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In vitro evaluation of chemical fungicides and bio agents against *Colletotrichum gloeosporioides* causing anthracnose disease in black pepper

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Abstract
An experiment was conducted at Department of Plant Pathology, College of Agriculture, OUAT, Bhubaneswar and High Altitude Research Station, Pottangi during 2017-18 to evaluate effectiveness of various chemical fungicides and bio agents in inhibiting the mycelia growth of *C. gloeosporioides* under *in vitro* conditions causing anthracnose disease in Black pepper crop by poisoned food technique and dual culture method, respectively. Among the fungicides maximum inhibition was obtained with the combination fungicides carbendazim + mancozeb (0.1%) which gave 97.26% growth inhibition over control. Among different bio-control agents *Trichoderma viridae* gave maximum growth inhibition 88.97%.

Keywords: Black pepper, *Colletotrichum gloeosporioides*, fungicides, biocontrol agents, poisoned food

Introduction
Black Pepper (*Piper nigrum* L.) is known as the "King of Spices" belongs to the family Piperaceae. India is one of the leading producers of black pepper in the world and Kerala contributes about 80-90% of total production in the country. Black pepper is affected by various diseases among which anthracnose caused by *Colletotrichum gloeosporioides*, is one of the important disease damaging the plant both in the nursery and the field. Though the disease may occur throughout the year but the maximum damage is caused during August to September with a range of 28% to 34% (Nair *et. al* 1987) [4]. The damage on the berries due to *C. gloeosporioides* has been reported up to 100% yield loss (Santha Kumari and Rajagopalan 2000) [10]. Keeping the view of the importance of the disease and severity of the damage caused by the pathogen, an experiment was conducted to evaluate effectiveness of various chemical fungicides and bio agents in inhibiting the mycelia growth of *C. gloeosporioides* under *in vitro* conditions.

Materials and Methods
The present experiment was conducted at Department of Plant Pathology, College of Agriculture, OUAT, Bhubaneswar and High Altitude Research Station, Pottangi during 2017-18 to evaluate effectiveness of various chemical fungicides and bio agents in inhibiting the mycelia growth of *Colletotrichum gloeosporioides* under *in vitro* conditions causing anthracnose disease in Black pepper crop by poisoned food technique and dual culture method, respectively.

a. Isolation and identification of causal agent.
The anthracnose infected leaves of Black pepper was collected and observed under microscope for the pathogen conformation. The infected leaves were first washed under running tap water and cut into small pieces of 2-3mm size. Then the bits were surface sterilized with 0.1% mercuric chloride ($HgCl_2$) followed by washing with sterile water for three times for the removal of excess chemicals. These bits were then transferred aseptically to sterile petriplates containing potato dextrose agar (PDA) media. The petriplates were incubated at room temperature for fungal growth. The culture was purified by transferring the growing tip of the fungal mycelium to PDA slant. The causal agent was identified based on morphological and cultural characteristics.
Three different chemical fungicides carbendazim (0.2%), mancozeb (0.2%) and mancozeb + carbendazim (0.1%) were evaluated to study their efficacies against *Colletotrichum gloeosporioides* causing anthracnose in black pepper by poisoned food technique.

100 ml of PDA was distributed in 250 ml of conical flasks for each treatment including control. The PDA was sterilized and recommended amount of fungicides was mixed with the PDA under aseptic conditions. PDA plate without fungicides served as control. Three replications were maintained for each treatment.

The test fungus was cut from periphery (5 mm diameter) by sterilized cork borer and aseptically inoculated at the centre of the petriplate. These plates were incubated in the incubator at 25 ±1 ^{0}C. The growth of the fungus was periodically observed and radial growth was taken 10 days after maximum growth was obtained at control. The growth inhibition percent was recorded in different poisoned media by using the following formula.

$$I = \frac{dc-dt}{dc} \times 100$$

Where, I= Inhibition percentage in surface growth of test fungus.
dc= Surface growth of test fungus in control.
dt= Surface growth of test fungus in treatment.

Similarly two different bio control agents *Trichoderma viridae* and *Pseudomonas fluorescense* were evaluated to study their efficacies against the same fungus. The experiment was conducted by following dual culture method. Pure culture of these above bio agents and test fungi were allowed to grow in petriplates. 5mm diameter of culture of each test fungi and bio agents were cut from periphery and placed in the side of sterilized petriplates. The PDA plate without any bioagents served as control. Three replications were made for each treatment. The growth of the test fungi and bio agents were periodically observed and radial growth of test fungus was measured on 7th day of their inoculation. The growth inhibition percent was recorded by using the following formula.

$$I = \frac{dc-dt}{dc} \times 100$$

Where, I= Inhibition percentage in surface growth of test fungus.
dc= Surface growth of test fungus in control.
dt= Surface growth of test fungus in treatment.

Results and Discussion

Statistical analysis of data revealed that there was a significant difference among the treatments in respect of their efficacy in inhibiting the mycelia growth of the test fungus. Recommended dose of the combination fungicide mancozeb + carbendazim (0.1%) was significantly better which gave 97.26% inhibition of mycelial growth of the test fungus over control followed by carbendazim (0.2%). Among bio control agents *Trichoderma viride* showed maximum inhibition of test fungus 88.97% followed by *Pseudomonas fluorescens* 67.84%. Among chemical fungicides mancozeb (0.2%) gave least result showing. 72.30% of mycelial inhibition. But in comparison with bio agents mancozeb has given better result over *Pseudomonas fluorescens*.

This was in confirmation with Pandey *et al*. (2012) [7, 8, 9] who reported that mancozeb + carbendazim at 100 ppm gave maximum mycelial growth inhibition for *C. gloeosporioides*. Similar results obtained by Patil *et al*. (2009) [6] is in confirmation with this. Ingle *et al*. (2014) reported that mancozeb + carbendazim (0.1%) had the highest curative effect against *Colletotrichum*. Regarding efficacy of bio agents similar results has been found by Devamma *et al*. (2012) [1] who conformed the efficacy of *T. viride* in mycelial growth inhibition of *C. gloeosporioides*.

Table 1: Effect of different chemical fungicides and bioagents on mycelial growth and mycelial growth inhibition of *Colletotrichum gloeosporioides*

Treatment	*Colletotrichum gloeosporioides* Mycelial growth (mm)	*Colletotrichum gloeosporioides* Mycelial growth inhibition (%)
Carbendazim (0.2%)	5	91.24
Mancozeb (0.2%)	17	72.30
Carbndazim + Mancozb (0.1%)	2	97.26
T. viride	7	88.97
P. fluorescens	19	67.84
Control	60	-
CD (0.05)		0.29
CV		0.18
SEm		0.09

* Value are mean of three replications.

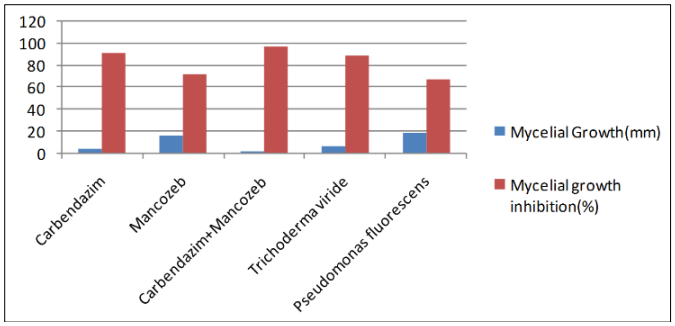

Fig 1: Effect of different chemical fungicides and bioagents on mycelial growth and mycelia growth inhibition of *Colletotrichum gloeosporioides*

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References

1. Devamma MN, Rajkumari JP, Devi PS. Fungicide compatible potential biocontrol agents against *Colletotrichum gloeosporioides* Penz. causing mango anthracnose. Current Biotica. 2012; 5(4):454-464.
2. Ingle YU, Patil CU, Thakur KD, Ingle K. Effect of fungicides and plant resistance activator on

Colletotrichum leaf spot of soybean. The Bioscan. 2014; 9(3):1187-1190.
3. Kurian PS, Josephrajkumar A, Backiyarani S, Sivakumar G, Shiva KN, Murugan M. Management of anthracnose disease (*Colletotrichum gloeosporioides* (Penz) Penz & Sac.) of black pepper (*Piper nigrum* L.) in the high ranges of Idukki District, Kerala. J Spices. Aromatic Crops. 2008; 17(1):21-23.
4. Nair PKU, Sasikumaran S, Pillai VS. Time of application of fungicides for control of anthracnose disease of pepper. Agric. Res. J Kerala. 1987; 25:136-139.
5. Patel DS. Chemical management of fruit spot of pomegranate caused by *Colletotrichum gloeosporioides* Penz. and Sacc. Indian Phytopathol. 2009; 62(2):252-253.
6. Patil CU, Zape AS, Wathore SD. Efficacy of Fungicides and Bioagents Against *Colletotrichum gloeosporioides* Causing Blight In *Piper longum*. Int. l Pl. Protec. 2009; 2(1):63-66.
7. Pandey A, Yadava LP, Manoharan M, Chauhan UK, Pandey BK. Effectiveness of cultural parameters on the growth and sporulation of *Colletotrichum gloeosporioides* causing anthracnose disease of mango (*Mangifera indica* L.). OJBS. 2012; 12(4):123-133.
8. Pandey A, Yadava LP, Mishra RK, Pandey BK, Muthukumar M, Chauhan UK. Studies on the incident and pathogenesis of *Colletotrichum gloeosporioides* Penz. Causes anthracnose of mango. I.J.S.N. 2012; 3(2):220-232.
9. Pandey A, Yadava LP, Manoharan M, Chuhan UK, Pandet BK. *In vitro* fungicides selectivity against *Colletotrichum gloeosporioides* (Penz.) isolates causing mango anthracnose. Pl. Sci. 2012; 2(3):72-85.
10. Santhakumari P, Rajagopalan B. Status of fungal foliar diseases of black pepper in Kerala. Contributory apers, Centennial Conference on Spices and Aromatic plants, 2000, 20-23.