Original Research Article

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

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.

Keywords
Coconut leaf spot, plant leaf extract, In vitro, Pestalotia palmarum, Poisoned food technique

Article Info
Accepted: 04 March 2020
Available Online: 10 April 2020

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 ± 1°C 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°C 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:

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

Where,

\(PI = \text{Per cent inhibition, } C = \text{Growth in control and } T = \text{Growth in treatment.}\)

**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), patharchatta (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 radial growth of the fungus i. e. 88.76 per cent which favors the present study.

![Pure culture of *P. palmarum*](image)
### 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            | Trachyspernum 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 sabdariffia   |
| 26.  | $T_{26}$  | Aadusa             | Justicia adhatoda      |
| 27.  | $T_{27}$  | Dalchini           | Cinnamamum verum       |
| 28.  | $T_{28}$  | Safed musli        | Chlorophytum borivilianum|
| 29.  | $T_{29}$  | Hathjodh           | Cissus quadrangularar  |
| 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 (%) |
|-----------|-------------------------------|-------------------------------------|
| T₁        | 17.000                        | 46.87                               |
| T₂        | 01.333                        | 95.84                               |
| T₃        | 19.000                        | 40.62                               |
| T₄        | 04.000                        | 87.5                                |
| T₅        | 05.333                        | 83.34                               |
| T₆        | 00.000                        | 100                                 |
| T₇        | 17.333                        | 56.25                               |
| T₈        | 15.333                        | 52.09                               |
| T₉        | 12.000                        | 62.50                               |
| T₁₀       | 14.667                        | 54.18                               |
| T₁₁       | 12.000                        | 62.50                               |
| T₁₂       | 06.667                        | 79.18                               |
| T₁₃       | 06.667                        | 86.46                               |
| T₁₄       | 07.000                        | 78.12                               |
| T₁₅       | 11.000                        | 65.62                               |
| T₁₆       | 14.667                        | 54.18                               |
| T₁₇       | 10.333                        | 67.71                               |
| T₁₈       | 00.000                        | 100                                 |
| T₁₉       | 00.000                        | 100                                 |
| T₂₀       | 09.667                        | 69.81                               |
| T₂₁       | 12.667                        | 60.43                               |
| T₂₂       | 13.667                        | 57.46                               |
| T₂₃       | 11.667                        | 63.06                               |
| T₂₄       | 19.000                        | 40.62                               |
| T₂₅       | 20.333                        | 36.46                               |
| T₂₆       | 11.667                        | 63.68                               |
| T₂₇       | 12.333                        | 61.15                               |
| T₂₈       | 03.000                        | 90.62                               |
| T₂₉       | 04.333                        | 86.50                               |
| T₃₀       | 32.000                        |                                     |
| **C.D at 5 %** | **4.339**  |                                     |
| **SE(m) ±** | **1.530**  |                                     |
Acknowledgement

The author is grateful to acknowledge Mr. Rajesh Kumar Patel for providing technical guidance during entire course of work. The authors are also put sincere thanks to the AICRP on Palms, Jagdalpur and S. G. College of Agriculture and Research Station, Jagdalpur for proving necessary amenities during entire research of work.

References

Anonymous (1968) Plant Pathologist’s Pocket Book. Commonwealth Mycological Institute 394-395.

Barnett HL and Hunter BB. 1972. Illustrated Genera of imperfect Fungi. Burgess Publishing Company USA.

Dhingra, OD and Sinclair JB .1985. Basic Plant Pathology Methods. CRC Press, Boca Raton, USA.

Islam M.R., Hossain M.K., Bafar M.H. and Ali M.R. 2004. Identification of causal agent of leaf spot of betelnut and in vitro evaluation of fungicides and plant extract against it. Pakistan journal of biological science. 7(10): 1758-1761.

Raghavi. MD , Sakthi Balaa. M , Surender. S , Lokesh. P and Kalidas. K 2019. review
on area, production and productivity of coconut in India. International Journal of Research in Business Management. 7(1):1-6.

Rahman S, Adhikary SK, Sultana S, Yesmin S, Jahan N. (2013) In vitro evaluation of some selected fungicides against Pestalotia palmarum (Cooke.) Causal Agent of Grey Leaf Spot of Coconut. J Plant Pathol Microb 4: 197 doi:10.4172/2157-7471.1000197

Schmitz H. 1930. A suggested body metric method for food preservation industries and engineering chem. Analyst Ed 3:361-365.

Tuite. 1969 Plant Pathological method; Fungi and Bacteria. Burgess Publishing Company, Inneapolis, Minn, USA.

Vincent JH. 1947. Distortion of Fungal Hyphae in the Presence of Certain Inhibitors. Nature, 15: 850.

How to cite this article:

Vandana Chadar, R. R. Bhanwar, Rajendra Kashyap, Shraddha Karcho and Luchika Rana. 2020. In-vitro Evaluation of Some Plant Leaf Extract against Coconut Leaf Spot Caused by Pestalotia palmarum (Cooke) in Bastar Plateau of Chhattisgarh. Int.J.Curr.Microbiol.App.Sci. 9(04): 218-224. doi: https://doi.org/10.20546/ijcmas.2020.904.026