



# EFFECT OF LEAF EXTRACTS ON MYCELIAL GROWTH & SPORULATION OF *COLLETOTRICHUM GLOEOSPORIOIDES* (PENZ.) SACC.

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**Abstract:** *Colletotrichum gloeosporioides* is a fungus that causes anthracnose disease in soybeans, resulting in pod damage and reduced seed yield and quality. Chemical fungicides are commonly used to treat this disease, but it also results in the development of fungal resistance to the chemicals. Therefore, this study aims to *in vitro* evaluate the efficacy of 5 crude leaf extracts against *C. gloeosporioides*. The results showed that *Calotropis gigantea* leaf extracts were found to have very high antifungal activities. Crude extract of *Calotropis gigantea* leaves could effectively inhibit the growth of fungal mycelium (96.15%), followed by *Nicotiana tabacum* (90.38%) & *Tinospora cordifolia* (81.85%). In conclusion, crude extracts of *Calotropis gigantea* leaves were found to be highly effective in inhibiting both *C. gloeosporioides* mycelium growth and spore germination, and they have the potential as the new natural fungicides for the management of anthracnose disease.

**Index Terms - Plant Extract, Antifungal Activity, Medicinal Plants**

## I. INTRODUCTION

*Colletotrichum gloeosporioides* is a fungus that causes anthracnose disease in several plants, including mangoes, papayas, and soybeans. This disease is extremely harmful and can cause fruit plant deterioration and rotting, resulting in low production and poor fruit quality. Chemical fungicides are the most often used method for treating anthracnose disease, but they can induce fungal resistance. Furthermore, the continuous and inappropriate use of chemical fungicides to manage anthracnose disease is not considered a long-term solution because it increases investment costs, the risk of toxic residues, and concerns in human health and environmental settings. As a result of these factors, there have been various attempts to find alternate strategies to control anthracnose disease.

## II. MATERIAL AND METHODS

### 2.1 Isolation & Identification of Fungal Culture:

*Colletotrichum gloeosporioides* was isolated from the rhizosphere soil of Nashik by dilution method. The Soil sample weighed 1 g and was diluted in 10 ml of distilled water. One ml of diluted sample was poured and spread on sterilized PDA medium petri plates. To suppress bacterial development, a 1% Streptomycin sulfate solution was added to the medium before

pouring it into Petri plates. The inoculation plates were kept at room temperature (25°C) for 4 to 5 days. Colonies with distinct morphologies growing on PDA plates were counted individually. The fungal cultures were then transferred. After that, the fungal cultures were transferred, subcultured, and the pure cultures were kept on a PDA medium. Microscopically observed under a compound microscope after staining with Lacto phenol cotton blue. Aside from the hyphal structure, spore size, forms, spore-carrying structures, colony color, and morphology were studied. They were compared with the standard works of Manual of Soil Fungi (Gillman,1957) and Soil Fungi (Domsch et al., 1980).

## 2.2 Plant Materials & Extraction

Fresh plant leaves from *Azadirachta indica*, *Calotropis gigantea*, *Carica papaya*, *Nicotiana tabacum*, and *Tinospora cordifolia* were collected. The Plant extract was made by carefully washing it in distilled water and grinning it in distilled water. A Glucose nitrate medium was prepared in the flask. To this medium, the requisite quantity of plant extracts like *Azadirachta indica*, *Calotropis gigantea*, *Carica papaya*, *Nicotiana tabacum*, and *Tinospora cordifolia* was added. The medium was then autoclaved at 15 lb pressure for 20 minutes.

## 2.3 Effect of leaf extracts of Medicinal plants on mycelial growth and Sporulation of Selected Fungi

The Fungal toxicity of plant extract was studied by the poisoned food technique described by Nene and Thapliyal (1993). After cooling the medium, fungi were inoculated in an aseptic condition and incubated for 6 days at room temperature. Medium without leaf extract served as a control. Mycelial growth and sporulation of the test fungi were measured after harvesting.

Inhibition of mycelial growth was calculated by using the following formula

$$\text{Percentage Disease Inhibition (\%)} = \frac{D_c - D_t}{D_c} \times 100$$

D<sub>c</sub>: average weight of fungal colony (control),

D<sub>t</sub>: average weight of fungal colony with plant extract.

## III. RESULTS & DISCUSSION

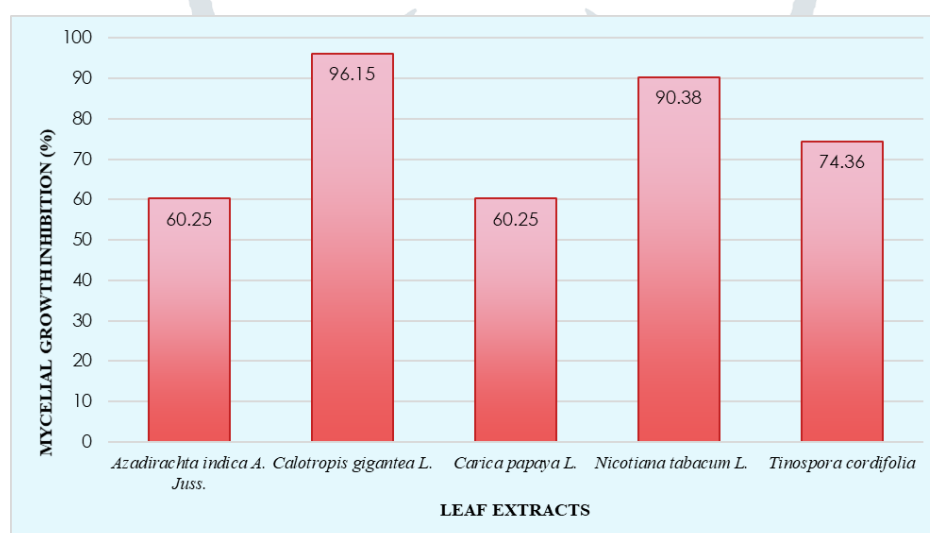
**Table No. 1: List of Plants**

Scientific Name	Family	Common Name
<i>Azadirachta indica</i> A. Juss.	Meliaceae	Neem
<i>Calotropis gigantea</i> L.	Apocynaceae	Crown Flower/ Giant Milkweed
<i>Carica papaya</i> L.	Caricaceae	Papaya Melon Tree
<i>Nicotiana tabacum</i> L.	Solanaceae	Tabacco
<i>Tinospora cordifolia</i>	Menispermaceae	Guduchi / Giloy

**Table No. 2 Effect of leaf extracts Medicinal plants on mycelial growth and Sporulation of *Colletotrichum gloeosporioides***

Plant Name	Degree of sporulation (Visual)	Mycelium dry weight (mg)	Mycelial growth inhibition (%)
<i>Azadirachta indica</i> A. Juss.	++	0.062	60.25
<i>Calotropis gigantea</i> L.	-	0.006	96.15
<i>Carica papaya</i> L.	++	0.062	60.25
<i>Nicotiana tabacum</i> L.	+	0.015	90.38
<i>Tinospora cordifolia</i>	++	0.040	74.36
Control	+++	0.156	

\*Sporulation:- Absent = - , Minimum = +, Moderate = ++, Maximum = +++



**Figure 1: The effect of plant extract on mycelial growth of *C. gloeosporioides***

Though all plant extracts exhibited different levels of antifungal activities against *C. gloeosporioides* mycelia shows less effectively prevent mycelia growth. Interestingly the *Calotropis gigantea* leaf extract exhibited the highest inhibition activities against *C. gloeosporioides* mycelial growth (96.15%) followed by *Nicotiana tabacum* (90.38%), *Tinospora cordifolia* (74.36%), *Azadirachta indica* & *Carica papaya* (60.25%).

Marigold (36.71%)

#### IV.CONCLUSION:

The effect of leaf extract of selected plants such as *Azadirachta indica*, *Calotropis gigantean*, *Carica papaya*, *Nicotiana tabacum*, and *Tinospora cordifolia* on mycelial growth and sporulation of *Colletotrichum gloeosporioides*, was tested and results were summarised in tables. Among selected plants *Calotropis gigantea*, and *Nicotiana tabacum* were found promising.

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## REFERENCES

- [1] Bhagavat, M.G. 2017 Studies on Diversity of soil micro-fungi from Nashik Tehsil, Maharashtra, India.' Ph.D. Thesis, Savitribai Phule Pune University, Pune.
- [2] Doran J. W. and Zeiss, M. R. 2000. Soil health and sustainability: Managing the biotic component of soil quality, *Appl. Soil Ecol.* 3–11.
- [3] Alexander, M. 1977. *Introduction to soil Microbiology*, John Wiley & Sons, New York,
- [4] Shen W, Lin X, Gao N, Zhang H, Yin R, ShiW, Duan Z, 2008. Land-use intensification affects soil microbial populations, functional diversity, and related suppressiveness of cucumber Fusarium wilt in China's Yangtze River Delta. *Plant Soil* 306(1–2):117–127. doi:10.1007/s11104-007-9472-5
- [5] Subba Rao, N.S. 2004 *Soil Microbiology*, Oxford & IBH Publishing Co. Pvt. Ltd. New Delhi,
- [6] G. P. Oyeyiola, 2016 Rhizosphere mycoflora of Okro (*Hibiscus esculentus*)' *Research journal of soil Biology.* 1 31-36
- [7] Maheshwari S.K., Singh D.V, Sahu A.K.1999. Effect of several nutrient media, pH & carbon sources on growth & sporulation of *Alternaria alternata*', *J. Mycopathol. Res.* 37 21-23.
- [8] Barnette, H. L. and Hunter, B. B.1972. *Illustrated genera of Imperfect Fungi*', Burgess Publishing Company, Minnesota, 241.
- [9] Gilman, J.C.200. *A manual of Soil fungi*' 2nd Indian edition, Biotech Books, Delhi, ().
- [10] Mukadam, D. S. et al.2006. *The illustrations of Fungi*', 1st edition, Saraswati Printing Press.
- [11] Paul, E. Nelson, et al 1927 *Fusarium species, An Illustrated Manual for Identification*', The Pennsylvania State University Press, University Park, and London ().
- [12] Stefanis, C., Alexopoulos, A., Voidarou, C., Vavias, S. and Bezirtzoglou, E. 2013. Principal methods for isolation and identification of soil microbial communities'. *Folia Microbiol. (Praha)*.58(1), 61-81.
- [13] Watanabe, Tsuneo.2002. *Pictorial Atlas of Soil and Seed Fungi, Morphologies of Cultured Fungi and Key to Species*', Second Edition, CRC Press, Boca Raon. 504.
- [14] Waksman S. A. 1922. A method of counting the number of fungi in the soil'. *J Bacteriol* 7 339-341.
- [15] Rangaswami, G., Seshadri, V.S. & Lucy Channamma, K.A., 1970. *Fungi of South India*'. UAS, Agricultural Res Service, W.Q. Judge Press, () 193.
- [16] Nene and Thapliyal, 'Fungicides in plant disease control', 2017 4th edition, MEDTEC 25-35.

