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Efficacy of different bioagents seed treatment for controlling seed-borne *Colletotricum* sp. in chilli (*Capsicum annum* L.)

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Abstract
In tropical and subtropical areas, *Colletotrichum capsici*, which causes chilli anthracnose, is a significant barrier to the production of chillies. Study was aimed mainly to find the efficacy of various bioagents on the germination, seedling length and seedling vigour of *C. capsici* infected chilli seeds. Rudra and Byadagi are the two agronomically superior varieties selected for the study. Among all the treatments *Trichoderma viride* were found superior and significantly higher seed germination, seedling length, seed vigour index as compared to other treatments and control. Among the various bioagents, In the variety Rudra the highest germination percentage of 97.66%, The root length, shoot length, seedling length of T_1 were 6.4 cm, 5.7 cm, 12.19 cm and seed vigour index of 1184.49 were obtained in T_1 (2.5 ml of *Trichoderma viride*). In the variety Byadagi highest germination percentage of 95.33% , The root length, shoot length, seedling length were 6.87 cm, 5.6 m, 12.47 cm, The seed vigour index of 1196.79 were obtained in T_2 (5 ml of *Trichoderma viride*). Among the bioagents *Trichoderma viride*, *Pseudomonas fluorescens* performs well and antagonists against the pathogen.

Keywords: Bioagents seed treatment, controlling seed-borne *Colletotricum* sp., chilli, *Capsicum annum* L.

Introduction
The chilli plant, *Capsicum annum* L., is mostly grown for its edible green fruits and for the dried chilli that is used as a commercial spice. It contains a lot of vitamin C, A, and B. Most tropical and subtropical nations, including those in India, Japan, Mexico, Turkey, the United States of America, and Africa, cultivate chillies. It offers a number of therapeutic benefits and is also effective for treating atomic dyspepsia, lumbago, neuralgia, and rheumatic illnesses (Pruthi, 1993) [12]. Sweet bell peppers, paprika, pimento, and other red pepper-based items all include *Capsicum annuum* L. The flavour of capsicum is what makes it stand out, as it is a fantastic source of vitamins A and C in addition to its pungency and therapeutic benefits. Its production and productivity have been impeded by a number of viral, bacterial, and fungal diseases that are susceptible to it. Early symptoms of the disease include a loss in photosynthetic area, while later symptoms include infestation of reproductive organs and commercially valuable produce. The illness reduces the plant's ability to photosynthesize. One of the main production bottlenecks in tropical and subtropical regions is *Colletotrichum capsici*, which causes a variety of disease symptoms such damping off, anthracnose or fruit rot, dieback in chilli. According to Pakdeevaraporn *et al.* (2005) [11], this fungus severely damages chilli fruits both before and after harvest, accounting for more than half of crop losses. As the fungus causes necrosis of tender twigs from the tip backwards the disease is called die-back, when the crop is in flowering stage the infection typically starts. Flowers wither and wilt. Flowers are being shed in great abundance. The flower stalk withers and becomes dry. This withering spreads from the flower stalks to the stem, which results in the dieback of the branches and stem and the withering of the branches. Partially afflicted plants produce few, poor-quality fruits. On such dark lesions, Colletotrichum will develop a lot of fruiting bodies and spores. Pre-emergent and post emergent damping-off of seedlings in nurseries and transplanted fields is caused by the sowing of such infected seeds.

Farmers usually use fungicides to control this disease one of the alternative to overcome these problems is to replace the use of the chemical approach with the search for alternative control methods. Chemicals have been used for many years to manage the chilli anthracnose fruit rot, which has led to several unfavourable issues. Alternative control measures that are efficient at the farm level must be incorporated. The use of fungicides and locally accessible bio agents to control this seed-borne infection at the farm level has not been the subject of any systematic research. In order to determine the effectiveness of different bioagents on the germination of chilli seeds contaminated with *C. capsici*, the current investigation was carried out.

Materials and Methods

The experiment was conducted during May 2022 in Department of seed science and technology, College of Horticulture, Bagalkot, Karnataka. The two agronomically superior varieties Rudra, and Byadagi were selected for the study in completely randomized design for 3 replications. All the observations were taken 10 days after sowing

Isolation and Pathogenicity test of the pathogen

C. capsici, a causal agent of anthracnose was isolated from two hundred anthracnose infected chilli seeds obtained; from Department of Plant pathology, College of Horticulture, Bagalkot, Karnataka. Anthracnose fungus was isolated by Blotter Method and Agar Plate Method described by ISTA (1976). After the pathogenecity test, isolates of *C. capsici* were cultured on PDA for 3 days. Then a most pathogenic isolate was used for further study.

Sources of bioagents and fungicides

The liquid form of three bioagents namely *Trichoderma viride*, *Pseudomonas fluorescens*, *Bacillus subtilis* at the rate of 2.5 ml and 5 ml were used to treat the seeds of two varieties Rudra and byadagi (Table 1) For studied the efficacy of bioagents as seed protectants against *C. capsici*, apparently healthy sterilized seeds of chilli were artificially contaminated with isolated pathogenic species of *C. capsici*. Hundred seeds per petri plate of such contaminated seeds were then treated with bioagents at the rate of 2.5 ml and 5 ml for 30 minutes in above solutions. The treated seeds were fine shade dried before sowing. The treated and untreated inoculated seeds were then placed in the petri plate under ambient conditions. Observations were recorded after 10 days on seed germination, root/shoot length and seed vigour index were noted.

Statistical analysis

Seedling vigour was also calculated by formula suggested by Abdul Bakshi and Anderson (1973) [1].

Vigour index = Germination (%) x (Root length + shoot length)

All other data were analyse using the computer package program DSAASTAT.

Table 1: (Treatments details for Rudra and Byadagi)

T_1	2.5 ml of *Trichoderma viride*,
T_2	5 ml of *Trichoderma viride*,
T_3	2.5 ml of *Bacillus subtilis*
T_4	5 ml *Bacillus subtilis*
T_5	2.5 ml of *Pseudomonas fluorescens*
T_6	5 ml of *Pseudomonas fluorescens*
T_7	Control

Results and Discussion

Table 2: Germination percentage, Shoot length, Seedling length and Seed vigour index of variety Rudra

Treatments	Germination %	Shoot length (cm)	Root length (cm)	Seedling length (cm)	Seed vigour index
T_1	97.66	6.44	5.7	12.19	1184.49
T_2	94.66	6.05	4.84	10.06	959.64
T_3	96	4.81	3.87	8.89	846.85
T_4	97	5.63	2.77	8.23	785.76
T_5	95.33	4.85	3.26	9.71	937.77
T_6	96.66	6.27	5.11	11.18	1023.98
T_7	77.65	2.10	1.47	3.49	238.44
C.D (5%)	3.41	1.11	1.76	0.74	32.95
C.D (1%)	4.74	1.51	2.44	1.03	45.74

Table 3: Germination percentage, Shoot length, Seedling length and Seed vigour index of variety Byadagi

Treatments	Germination %	Shoot length (cm)	Root length (cm)	Seedling length (cm)	Seed vigour index
T_1	91.07	5.33	4.63	10.12	934.38
T_2	95.33	6.87	5.6	12.47	1196.79
T_3	87.33	6.43	5.27	11.7	1009.25
T_4	90.66	6.02	4.91	10.84	974.55
T_5	93	6.11	4.01	10.11	928.51
T_6	91.92	6.61	4.87	11.38	1019.713
T_7	79.11	1.83	1.36	4.12	315.11
C.D (5%)	2.83	0.88	0.89	0.65	38.96
C.D (1%)	3.93	1.21	1.24	0.91	54.08

Based on the present investigation It became clear during the experiment that all seed treatments (bioagents) increased seed germination and seedling vigour in comparison to the control. According to the results, In the variety Rudra the highest germination percentage 97.66 was obtained in T_1 (2.5 ml of *Trichoderma viride*) Trichoderma species have been applied to control Colletotrichum species in chilli (Boonratkwang *et al*., 2007) [2] followed by T_6 (5 ml of *Pseudomonas fluorescens*) with 96.66%. *P. fluorescens* significantly decreased the seedling mortality (Padder *et al*., 2010) [10] it is prominently higher when compared to control. The root length, shoot length, seedling length of T_1 were 6.4 cm, 5.7 cm, 12.19 cm respectively. Antagonism with Trichoderma species against an array of phytopathogens has been reported (Kaur *et al*., 2006) [8] and the root length, shoot length, seedling length of T_6 were 6.27 cm, 5.11 cm, 11.18 cm respectively. The seed vigour index of T_1 and T_6 were 1184.49 and 1023.98 respectively. (Table 2)

Based on the results obtained in the variety Byadagi the highest germination percentage 95.33 were obtained in T_2 (5 ml of *Trichoderma viride*), followed by T_6 (5 ml of *Pseudomonas fluorescens*) with 91.92%. Pseudomonas fluorescens was found better in increasing the germination percentage, dry weight, leaf area and chlorophyll content (Dutta *et al*., 2005) [4] it is prominently higher when compared to control. The root length, shoot length, seedling length of T_2 were 6.87 cm, 5.6 m, 12.47 cm respectively. Volatile and non-volatile compounds produced by *T. viride* and mycoparasitism might be responsible for the suppression of the pathogen (El-katalny el., 2001; Eziashi *et al*., 2007; Ghildiyal *et al*., 2008) [5-7]. The root length, shoot length, seedling length of T_6 were 6.61 cm, 54.87 cm, 11.38 cm respectively. *P. fluorescens* as observation in the present study might have tolerated or escaped from root infection facilitating

an active absorption of nutrients thereby promoting the plant growth and health (Muthukumar *et al*., 2010) [9] The seed vigour index of T_2 and T_6 were 1196.79 and 1019.71 respectively (Table 3.). The results of treating chilli seeds that had been intentionally contaminated with isolated pathogenic species of *C. capsici* with various bioagents showed that all of the bioagents and antagonists had a significant impact on the percentage of germination, the length of the seedlings, and the vigour of the seedlings.

Conclusion

In conclusion, this investigation unequivocally demonstrates the efficacy of using bioagents, particularly *Trichoderma viride* and *Pseudomonas fluorescens*, in enhancing seed germination, seedling vigour, and overall plant health in chilli varieties Rudra and Byadagi. The application of these bioagents significantly outperformed the control in terms of germination percentage, seedling growth metrics, and seed vigour index. Specifically, *Trichoderma viride* showed remarkable results in promoting seed germination and growth, with the highest germination percentages recorded in both chilli varieties. *Pseudomonas fluorescens* also played a crucial role in reducing seedling mortality and enhancing plant growth, indicating its potential as a biocontrol agent against phytopathogens. The findings underscore the importance of integrating bioagents into agricultural practices to combat pathogens and promote sustainable crop production. This study not only reaffirms the antagonistic properties of *Trichoderma* species against various phytopathogens but also highlights the beneficial effects of *Pseudomonas fluorescens* in supporting plant health, thereby offering a viable and eco-friendly alternative to chemical treatments in agriculture.

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