The Bioscan 9(1): 283-286, 2014 (Supplement on Plant Pathology)
AN INTERNATIONAL QUARTERLY JOURNAL OF LIFE SCIENCES
www.thebioscan.in

ANTIFUNGAL ACTIVITY OF PLANT EXTRACTS ON *COLLETOTRICHUM GLOEOSPORIOIDES* INFECTING *JATROPHA CURCAS*

YASHODA R. HEGDE*, TIPPESHI L. CHAVHAN AND RAJALAXMI S. KESHGOND
Department of Plant Pathology, University of Agricultural Sciences,
Dharwad- 580 005, Karnataka
e-mail: uasyashoda@rediffmail.com

KEYWORDS
Plant extracts
Antifungal activity
Colletotrichum gloeosporioides
Jatropha curcas

Received on :
19.10.2013

Accepted on :
25.02.2014

***Corresponding author**

ABSTRACT
Nine different plant extracts were evaluated against *Colletotrichum gloeosporioides*, *viz*. Glyricidia, neem, onion, eucalyptus, duranta, ocimum, garlic, ginger at two concentrations (5 and 10 per cent) and Azadirachtin (2.5 and 5.0 per cent). Azadirachtin was found to be the best in mycelial growth inhibition (89.25%) followed by ginger (48.05%) and onion (38.05%) whereas Azadirachtin and garlic bulb extract were best in complete inhibition of sporulation of *C. gloeosporioides*.

INTRODUCTION

Physic nut (*Jatropha curcas* L.) globally known as jatropha belonging to the family Euphorbiaceae is a large shrub or small tropical tree widely distributed in arid and semiarid areas. It is a multipurpose, stress resistant, zero-waste perennial and monoecious plant which is considered as a potential source of non-edible fuel-producing plant. (Ram *et al*., 2012)

Moreover, parts of the shrub are used in traditional medicine and as raw material for pharmaceutical and cosmetic industries (Paramathma *et al*., 2006). Anthracnose is one of the important diseases observed on *Jatropha curcas* incited by *Colletotrichum gloeosporioides* causing damages on leaves, stems and fruits and consequently a decrease in seed quantity and quality (Chavhan, 2007; Pinto *et al*., 2011). *Colletotrichum* is an important pathogen in different crops. (Pasuvaraji *et al*., 2013). The symptoms observed are small lesions on leaves which later lead to complete necrosis or blighting of leaves. Lesions can also be seen on the fruits leading to mummification of the fruits with pinkish discoloration and the seeds developed from such fruits are much smaller and shriveled compared to seeds from healthy fruits (Plate 1 and 2).

Kwon *et al*. (2012) first reported anthracnose disease on *Jatropha curcas* caused by *Colletotrichum gloeosporioides* in Korea. Santos *et al*. (2013) isolated and characterized *C. gloeosporioides* and *C. capsici* from physic nut seeds causing anthracnose from Brazil. No doubt in the past few decades chemical pesticides have protected the plants from diseases, their continuous and overuse have led to some serious ecological problems, *viz*. hazardous effects on beneficial organisms in soil, residual effects, pollution and resistant strain development in pathogen. Hence it is much better and safer to use the naturally available bioagents and the plant extracts with antifungal activity. The paper deals with management of *C. gloeosporioides* by using different plant extracts and its role on sporulation of *C. gloeosporioides*

MATERIALS AND METHODS

Pathogen was isolated on potato dextrose agar (PDA) by tissue isolation method and pure culture was obtained by following single spore isolation(Choi *et al*.,1999).The pathogen was identified as *Colletotrichum gloeosporioides* based on morphological characters. It produced single celled conidia in acervuli (Plate 3). Pathogenicity was proved by employing Koch's postulates.

The antifungal activity of eight plant extracts belonging to different families and one commercial product Multineem containing Azadirachtin on the mycelial growth and sporulation of *C. gloeosporioides* was studied *in vitro* by poisoned food technique (Nene and Thapliyal, 1982) at two concentrations, *i*. e. 5 and 10 per cent (Table 1). Fresh healthy leaves or bulbs were washed thoroughly with clean tap water and subsequently with sterile distilled water. Hundred gram of either leaves or bulbs were crushed in a pestle and mortar

Table 1: Effect of plant extracts on per cent inhibition of mycelial growth and sporulation of *Colletotrichum gloeosporioides*

Treatments	Per cent inhibition (%) Concentration 5 %	Sporulation	10 %	Sporulation	Mean
Glyricidia	13.33 (21.39)	+ +	32.77 (34.90)	+ +	23.05 (28.15)*
Neem	24.52 (29.49)	+ + +	34.62 (35.98)	+ + +	29.44 (32.73)
Onion	36.29 (37.01)	+	39.81 (39.10)	+	38.05 (38.06)
Eucalyptus	27.07 (31.32)	+ + +	38.70 (38.31)	+ + +	32.88 (34.81)
Duranta	23.16 (28.74)	+	41.07 (39.83)	+	32.11 (34.29)
Ocimum	30.18 (33.30)	+ + +	20.55 (26.93)	+ + +	25.36 (30.12)
Azadirachtin (2.5 & 5.0%)	88.51 (70.16)	-	89.99 (71.53)	-	89.25 (70.85)
Garlic	23.69 (28.87)	-	18.88 (25.62)	-	21.29 (27.24)
Ginger	39.99 (39.20)	+ +	56.10 (48.48)	+ +	48.05 (43.84)
Mean	30.75 (32.52)		36.64 (37.35)		
	Plant extract (P)	Concentrations (C)		PXC	
S.Em ±	0.99	0.44		1.40	
CD at 1%	2.43	1.08		3.42	

*Figures in the parentheses indicate arc sine transformed values; + + + : Heavy sporulation > 50 conidia per microscopic field; + + : Moderate sporulation 10-50 conidia per microscopic field; + : Scanty sporulation < 10 conidia per microscopic field; - : No sporulation

by adding 100 ml sterile distilled water. The resultant 100 per cent plant extract was filtered through double layered muslin cloth and then through sterilized Whatman No. 1 filter paper. From this standard stock solution, required concentrations were prepared by adding to PDA. It was thoroughly mixed and poured to sterilized Petriplates and allowed to solidify. Mycelial discs of 5mm diameter were cut from 8 days old culture of *C. gloeosporioides* with the help of sterilized cork borer and transferred aseptically to the centre of each Petri plate containing poisoned medium such that the mycelium of the fungus touched the medium. Medium devoid of plant extract served as control. Petri plates were incubated at 27 ± 1ºC in BOD incubator. Three replications were maintained for each treatment. The observations on colony diameter were recorded when control plate was completely covered with the test fungus. Per cent inhibition of mycelial growth of test fungus was calculated by using the formula given by Vincent (1947).

$$I = \frac{C - T}{C} \times 100$$

Where,

I: Percent inhibition

C: Radial growth in control

T: Radial growth in treatment

Plate 1: Malformed fruits of jatropha due to anthracnose

RESULTS AND DISCUSSION

The data presented in Table 1 reveals that among the plant extracts evaluated, azadirachtin showed maximum inhibition of mycelial growth (89.99%) at 5 percent and 2.5 per cent concentrations (88.51%) followed by ginger, 56.10 per cent (10%) and 39.99 per cent (5%). Next best was onion bulb extract at 10 per cent (39.81%) and 5 per cent (36.29%). Rest of the botanicals gave comparatively least growth inhibition (Plate 4a). Azadirachtin and garlic bulb extract completely inhibited the sporulation of *C. gloeosporioides*. These results are in agreement with those of earlier workers, *viz*. Shekhavat and Prasad (1971) and Mesta (1996). Garlic bulb extract inhibited mycelial growth of *C. gloeosporioides* to an extent of 60 per cent (Mukherjee *et al*., 2011) and complete mycelium

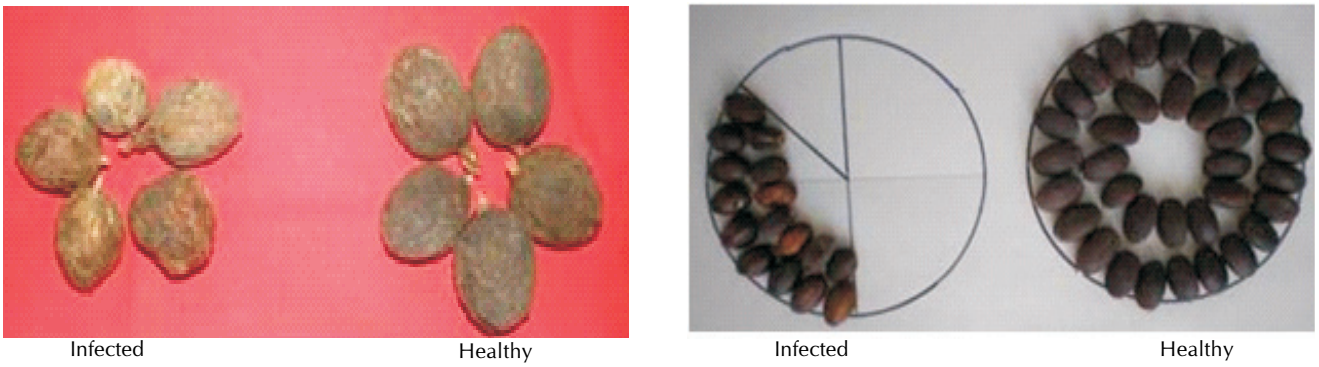


Plate 2: Healthy and infected fruits and seeds

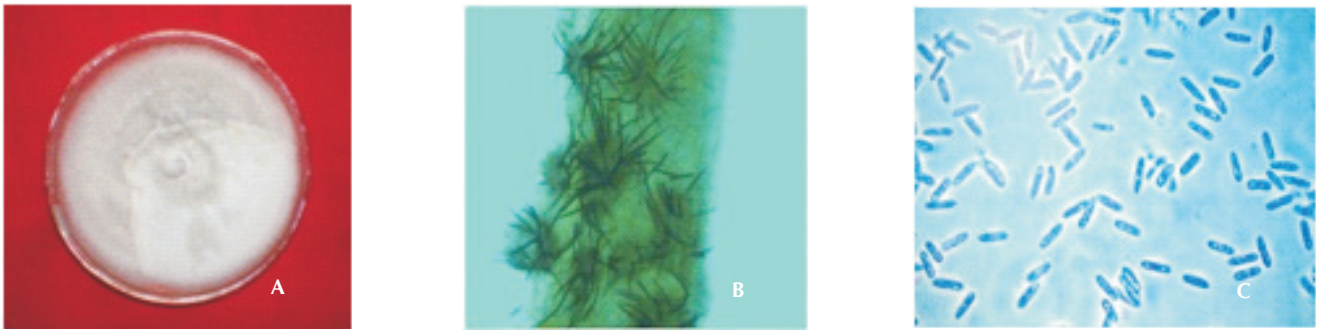
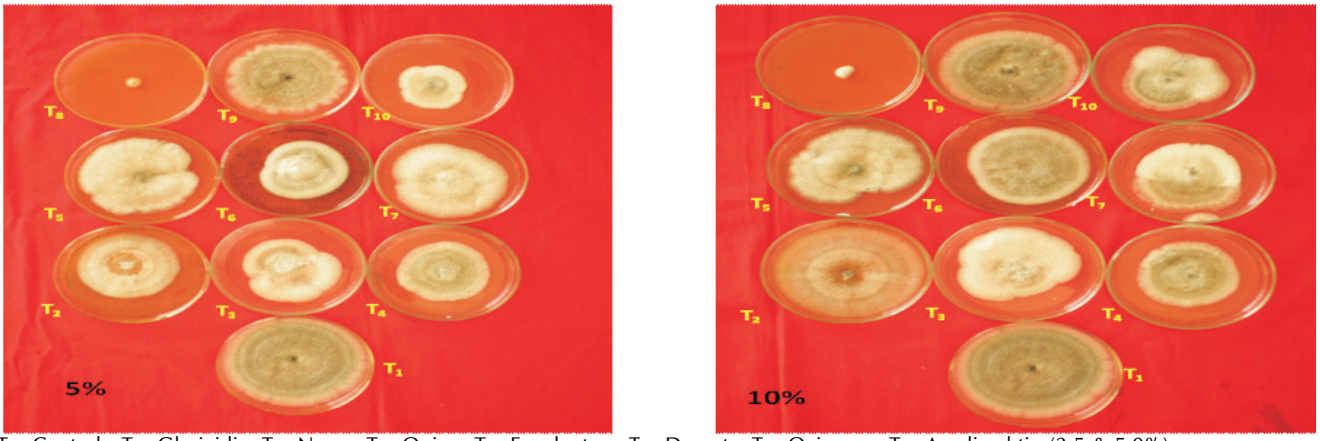
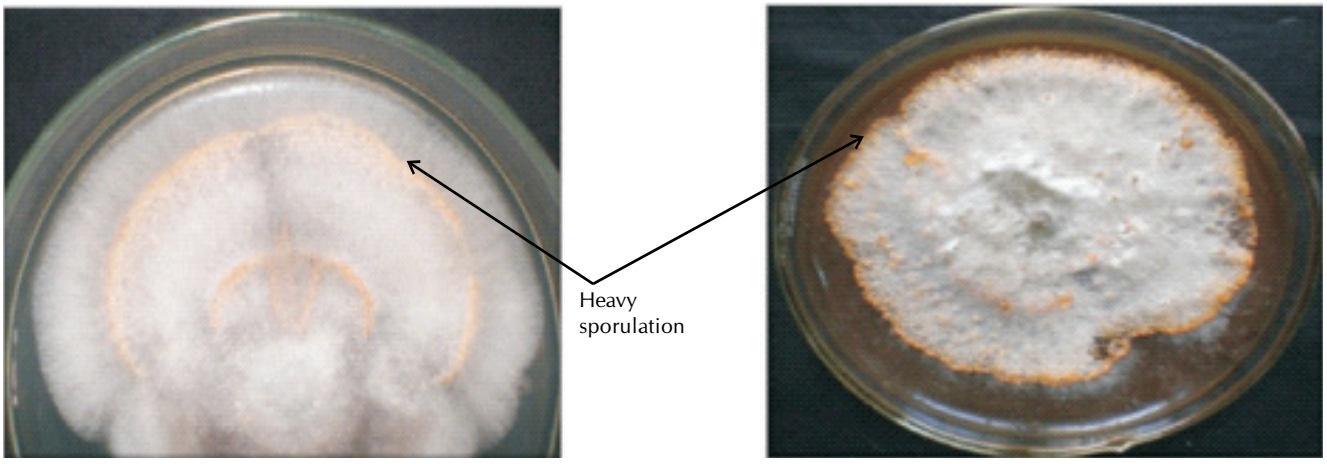


Plate 3: ***Colletotrichum gloeosporioides*****- A: pure culture; B: acervuli; C: conidia**



T_1 - Control, T_2 - Glyricidia, T_3 - Neem, T_4 - Onion, T_5 - Eucalyptus, T_6 - Duranta, T_7 - Ocimum, T_8 - Azadirachtin (2.5 & 5.0%), T_9 - Garlic, T_{10} - Ginger

Plate 4a: ***In vitro*** **evaluation of botanicals against** ***Colletotrichum gloeosporioides***



Neem leaf extract

Eucalyptus leaf extract

Plate 4b: Heavy sporulation induced by Neem and Eucalyptus leaf extracts

inhibition in *C. dematium* (Bhuiyan *et al.,* 2008). Ethanol extracts of *Allium cepa, Allium sativum*, *Azadirachta indica* and *Ocimum sanctum* showed fungitoxic properties against *C. capsici* (Ashashivapuri *et al.,* 1997). Jadav *et al.* (2008) reported that garlic bulb (10%) extract was effective in inhibiting the growth of *C. gloeosporioides*. Watve *et al.* (2009) reported that maximum inhibition was achieved due to neem leaf extract (78.15%) followed by garlic (58.89%) and tulsi (55.93%) and the least colony diameter was observed in glyricidia (25.93%) against jatropha leaf spot caused by *Colletotrichum gloeosporioides*. Ojha *et al.* (2008) recorded 94 per cent mycelial inhibition of *C. gloeosporioides* infecting *Saraca asoca* with garlic bulb extract.

Though neem, eucalyptus and ocimum extracts reduced the mycelial growth, they resulted in increased sporulation when compared to control (Plate 4b). This warrants us that such

plant extracts that encourage sporulation should not be used in plant disease management because they increase the disease incidence and severity through increased production of spores which subsequently spread to new areas and crops.

The present investigation revealed that azadirachtin was effective in inhibiting mycelial growth as well as sporulation and garlic bulb extract was effective in reducing sporulation of *C. gloeosporioides* infecting *Jatropha curcas*.

ACKNOWLEDGEMENT

We are grateful to National Oil seeds and Vegetables Oils Development board (NOVOD), Gurgaon, Haryana, for providing financial assistance during investigation.

REFERENCES

Ashashivapuri, O. P., Sharma. and Jhamana, S. L. 1997. Fungitoxic properties of plant extracts against pathogenic fungi. *J. Mycol. Pl. Pathol*. **27**: 29-31.

Bhuiyan, M. K. A., Shovan, L. R., Begum, J. A. and Pervez, Z. 2008. *In vitro* control of *Colletotrichum dematium* causing anthracnose of soybean by fungicides, plant extracts and *Trichoderma harzianum*. *International J. Sustainable Crop Production*. **3(3)**: 10-17.

Chavhan, T. L. 2007. Survey, characterization and management of diseases of *Jatropha curcas* L., M.Sc (Agri) Thesis, University of Agricultural Sciences, Dharwad.

Choi, Y. W., Hyde, K. D. and Ho,W. H. 1999. Single spore isolation of fungi. *Fungal Diversity*. **3:** 29-38.

Jadav, S. K., Diwakar, M. P., Sawant, U. K. and Kadam, J. J. 2008. Management of leaf spot disease of Kokum incited by *Colletotrichum gloeosporioides*. *J. Pl. Dis. Sci.* **3(2)**: 193-196.

Kwon, J., Choi, O., Kim, J. and Kwak, Y. 2012. First Report of Anthracnose Disease on *Jatropha curcas* Caused by *Colletotrichum gloeosporioides* in Korea. *J. Phytopathol*. **160(5)**: 255-257.

Mesta, R. K. 1996. Studies on fruit rot of chilli (*Capsicum annuum* L.) caused by *Colletotrichum capsici* (Sydow.) Butler and Bisby, M. Sc (Agri) Thesis, University of Agricultural Sciences, Dharwad. p. 65.

Mukherjee, S., Khandker, M. R., Islam. and Shahid, S. B. 2011. Efficacy of some plant extracts on the mycelial growth of *Colletotrichum gloeosporioides*. *J. Bangladesh Agril. Univ*. **9(1)**: 43-47.

Nene, V. L. and Thapliyal, P. N. 1982. Fungicides in plant disease control. *Oxford and IBH pub Co. Pvt. Ltd. New Delhi*. pp. 212-349.

Ojha, S., Chakraborty, M. R., Chakrabarti, J. and Chatterjee, N. C. 2008. Evaluation of different fungicides and biocides against *Colletotrichum gloeosporioides* causing Anthracnose of *Saraca asoca*. *Plant Archives*. **8(2)**: 733-736.

Paramathma, M., Venkatachalam, P., Sampathrajan, A., Vairavan, K., Sudhagar, J. R., Parthiban, K. T., Subramanian, P. and Kulanthaisamy, P. 2006. In: *Cultivation of Jatropha and biodiesel production"*. Sri Sakthi Promotional Litho Process. p. 40.

Pasuvaraji Adhipati, S. Nakkeeran and Chandrasekaran, A. 2013. Morphological characterization and molecular phylogeny of *Colletotrichum capsici* causing leaf spot disease of turmeric. *The Bioscan*. **8(1)**: 331-337.

Pinto, M. M., Silva, M. J. and Santos, M. R. 2011. Screening of *Jatropha curcas* genotypes to anthracnose caused by *Colletotrichum gloeosporioides*. *Commun. Agric. Appl. Biol. Sci.* **76(4):** 629-634.

Ram, G. D., Chauhan, S. S. and Verma, D. K. 2012. In vitro propagation of *Jatropha curcas* from embryo and nodal explants.*The Bioscan.* **7(2):** 251-254.

Santos, G. R., Junior, H. J. T., Sa, D. A. C., Furtado, G. Q. and Junior, N. S. M. 2013. Etiology and pathogenicity of two different isolates of *Colletotrichum* spp. obtained from physic nut seeds. *J. Seed Sci*. **35(2):** 139-146.

Shekhawat, P. S. and Prasad, R. 1971. Antifungal properties of some plant extracts and their inhibition of spore germination. *Indian Phytopath*. **24**: 800-802.

Vincent, J. M. 1947. Distortion of fungal hyphae in the presence of certain inhibitors. *Nature.* **159:** 850.

Watve, Y. G., Diwakar, M. P., Sawant, U. K. and Kadam, J. J. 2009. Studies on effect of different fungicides on *Colletotrichum gloeosporioides* Penz. causing leaf spot of jatropha. *J. Pl. Dis. Sci*. **4(1)**: 95-98.