www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; 11(1): 410-412

www.thepharmajournal.com
Received: 07-11-2021
Accepted: 29-12-2021

GV Bhosale
Department of Plant Pathology, College of Agriculture, Parbhani Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani, Maharashtra, India

DS Bharti
Department of Plant Pathology, College of Agriculture, Parbhani Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani, Maharashtra, India

GB Bhosale
Department of Extension education, College of Agriculture, VNMKV, Parbhani, Maharashtra, India

GP Jagtap
Department of Plant Pathology, College of Agriculture, Parbhani Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani, Maharashtra, India

SN Banne
Department of Plant Pathology, College of Agriculture, Parbhani Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani, Maharashtra, India

Corresponding Author:
GV Bhosale
Department of Plant Pathology, College of Agriculture, Parbhani Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani, Maharashtra, India

In vitro evaluation of bio control agents against *Claviceps fusiformis*, causing Ergot of Bajra

GV Bhosale, DS Bharti, GB Bhosale, GP Jagtap and SN Banne

Abstract
Biotic and abiotic stresses are major constraints in the production of Pearl millet. Among biotic stresses apart from bacterial and viral diseases, many fungal diseases are of economic importance. Ergot was the most important and destructive disease of Pearl millet. Ergot caused by *Claviceps fusiformis* a serious threat to successful cultivation of Pearl millet. For the management of ergot of Bajra, an experiment was conducted to study the efficacy of antagonistic organism against *Claviceps fusiformis*. The bio-agents i.e. *Trichoderma viride, T harzianum, T hamatum, Aspergillus niger, T. longibrachiatum, T. koningii, Pseudomonas fluorescens* and *Bacillus subtilis* etc. were evaluated *in vitro*, found antifungal to *Claviceps fusiformis*. However, *T. viride* was found most significant with highest mycelial growth inhibition (59.52%) of the test pathogen. The second and third inhibitoriest antagonists found were *T. harzianum* and *T. hamatum* with mycelia growth inhibition of 58.76 and 48.44%, respectively.

Keywords: Biocontrol, *Claviceps fusiformis*, *in vitro*, inhibition

Introduction
Pearl millet *Pennisetum glaucum* (L.) R. Br. is the fourth most important staple cereal crop in the world. Best suited to the harsh climate of the seasonably hot drought prone semi–arid regions of Indian sub-continent. (Loveless, 1967) [2].
Rajasthan, Maharashtra, Gujarat, and Uttar Pradesh share the principal pearl millet growing states. They grew 68.8% of national production on 76.7% of the pearl millet area. In the year (2015-16) total area under pearl millet crop was 8.16m/ha, with grain production of 9.56 million tone and productivity 1172kg/ha. And other important pearl millet growing states are Rajasthan (4.04 million ha), and Maharashtra (0.64million ha), with all India share (57.92), (9.15) respectively and U.P. (0.98 million ha). Pearl millet is also grown in Gujarat (0.39 million ha), Madhya Pradesh (0.27 million ha) and Haryana (0.37 million ha), Karnataka (0.1 million ha). (Agriculture statistics at glance 2016).
Pearl millet is a principal source of energy, protein, vitamins and minerals for millions of poorest people in the regions where it is cultivated. It generally has 9 to13% proteins, but large variation among genotypes ranging from 6 to 21% has been observed Pearl millet contains more calories than wheat, probably because of its higher oil content of 5%, of which 50% are polyunsaturated fatty acids. It is rich in calcium, potassium, magnesium, iron, zinc, manganese, riboflavin, thiamine, niacin, lysine and tryptophan. Pearl millet grain is gluten-free and thus is the only grain that retains its alkaline properties after being cooked which is ideal for people with gluten allergies. (Khairwal *et al*. 2007) [1].
The most important factors responsible are the diseases like Ergot of Bajra, Pyricularia leaf spot, Rust, Downey mildew, Smut and *Rhizoctonia* blight and non-availability of leading varieties, which are resistant to biotic and abiotic stresses. Among these diseases, Ergot of Bajra caused by *Claviceps fusiformis* is one of the most destructive diseases. *Claviceps fusiformis* occurs in most of the African and Asian countries where pearl millet is grown (Ramkrishnan, 1971) [6].
Members of the fungal ascomycetous genus *Claviceps* parasitize more than 600 monocotyledonous plants of the *Poaceae, Juncaceae* and *Cyperaceae*, including economically important crop plant such as ray, wheat, barley, rice, corn, millet and oat. As it infects more than 400 plant species with a disease known as ergot. The common name ergot fungus is derived from the French word for spur (‘argot’) and refers to the dark sclerotia protruding from the ripe grass ear in the final disease stages (Taber, 1985) [7].
Ergot disease can readily be identified when cream to pink mucilaginous droplets called 'honeydew' ooze out of the infected florets on pearl millet panicles.

These droplets contain numerous asexual spores called conidia. Within 10-15 days these droplets dry out and hard, dark brown to black structures, larger than seed and with a pointed apex develop, which protrude from the florets in place of grain. These are called sclerotia (singular sclerotium). During harvesting and threshing, sclerotia get mixed with the grain or fallen to the ground.

Ergot causing fungi infect young, usually unfertilized ovaries, replacing the seeds by dark mycelial masses known as sclerotia. The alkaloids produced by the fungus severely affect health of humans and warm blooded animals and in pearl millet ergot become a problem when growers adopted hybrid technology. (Thomas *et al*. 2015) [8].

Therefore, the present investigation was undertaken for the management of Ergot disease of Bajra causing pathogen *in vitro* by dual culture technique to identify the new effective bio-control agents, which derive maximum benefit to the farmers.

Material and Methods

In dual culture technique, 20 ml of sterilized and cooled potato dextrose agar were poured into sterile Petri plates. Fungal antagonists were evaluated by inoculating the pathogen at one side of Petri plate and the antagonist inoculated at exactly opposite side of the same plate by leaving 3 cm gap. For this actively growing cultures will be used. In case of bacterial antagonist evaluation, two mycelia discs of pathogen were inoculated and bacterial antagonist were streaked in the center of the plate. Each treatment were replicated three times. After required period of incubation i.e., in the control plate growth reached 90 mm diameter, the radial growth of pathogen were measured.

Per cent inhibition over control were as calculated by applying the formula given by Vincent (1927) [9].

$$R = \{(C-T)/C\} \times 100$$

Where

R = Per cent inhibition.

C = Radial growth of pathogen colony in control.

T = Radial growth of pathogen colony in treatment.

Details of experiment

Design : CRD

Replications : Three

Treatments : Nine

Treatment details

Treatment	Bioagents
T_1	*Trichoderma viride*
T_2	*Trichiderma harzianum*
T_3	*Trichoderma hamatum*
T_4	*Trichoderma koningii*
T_5	*Trichoderma longibrachiatum*
T_6	*Aspergillus niger*
T_7	*Bacillus Subtilis*
T_8	*Pseudomonas fluorescens*
T_9	Contol

Observations on radial mycelial growth of the test fungus and bioagents will be recorded at 24hrs interval and will be continued till growth of the test pathogen in untreated control plate is fully covered. Per cent inhibition of the test pathogen will be calculated by applying formula given by Vincent (1927) [9].

Where

R = Per cent inhibition.

C = Growth of the test pathogen in untreated.

Control plates

T = Growth of the test pathogen a in treated plates.

$$R = \{(C - T)/C\} \times 100$$

Results

In vitro evaluation of bioagents

The results obtained on mycelial growth and inhibition of *C. fusiformis* with six fungal and two bacterial antagonists are presented in (Table 1 depicted in fig. 1 and PLATE I) Results revealed that all the bioagents evaluated exhibited fungistatic/ antifungal activity against *C. fusiformis* and significantly inhibited its growth over untreated control.

Table 1: *In vitro* efficacy of bioagents against *C. fusiformis*

Treatments	Average colony Dia. of test pathogen (mm)	#Average % inhibition over control
Trichoderma viride	36.44	59.52 (50.48)
T. harzianum	37.16	58.76 (50.04)
T. hamatum	46.40	48.44 (44.10)
T. koningii	51.63	42.63 (40.76)
T. longibrachiatum	50.51	44.03 (41.57)
Aspergillus niger	48.27	46.38 (42.92)
Bacillus subtilis	57.92	35.70 (36.69)
Pseudomonas fluorescens	58.89	34.32 (35.86)
Contol	90.00	00.00 (00.00)
S. E. ±	1.06	1.17
C.D. @ 1%	4.06	4.49

*Mean of three replication, Dia: Diameter.

Figures in parenthesis are arc sin transformed value.

Among bioagents tested *T. viride* was found most effective with significantly least mycelial growth and highest mycelial growth inhibition of the test pathogen (36.44 mm) followed by *T. harzianum* (37.16 mm). Which was at par each other, and highest mycelial growth inhibition (59.52%) of the test pathogen followed by *T. harzianum* (58.76%) The third best antagonist found *T. hamatum* with mycelial growth of (46.40 mm) and inhibition of 48.44 percent. This was followed by *Aspergillus* niger, *T. longibrachiatum* and *T. koningii* (48.27 mm, 50.51 mm and 51.63 mm). and (46.38%, 44.03% and 42.63%). *Pseudomonas fluorescens* and *Bacillus subtilis* was found less effective with mycelial growth (58.89 mm and 57.72 mm) and inhibition (34.32% and 35.70%).

Results of the present study on antifungal activity of *the T. viride. T. harzianum, T. hamatum T. koningii T. longibrachiatum, Aspergillus niger* and two bacterial antagonists *viz*., *P. fluorescens* and *Bacillus subtilis* against *C. fusiformis* are in conformity with those reported earlier by several workers. Mohan and Jayrajan (1990) [5] found that three antagonists, *Trichoderma harzianum*, *Trichoderma viride* and *gliocladium virens*, significantly reduced sclerotial

germination. Mahadevamurthy *et al*., (1990) [3] treatment with *Aspergillus niger* or *T. viride* completely inhibited the germination of sclerotia. Mahadevamurthy *et al*. (2006) [4] observed under field condition spore suspension and culture filtrates of a nuber of fungi and bacteria (*F. chlamydosporum, Fusarium heterosporum, T. harzianum, T. viride, A. niger, Bacillus subtilis*) reduced the incidence of ergot when applied to flowering heads suppressed due *T. viride* improved the seed germination, seedling vigour and reduced the incidence of seed-borne fungal pathogens. They also reported that *Trichoderma harzianum* showed its efficacy against all *Fusarium* species.

Plate I: *In vitro* efficacy of various bio-agents against *C. fusiformis*

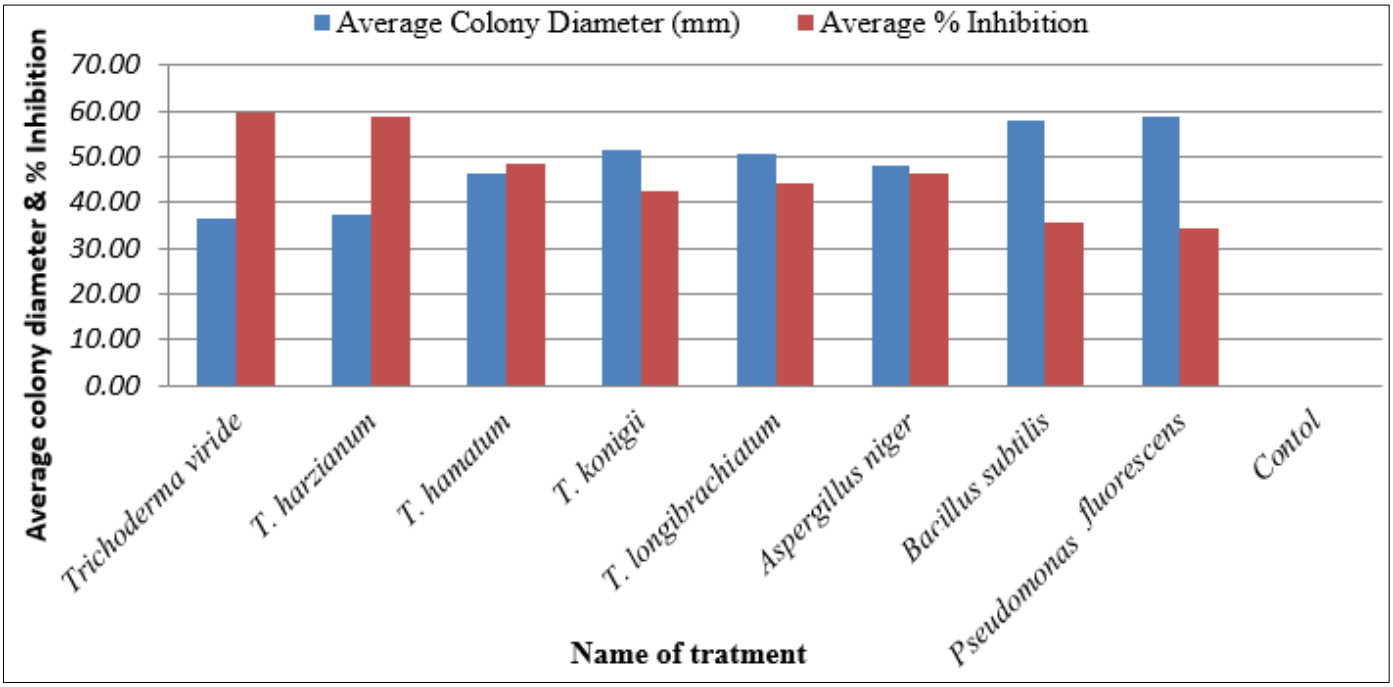

Fig 1: Bioefficacy of bioagents on mycelial growth and % inhibition in *C. fusiformis*

Conclusion

From the present study, it may be concluded that, in biological control, *T. viride* was found most significant.

Acknowledgment

We are thankful to the Head of the Department of Plant Pathology, College of Agriculture, Parbhani for providing the laboratory facilities.

Reference

1. Khairwal IS, Rai KN, Diwakar B, Sharma YK, Rajpurohit BS, Bindu Nirwan, *et al*. Pearl millet: Crop management and seed production Manual. Patancheru 502 324, Andhra Pradesh, India: International Crops Research, 2007.
2. Loveless AR. *Claviceps fusiformis* sp. Nov., the causal agent of an agalactia of sows. Transactions of the British Mycological Society. 1967;50:15-18.
3. Mahadevamurthy S, Prakash HS, Shetty HS. Effect of fertilizer amendment of soil and antagonist treatment on sclerotial germination on *Claviceps fusiformis.* Plant Disease Research. 1990;5(2):212-215.
4. Mahadevamurthy S, Prakash HS, Shetty HS. Effect of spore suspensions and culture filtrates of antagonists on the development of ergot on pearl millet. Asian Journal of Microbiology, Biotechnology and Environmental Sciences. 2006;8:393.
5. Mohan L, Jayarajan R. An *in vitro* test for evaluating the efficacy of mycoparasites on the sclerotial germination of ergot (*Claviceps fusiformis* Lov.) of pearl millet. Journal of Biological Control. 1990;4(1):75-76.
6. Ramkrishnan TS. Diseases of Millets. New Delhi India: Indian Council of Agriculture Research, 1971.
7. Taber WA. Biology of *Claviceps*. I: Biotechnology Series. 1985;6:449-486.
8. Thomas Miedaner, Hartwig H. Biology, Genetics, and management of ergot (*Claviceps* spp.) in Rye, Sorghum, and Pearl Millet. 2015;7:659-678.
9. Vincent JM. Distortion of fungal hyphae in the presence of certain inhibitors. Nature, 1927, 159-180.