Original Research Article

Evaluation of Fungicides against *Myrothecium roridum* Causing Leaf Spot of Soybean under *in vitro* Conditions

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**Abstract**

Introducation

Soybean (*Glycine max.* L. Merril) belonging to family Leguminaceae is designated as miracle bean established its potential as an industrially vital and viable oilseed crop in many areas of India. Leaf spot of soybean caused by *Myrothecium roridum* Tode ex. Fries is an important disease, which occurred in epidemic proportion entailing into colossal losses to soybean crop in Madhya Pradesh (Shrivastava and Khan, 1994, Singh and Shrivastava, 1994). Myrothecium leaf spot of soybean is occurring in almost all the major soybean growing areas of India causing about 30 per cent yield loss (Shrivastava and Khan 1994). The disease severity of myrothecium leaf spot soybean was in the range of 35 to 45 % and disease incidence of myrothecium leaf spot soybean was in the range of 30 to 55 % (Singh and Shrivastava, 1994). Myrothecium *roridum* is ordinary soil fungi, and survive in this environment as saprophytes in decaying plant tissues (Ellis, 1971). Initial symptoms of the disease appear as small round or oval, brown spots with dark brown margin on leaves in the infected plant. Since it is an economically important disease, management of the disease plays crucial role. Duval *et al.*, (2010) tested some fungicides *in vitro* against an isolate of *M. roridum*. Talukar *et al.*, (2013) tested seven fungicides at three concentrations i.e., 50 ppm, 100 ppm, of each treatment were used. In this study, *in vitro* evaluation of six fungicides at two concentrations (50 and 100 ppm) showed that Propiconazole, and Pyraclostrobin most effective in inhibiting the mycelia growth of *M. roridum*.

**Keywords**

Myrothecium leaf spot, Soybean, *Myrothecium roridum*, Fungicides.

**Article Info**

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Myrothecium leaf spot of soybean caused by *Myrothecium roridum*. Myrothecium leaf spot of soybean is occurring in almost all the major soybean growing areas of India causing about 30 per cent yield loss. Initial symptoms of the disease appear as small round or oval, brown spots with dark brown margin on leaves in the infected plant. Since it is an economically important disease, management of the disease plays crucial role. Poisoned food technique was employed for the evaluation of fungicides in the laboratory. Six fungicides viz. Dithan M45 (Mancozeb75%WP), Hexaconazole 5SC (Contaf), Propiconazole 25EC (Tilt), Tebuconazole 250EC (Folicur), Pyraclostrobin, Fluxapyroxid were evaluated against *M. roridum*. Two concentrations i.e., 50 ppm, 100 ppm, of each treatment were used. In this study, *in vitro* evaluation of six fungicides at two concentrations (50 and 100 ppm) showed that Propiconazole, and Pyraclostrobin most effective in inhibiting the mycelia growth of *M. roridum*. 

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concentrations (250, 500 and 1000 ppm) under in-vitro condition against M. roridum. There are numerous reports on fungicides in vitro condition to control the disease.

**Materials and Methods**

**Experimental site**

The laboratory experiment was carried out at Department of Plant Pathology, IGAU, Raipur (C.G.).

**Isolation of test fungus**

The fresh infected leaves of soybean plant samples were cut into small pieces, surface sterilized with 0.1% mercuric chloride (HgCl$_2$) solution followed by three washing with sterile distilled water and placing in moist chamber than after 1 to 2 days fungal mycelium growth were seen than finally small bits of fungus kept on the previously poured and solidified potato dextrose agar medium in Petri plates for isolation of the pathogen. The plates were incubated at 25°C in BOD incubator.

**In vitro evaluation of fungicides**

Poisoned food technique was employed for the evaluation of fungicides in the laboratory. Six fungicides viz. Dithan M45 (Mancozeb 75% WP), Hexaconazole 5SC (Contaf), Propiconazole 25EC (Tilt), Tebuconazole 250EC (Folicur), Pyraclostrobin, Fluxapyroxad were evaluated against M. roridum. Two concentrations i.e., 50 ppm, 100 ppm, of each treatment were used. The required quantity of fungicide was mixed with PDA at the time of pouring. Three replications were maintained for each fungicide for each of its concentration in Complete Randomized Design (CRD). The media was shaken well so as to enhance proper mixing of the fungicides. To avoid bacterial contamination a little amount of streptomycin was added in each flask before plating; five mm disc was cut with the help of sterilized cork borer from seven days old culture of the test fungus and was placed in the center of the medium in the reversed position to maintain continuous contact of the pathogen with poisoned medium. PDA plates without fungicide served as control. The radial growth of the colony was measured when the growth in control plates reached the rim of the Petri plates. Percent growth inhibition under the influence of different fungicides was calculated on the basis of the control. Observation was recorded at 10 days and 15 days after inoculation.

Percent inhibition of radial growth were calculated by the following

Inhibition % = C–T/C × 100

Whereas

C = Diameter of fungus colony (mm) in control plate,
T = Diameter of fungus colony (mm) in treated plate

**Results and Discussion**

**In vitro evaluation of fungicides**

Data in table 1 indicates that all fungicides were significantly superior in reducing the mycelial growth in comparison to control. In 50 ppm concentration percent growth inhibition of M. roridum recorded ranged between 100% to 62.33% in at 10DAI while it was 100% to 63.33% in 15 DAI. Complete inhibition of mycelial growth was recorded in the propiconazole and pyraclostrobin (100 %) at 10 DAI as well as 15 DAI. The minimum inhibition was recorded at 10 DAI in the Mancozeb (62.33%) followed by
Fluxapyroxad (64.25%), Hexaconazole (66.66%) and Tebuconazole (83.33%). The minimum inhibition was recorded at 15 DAI in the Mencozeb (63.33%) followed by Fluxapyroxad (66.11%), Hexaconazole (68.75%) and Tebuconazole (85.12%).

In 100 ppm concentration all fungicides were significantly superior in reducing the mycelial growth over control at 10 DAI and 15 DAI (Table 1; Plates 1 and 2). Percent inhibition in growth of *M. roridum* recorded in ranged 100% to 81.11% in 10 DAI and 100% to 73.70% in 15 DAI. Complete inhibition of mycelia growth was recorded in the Propiconazole and Pyraclostrobin (100%) at 10 DAI as well as 15 DAI (Figs. 1 and 2).

**Plate.1** Effect of different fungicides on radial growth of *Myrothecium roridum* (50ppm)
Plate.2 Effect of different fungicides on radial growth of *Myrothecium roridum* (100ppm)

| S. N. | Fungicides      | Radial growth (mm) | Radial growth (mm) | Radial growth (mm) | Radial growth (mm) |
|-------|-----------------|--------------------|--------------------|--------------------|--------------------|
|       |                 | 10 DAI**(50ppm)    | 15 DAI             | 10 DAI (100ppm)    | 15 DAI             |
|       |                 | G(mm)*             | I (%)              | G(mm)*             | I (%)              | G(mm)*             | I (%)              |
| 1     | Mencozeb        | 22.66              | 62.23              | 29.33              | 63.33              | 11.33              | 81.11              | 22.00              | 73.70              |
| 2     | Hexaconazole    | 20.00              | 66.66              | 25.00              | 68.75              | 10.00              | 83.33              | 20.00              | 76.09              |
| 3     | Propiconazole   | 0.00               | 100.00             | 0.000              | 100.00             | 0.00               | 100.00             | 0.00               | 100.00             |
| 4     | Tebuconazole    | 10.00              | 83.33              | 12.00              | 85.12              | 8.00               | 86.66              | 9.00               | 89.24              |
| 5     | Pyraclostrobin  | 0.00               | 100.00             | 0.00               | 100.00             | 0.00               | 100.00             | 0.00               | 100.00             |
| 6     | Fluxapyroxad    | 21.33              | 64.45              | 27.33              | 66.11              | 11.00              | 82.71              | 21.66              | 74.10              |
| 7     | Control         | 60.00              | 80.66              | 60.00              | 83.66              | 0.668              | 2.471              | 0.807              |
|       | CD (5%)         | 1.726              | 1.158              | 0.668              | 2.471              | 0.563              | 0.378              | 0.218              |

* Means of three replications ** Days after inoculation, G- Growth, I- Inhibition
Fig.1 Effect of different fungicides on radial growth of *Myrothecium roridum* (50ppm)

![Graph showing the effect of different fungicides on radial growth of Myrothecium roridum at 10 DAI and 15 DAI.]

The minimum inhibition was recorded at 10 DAI in the Mencozeb (81.11. %) followed by Fluxapyroxad (82.71%) Hexaconazole (83.33%) and Tebuconazole (86.66. %), the minimum inhibition was recorded at 15 DAI in the Mencozeb (73.70%) followed by Fluxapyroxad (74.10%), Hexaconazole (76.09%) and Tebuconazole (89.24%)

Similarly, Duval *et al.*, (2010) reported that the fungicide Tebuconazole (85.50%) highly effective in inhibiting the mycelial growth of *Myrothecium roridum*. Kale and Ukesh (2015) reported the fungicides Propiconazole and Hexaconazole as best in maximum

Fig.2 Effect of different fungicides on radial growth of *Myrothecium roridum* (100ppm)

![Graph showing the effect of different fungicides on radial growth of Myrothecium roridum at 10 DAI and 15 DAI.]

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inhibition against *M. roridum*. Daivasikamani *et al.*, (2013) reported the mycelia growth of *M. roridum* by Tebuconazole (90.10%) and Propiconazole (83.9%).

In conclusion, the present study in evaluation of fungicides against *Myrothecium roridum* causing leaf spot of soybean under in vitro conditions result showed Propiconazole, Pyraclostrobin and Tebuconazole were very effective in two (50 ppm and 100 ppm) concentrations in reducing mycelia growth of *M. roridu* under lab conditions.

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