5	Jetropha	*Jatropha curcas*
6.	T_6	Dhatura	*Datura stramonium*
7.	T_7	Caster	*Ricinus communis*
8.	T_8	Pattharchatta	*Bryophyllum pinnatum*
9.	T_9	Vringraj	*Eclipta prostrate*
10.	T_{10}	Neem	*Azadirachta indica*
11.	T_{11}	Karanj	*Millettia pinnata*
12.	T_{12}	Neem bark	*Azadirachta indica*
13.	T_{13}	Kalmegh	*Andrographis paniculata*
14.	T_{14}	Satavar	*Asparagus racemosus*
15.	T_{15}	Ashoka	*Saraca asoca*
16.	T_{16}	Nilgiri	*Eucalyptus spp.*
17.	T_{17}	Laung	*Syzygium aromaticum*
18.	T_{18}	Anjwain	*Trachyspermum ammi*
19.	T_{19}	Tobacco	*Nicotiana tabacum*
20.	T_{20}	Lemongrass	*Cymbopogan spp.*
21.	T_{21}	Beshram	*Ipomoea carnea*
22.	T_{22}	Brijdanti	*Banteria prionitis*
23.	T_{23}	Karanj bark	*Millettia pinnata*
24.	T_{24}	Aloevera	*Aloe barbadensis*
25.	T_{25}	Amari (Gangura)	*Hibiscus sabdariffa*
26.	T_{26}	Aadusa	*Justicia adhatoda*
27.	T_{27}	Dalchini	*Cinnamamum verum*
28.	T_{28}	Safed musli	*Chlorophytum borivilianum*
29.	T_{29}	Hathjodh	*Cissus quadrangularis*
30.	**T_{30}**	**Control**	**Without phyto extract**

Table.2 Percent inhibition of the radial growth of the pathogen of coconut leaf spot in *in-vitro*

Treatment	Mean growth (mm) of pathogens	Percent inhibition of pathogens (%)
T_1	17.000	46.87
T_2	01.333	95.84
T_3	19.000	40.62
T_4	04.000	87.5
T_5	05.333	83.34
T_6	00.000	100
T_7	17.333	56.25
T_8	15.333	52.09
T_9	12.000	62.50
T_{10}	14.667	54.18
T_{11}	12.000	62.50
T_{12}	06.667	79.18
T_{13}	06.667	86.46
T_{14}	07.000	78.12
T_{15}	11.000	65.62
T_{16}	14.667	54.18
T_{17}	10.333	67.71
T_{18}	00.000	100
T_{19}	00.000	100
T_{20}	09.667	69.81
T_{21}	12.667	60.43
T_{22}	13.667	57.46
T_{23}	11.667	63.06
T_{24}	19.000	40.62
T_{25}	20.333	36.46
T_{26}	11.667	63.68
T_{27}	12.333	61.15
T_{28}	03.000	90.62
T_{29}	04.333	86.50
T_{30}	32.000	-
C.D at 5 %	**4.339**	
SE(m) ±	**1.530**	

Fig.2 Infected coconut leaf

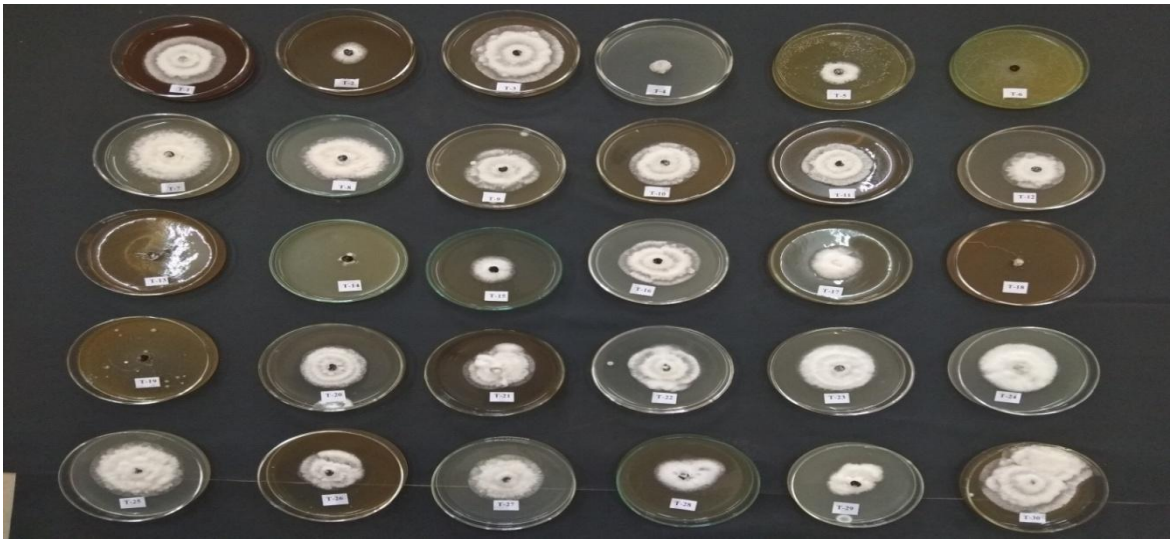

Fig.3 Effect of different plant leaf extract on *P. palmarum*

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