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Efficacy of biocontrol agents with mancozeb against Cercospora leaf spot (*Mycosphaerella cruenta*) disease in Cowpea (*Vigna unguiculata*)

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Abstract

The present investigation was carried out with title Efficacy of biocontrol agents with mancozeb against Cercospora leaf spot (Mycosphaerella cruenta) disease in Cowpea (Vigna unguiculata) at Central Research Field of Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj during the Kharif season of 2022 to evaluate the efficacy of combination of Biocontrol agents with mancozeb on Cercospora leaf spot disease of cowpea. The experimental design used was Randomized Block Design replicated three times with 7 treatments which are mancozeb (1%), mancozeb (1%) + Bacillus subtilis (4%), mancozeb (1%) + Pseudomonas fluorescens (4%), mancozeb (1%) + Trichoderma harzianum (4%), mancozeb (1%) + Pseudomonas fluorescens (4%) + Bacillus subtilis (4%), mancozeb (1%) + Pseudomonas fluorescens (4%) + Trichoderma harzianum (4%), mancozeb (1%) + Bacillus subtilis (4%) + Trichoderma harzianum (4%) and control (untreated). The parameters assessed were plant height, number of pods per stand, pod length and disease intensity. The growth parameters were highest in cowpea treated with the biocontrol agents with fungicides particularly, mancozeb (1%) + Pseudomonas fluorescens (4%) + Trichoderma harzianum (4%) compared to other treatments of cowpea. Disease intensity were found to be significantly (P>0.05) lower in cowpea treated with the biocontrol agents with fungicides particularly, mancozeb (1%) + Pseudomonas fluorescens (4%) + Trichoderma harzianum (4%) and higher in treatment control (untreated) cowpea.

Keywords: Cowpea, biocontrol agents, mancozeb, cercospora leaf spot, disease intensity

Introduction

Cowpea (*Vigna unguiculata* L. Walp.) (2n = 2x = 22) is a member of the Phaseoleae tribe of the Leguminosae family.

Cowpea is an indigenous African legume crop belonging to the Fabaceae/Leguminosae family (Wiersema & León, 1999)^[12]. Cowpea is widely cultivated and adapted crop is of great importance in the tropical and subtropical countries of Asia, Oceania, the Middle East, southern Europe, Africa, southern United States of America and Central and South America (Brader, 2002)^[13].

The leaves, pods and seeds provide a good source of protein, vitamins and carbohydrates. The seed particularly contains on average 23-25% protein and 50-67% starch (Quin, 1997)^[14]. The seed provides an important source of nourishment, especially protein, for relatively poor people who cannot afford milk and meat products (Brader, 2002)^[13].

Cowpea (*Vigna unguiculata* (L.) Walp) is known to be affected by several fungal, viral and bacterial diseases. One of such fungal diseases reported to cause serious yield loss in Cowpea is the Cercospora leaf spot caused by two Fungi namely *Cercospora canescens Ellis and Martin* and *Mycosphaerella cruenta Lanthanus* (Akande, 2007)^[1].

The cultivation of cowpea is affected by a number of seed borne, soil borne and wind borne/foliar fungal diseases. Among the foliar diseases, affecting the crop, Cercospora leaf spots, powdery mildew and rust are of major concern. The Cercospora leaf spots appear as spots varying in colour and shape on the leaves and as lesions of stem and pods. In India, Cercospora leaf spot was first reported in Delhi, (Munjal *et al.*, 1960)^[15].

Two species of Cercospora have been reported to induce leaf spots in cowpea: *Cercospora canescens and Pseudocercospora cruenta*. In either case, air borne spores produced on the underside of the leaf help in transmission. The fungus is carried over from one season to the next in left-over planting material.

The fungus has a wide host range, attacking other legumes such as *Glycine max* (soybean), *Amaranthus tricolor* (grain amaranth), *Lablab purpureus* (hyacinth bean), *Mucuna pruriens* (buffalo bean) and the minor host include *Crotalaria juncea* (sunhemp), *Lycopersicon esculentum* (tomato) *Psophocarpus tetragonolobus*, (winged bean), *Vigna angularis* (adzuki bean), *Vigna mungo* (black gram) *and Vigna radiata* (mung bean). These alternative hosts extend the reservoir of plants which can cany over infection to the next growing season.

Cercospora leaf spot is a serious limitation to cowpea production, resulting in yield loss of as much as 42%. There is the need to reduce the yield losses from crop disease since it is possible that the cultural practices including use of resistant varieties may fail to provide the desired level of disease control or in the case of disease epidemic outbreak, the last option is use of chemical (fungicides) control. Therefore, the use of different fungicides to test their efficacy on disease is a step in the right direction. The chemicals are applied singly or in combination to determine the most effective method in the control of Cercospora leaf spot.

Materials and Methods

The present study entitled "Efficacy of biocontrol agents with mancozeb against Cercospora leaf spot (*Mycosphaerella cruenta*) disease in Cowpea (*Vigna unguiculata*)" was conducted at Central Research Field of Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj during the Kharif season of 2022. The selected field was uniform, cultivable with black soil having good drainage. The extremes of both summer and winter are experienced here. The maximum temperature was recorded during summer up to 47 °C and the minimum temperature was recorded during winter up to 2.5 °C.

Application of treatment

The selected chemical fungicide mancozeb was sprayed to the respective plots as foliar spray application. The subsequent spray was given at 15 days interval. The selected Bio control agents were applied as seed treatment.

Observations

Plant height, No. of pods, pod length and No. of infected leaves were recorded from 5 randomly selected and tagged plants at a interval of 30, 60, 75 and 90 DAS. The significance and non-significance of the treatments were judged with the help of 'F' (Variance ratio) test the significant differences between the means were tested with the critical differences at 5% probability.

Symptoms

The symptoms of Cercospora leaf spot of cowpea caused by *C. canescens* as round orange to light brown, silvery grey spots of 8-15 mm diameter. On the petiole, symptom appeared as uniform light brown spindle shaped lesion. Symptoms of *C. cruenta* appeared as brown colored spots with circular to irregular dark margin which later became necrotic, grey colored center with a brown margin. Cercospora leaf spot on faba beans mainly affects leaves, but also affect stems and pods.

Disease intensity

Disease intensity (%) to be recorded at 60, 75 and 90 days after sowing. It is calculated by using the following formula:

Disease intensity (%) = $\frac{\text{Sum of all disease rating}}{\text{Total no. of rating } \times \text{Maximum disease grade}} \times 100$

| 9 | | Plan | t height | (cm) | No. of pods per plant | | | Pod length (cm) | | | Disease intensity (%) | | |
|-----------------------|---------------------------------------------------------------------------------|---------------------|----------------------|---------------------|-----------------------|---------------------|---------------------|---------------------|---------------------|---------------------|-----------------------|----------------------|---------------------|
| Treatments | Treatment Details | 30 DAS | 60 DAS | 90 DAS | 60 DAS | 75 DAS | 90 DAS | 60 DAS | 75 DAS | 90 DAS | 60 DAS | 75 DAS | 90 DAS |
| T ₀ | Control | 11.4 ^f | 36.333 ^f | 42 ^g | 5.867 ^f | 6.533 ^f | 8.533 ^f | 9.833 ^f | 13.767 ^g | 18.267 ^g | 27.67 ^a | 36.957 ^a | 46.943ª |
| T_1 | Mancozeb (1%) | 32.333 ^b | 46.933 ^{bc} | 52.466° | 10 ^b | 11 ^b | 12.333° | 18.167 ^b | 23.333 ^t | 29.100 ^b | 18.993 ^d | 22.487 ^d | 26.357 ^e |
| T_2 | Mancozeb (1%) + Bacillus subtilis (4%) | 16.533e | 39.2 ^e | 44.533 ^f | 7 ^e | 7.533 ^e | 9.533 ^e | 12.533e | 17.667ª | 22.433° | 24.217 ^{bc} | 30.853 ^{bc} | 39.707° |
| T ₃ | Mancozeb (1%) + Pseudomonas fluorescens (4%) | 33.1 ^b | 48.133 ^b | 55.233 ^b | 10.733 ^b | 11.667 ^b | 15.333 ^b | 19.067 ^b | 24.267 ^t | 29.267 ^ь | 18.31 ^d | 21.037 ^d | 23.217 ^f |
| T_4 | Mancozeb (1%) + <i>Trichoderma</i> <i>harzianum</i> (4%) | 23.4° | 45.466° | 50.5 ^d | 8.867° | 10.133° | 11.067 ^d | 16.633° | 22.067° | 27.233° | 25.207 ^b | 30.163° | 37.607 ^d |
| T_5 | Mancozeb (1%) + Pseudomonas fluorescens (2%) + Bacillus subtilis (2%) | 22.433° | 43.4 ^d | 48.166 ^e | 7.867 ^d | 8.467 ^d | 10.6 ^d | 14.733 ^d | 19.733 ^d | 24.600 ^d | 22.88 ^c | 28.857° | 35.977 ^d |
| T_6 | Mancozeb (1%) + Pseudomonas fluorescens (2%) + Trichoderma harzianum (2%) | 35.666ª | 52.266ª | 60.833ª | 12.167ª | 12.933ª | 16.467ª | 20.567ª | 25.967ª | 30.667ª | 15.93 ^e | 17.51° | 19.117 ^g |
| T 7 | Mancozeb (1%) + Bacillus subtilis (2%) + Trichoderma harzianum (2%) | 19.333 ^d | 41.766 ^d | 47.833° | 6.933 ^e | 7.267° | 9.333 ^e | 11.900 ^e | 16.000 ^f | 20.767 ^f | 24.73 ^{bc} | 32.58 ^b | 41.537 ^b |
| CD (p=0.05) | | 1.75 | 1.85 | 1.45 | 0.76 | 0.73 | 0.79 | 1.44 | 1.13 | 1.24 | 1.93 | 2 | 1.82 |
| S.EM | | 0.58 | 0.61 | 0.48 | 0.25 | 0.24 | 0.26 | 0.47 | 0.37 | 0.41 | 0.64 | 0.66 | 0.6 |

 Table 1: Effect of biocontrol agents with mancozeb on plant height, no. of pods, pod length and disease intensity of cowpea at 60, 75 and 90 DAS.

Note: Values in the same column followed with similar alphabet are non-significant to each other at (p=0.05).

Results and Discussion

The present investigation was carried out with title "Efficacy of biocontrol agents with mancozeb against Cercospora leaf spot (*Mycosphaerella cruenta*) disease in Cowpea (*Vigna* *unguiculata*)" at Crop Research Field (CRF), Department of Plant Pathology, Naini Agricultural Institute, Sam Higginbottom University of Agriculture, Technology and Sciences (SHUATS), Prayagraj, Uttar Pradesh during the *Kharif* - 2022. The salient results of the study are summarized below:

Field samples of Cercospora leaf spot incidence in vegetable cowpea were collected from Crop Research Field (CRF). Studies on the symptomology and Etiology revealed that symptoms were observed on the leaves, pods and stem of cowpea. Symptoms of *Mycosphaerella cruenta* appeared initial as small light greenish yellow diffused spots on the adaxial surface of the leaf turned reddish brown with greenish yellow hallow and black mass of conidia on the upper and lower surfaces. At the final stage, the leaves became necrotic, dried up and hung down. On the stem, symptom appeared as black coloured lesions with mass of conidia and in the pods, irregular black spots appeared with powdery mass of fungal growth. Symptoms of *Mycosphaerella cruenta* on leaves appeared as rough, diffused and irregular, dark red spots with silver grey centre and a darker margin.

The data collected on different aspects were tabulated & analysed statistically using the methods of analysis of variance & critical difference. The significant & non-significant differences observed have been analysed critically within & between the treatment combinations.

The results of efficacy of bioagents with Mancozeb on growth parameters and disease intensity of Cercospora leaf spot of cowpea are summarized below:

- The plant height at 30, 60 and 90 DAS was recorded 35.66 cm, 52.26 cm and 60.83 cm respectively in T₆ [Mancozeb (1%) + *Pseudomonas fluorescens* (2%) + *Trichoderma harzianum* (2%)] which was found to be statistically significant compared to other treatments including T₀ (control).
- The number of pods at 60, 75 and 90 DAS were 12.16, 12.93 and 16.467 respectively in T₆ [Mancozeb (1%) + *Pseudomonas fluorescens* (2%) + *Trichoderma harzianum* (2%)] which was found to be statistically significant compared to other treatments including T₀ (control).
- Pod length at 60, 75 and 90 DAS was 20.56 cm, 25.96 and 30.67 cm respectively in T₆ [Mancozeb (1%) + *Pseudomonas fluorescens* (2%) + *Trichoderma harzianum* (2%)] which was found to be statistically significant compared to other treatments including T₀ (control).
- The disease intensity at 60, 75 and 90 DAS recorded was 15.93%, 17.51% and 19.117% in T₆ [Mancozeb (1%) + *Pseudomonas fluorescens* (2%) + *Trichoderma harzianum* (2%)] which was found to be statistically significant compared to other treatments including T₀ (control).

Conclusion

From this present study entitled "Efficacy of Biocontrol agents with mancozeb against Cercospora leaf spot (*Mycosphaerella cruenta*) disease in Cowpea (*Vigna unguiculata*)" it is concluded that T_6 was found to be significant. The application of these combined biocontrol agents with chemical fungicide particularly mancozeb (1%) (Foliar spray) + *Pseudomonas fluorescens* (4%) (Seed treatment) + *Trichoderma harzianum* (4%) (Seed treatment) was found to be effective on growth parameters such as increased plant height, more number of pods, increased pod length and reduced disease intensity caused by *Mycosphaerella cruenta*, while control was found to be less

effective on growth parameters and increased disease intensity.

Based on analysis, one of the combinations of biocontrol agents with chemical fungicide *viz*. T_6 – mancozeb (1%) + *Pseudomonas fluorescens* (4%) + *Trichoderma harzianum* (4%) is recommended to control the Cercospora leaf spot disease in cowpea.

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