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RESEARCH ARTICLE

EFFICACY OF TRICHODERMA AGAINST GREENGRAM ROOT ROT PATHOGEN MACROPHOMINA PHASEOLINA

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Abstract

Greengram (*Vigna radiata* L.) is one of India's most essential and widely cultivated pulse crops. Several biotic and abiotic factors cause the yield loss in Greengram. Among the biotic factors, root rot is one of the devasting factors which can cause maximum yield loss under severe conditions. Ten pathogenic isolates of *Macrophomina phaseolina* causing root rot in Greengram were collected from different locations in the Perambalur District of Tamil Nadu. The isolates produced root rot symptoms under artificial inoculation to confirm the pathogenicity. Among these isolates, *Macrophomina phaseolina 1* isolated from Valikandapuram was virulent, which took for further study. To test the efficacy of fungal biocontrol agents against the Greengram root rot pathogen, ten *Trichoderma viride* were isolated from ten different places in the Perambalur District of Tamil Nadu and evaluated *in vitro*. Among the ten isolates tested against *Macrophomina phaseolina*, the isolate Tv- 4 isolated from the Esanai village of Perambalur District effectively arrested the mycelial growth of *Macrophomina phaseolina*, which recorded 61.00 per cent growth inhibition over control.

Introduction:-

Greengram (*Vigna radiata* L.) is one of the most essential and widely cultivated pulse crops. It is grown all over India as a pure crop and in rice fallows.Greengram is attacked by numerous diseases caused by fungi, bacteria, and viruses. Among these diseases, dry root rot of mungbean caused by *Macrophomina phaseolina* is one of the most devastating diseases occurring in tropical and subtropical countries (Kashyap *et al.*, 2022).This disease causes substantial losses of mungbean causing seed infection ranging from 2.2 – 15.7 per cent, which leads to a decrease in the grain yield by 10.8 per cent as well as protein content (12.3 per cent) in seeds (Kumara *et al.*, 2021).

The pathogen is seed-borne and seed-to-seedling transmission of this disease has been documented when infected seeds are used (Deshmukh *et al.*, 2020). This pathogen attacks all the parts of the plant, like, root, stem, branches,

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petioles, leaves, pods and seeds.The disease symptom starts yellowing and drooping of the leaves, and later infected leaves fall off, and the plant dies within a week. This disease shows dark brown lesions on the stem at ground level, and the bark shows shredding symptoms. The affected plants can be easily pulled out, leaving dried, rotten portions in the ground. The rotten tissues of the stem and root contain many black minute sclerotia (Rangaswami, 1993). Disease incidence is often high when plants are stressed by drought and high temperatures. Sclerotia produced in infected plant tissue function as long-term survival structures in soil and as primary inoculum. *M. phaseolina* can be seed-borne and pycnidia and conidia formed on certain hosts enable aerial transmission. (Pratt *et al*., 1998).

Phytopathogenic fungi are usually managed using synthetic fungicides, but their applications are limited due to adverse environmental and health effects. Due to their harmful effects and the development of resistance to crop pathogens, biological-based alternatives were used to manage the diseases of various crops. Bioagents served as an ecologically safe and acceptable substitute for the fungicidal management of soil-borne diseases in recent years (Abada and Ahmad, 2014). Therefore, *Trichoderma* spp. has been used in this study as a microbial antagonist for the management of the root disease of Greengram. (Padmini, 2014).

Materials and Methods:-

This study was carried out in the Plant Pathology Laboratory, Department of Crop Protection, Thanthai Roever Institute of Agriculture and Rural Development (Affiliated to Tamil Nadu Agricultural University), Valikandapuram, Perambalur district, Tamil Nadu, during 2021 – 2022. In this study, the pathogen causing dry root rot in Greengram and fungal antagonists have been isolated and tested under *in vitro*.

Isolation of *Macrophomina phaseoloina*

The infected plant parts (Roots) showing characteristic symptoms of dry root rot were cut into small pieces (2-3mm). Washed three times with sterilized water and transferred into PDA medium in vitro. The inoculated Petri plates were incubated at 28±2°C for 7 to 10 days.

Isolation of *Trichoderma viride*

Trichoderma viride was isolated from the rhizosphere soil of healthy plants in different locations of the Perambalur district, Tamilnadu. Rhizosphere soils (10g) from different plants were suspended separately in 90 ml of sterile distilled water and vortexed for 5 minutes. 10^{-1} dilution was transferred to the test tube containing 9 ml of sterile water, and 1 ml of 10^{-2} dilution was transferred to 10^{-3} dilution and repeated up to 10^{-4}. 1 ml of 10^{-4} dilution was poured on Petri plates and added 20 ml of *Trichoderma viride* selective medium as described by (Elad and Chet, 1983). The inoculated Petri plates were incubated at 28+2°c for 7 days.

Testing the Antagonistic activity of *Trichoderma viride* against *Macrophomina phaseolina* (Dual culture technique):

Trichoderma species isolated from the rhizosphere were tested for antagonistic activity against *M. phasiolina* by dual culture technique. The different isolates of antagonist and *M.phaseolina* were grown separately on a PDA medium. The virulent strains of *Macrophomina phaseolina* were used for testing the antagonistic activity of *Trichoderma viride* isolates. A 9 mm disc of the test antagonists was taken using a sterile cork borer from the periphery of the 7-day-old colony and placed at one end of a sterile Petri plate already poured with 15 ml of sterile PDA medium (Webster, 1971). Similarly, a 9 mm disc of the pathogen was placed at the opposite end and approximately 75mm away from the antagonist disc. In this study,a control plate (without antagonist) was maintained for calculating the efficacy of *Trichoderma viride* against *Macrophomina phaseolina*. The plates were incubated at room temperature until the control plate was fully grown with the testing pathogen. The radial mycelial growth of the pathogen and test antagonist was measured, and the per cent inhibition of the pathogen was calculated using the formula below.

$$PI = \frac{C - T}{C} \times 100$$

Where,

PI - Percent inhibition of growth of test pathogen.

C - Radial growth (mm) in control.

T - Radial growth (mm) in treatment.

Results and Discussion:-

Isolation and identification of virulent strain of *Macrophomina phaseolina*

Karibasappa *et al.,* (2020) observed high variability in the virulence of the *Macrophomina phaseolina* isolates. The isolates of *Macrophomina phaseolina* obtained from 11 major growing localities in India showed great variability in seedling mortality under *in vitro* conditions (Dev and Sing, 2013). Hence, in this study, various isolates were collected from different localities of the Perambalur District and tested for their efficacy.

Ten isolates of *Macrophomina phaseolina* are isolated from infected Greengram root samples collected from different villages of Perambalur districts. Among the ten isolates, *Macrophomina phaseolina*1isolated from the Valikandapuram village of Perambalur District was a virulent isolate that recorded 9 cm growth on the fourth day after inoculation. (Table 1)

Table 1:- A growth rate of different isolates of *Macrophomina phaseolina*.

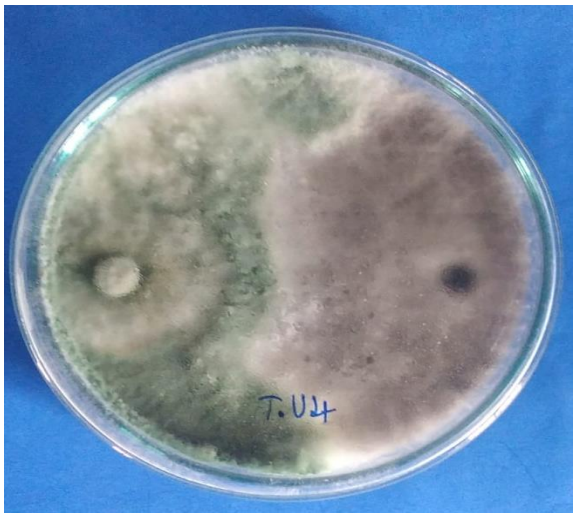
Name of isolates	Locality of Isolation	Day 1 (cm)	Day 2 (cm)	Day 3 (cm)	Day 4 (cm)	Day 5 (cm)
M.p 1	Valikandapuram	1.00	3.50	5.40	7.60	9.00 (17.45)[a]
M.p 2	Thevaiyur	0.50	1.30	3.40	5.70	7.90 (15.32)[f]
M.p 3	Ranjankudi	0.70	2.00	4.10	6.30	8.20 (15.90)[d]
M.p 4	Sengunam	0.65	1.90	3.90	6.00	8.10 (15.71)[e]
M.p 5	Elambalur	0.80	2.10	4.30	6.20	8.30 (16.10)[c]
M.p 6	TRIARD Farm	0.75	1.90	3.80	5.90	7.80 (15.12)[g]
M.p 7	Bhramadesh	0.60	1.80	3.70	5.60	7.70 (14.93)[h]
M.p 8	Veppanthattai	0.55	1.65	3.50	5.40	8.40 (16.30)[b]
M.p 9	V. Kalathur	0.85	2.70	5.10	7.30	7.40 (14.35)[j]
M.p 10	Mangalamedu	0.40	1.50	3.40	5.40	7.60 (14.74)[i]

Isolation and Efficacy of *Trichoderma viride* isolates against *Macrophomina phaseolina*

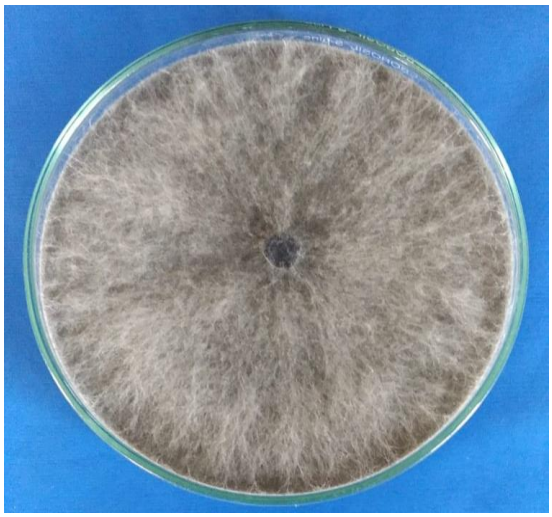
Trichoderma showing the obvious antagonistic effect on pathogens, could rapidly expand and colonize the mycelium of pathogens. In contrast, the mycelia of the pathogen stopped growing, and the colony stopped expanding. At the same time, the *Trichoderma* continued to grow forward until it completely covered the pathogen colony and the entire plate. (Liu et al., 2022)

Zivanovet al. (2017) found that the isolates of *Trichoderma viride* were effective in arresting the growth of soil-borne soil-borne pathogens like *Macrophomina, Fusarium, Pythium* etc. Matloob, 2019 reported that *Trichoderma harzianum* and *T. viride*, which are highly antagonistic against pathogenic fungi *F. solani* and *R. solani*, cause root rot in broad been and were found to increase the parameters of plant growth under the glass-house conditions.The induction of chitinase, b-1, 3 glucanases and an increase in total phenol content was observed, which signified their role in pathogen growth inhibition during antagonism (Gajera *et al.,* 2012).

Totally ten *Trichoderma viride* were isolated from the different villages of Perambalur District and morphologically confirmed by observing the mycelial characters, spore formation, spore characters and pigment formation. The antagonist efficacy of *Trichoderma viride* was tested by following dual culture techniques under *in vitro*. Among the ten isolates tested, *Trichoderma viride*-4 recorded 61.00 per cent of mycelium growth inhibition, followed by *Trichoderma viride*-3 was recorded 55per cent of mycelium growth inhibition over control.



Trichoderma viride -4



Control

Table 2:- Antagonistic activity of *Trichoderma viride* against *Macrophomina phaseolina*.

Name of isolates (Antagonist)	Mycelial growth of M.pat 4 DAI (cm)	Per cent reduction over control (%)
T. v 1	4.90	45.55 (22.77)[e]
T. v 2	5.50	38.88 (19.44)[g]
T. v3	4.00	55.55 (27.77)[b]
T. v4	3.50	61.11 (30.55)[a]
T. v5	6.00	33.33 (16.66)[i]
T. v6	5.80	35.55 (17.75)[h]
T. v7	4.30	52.22 (26.11)[c]
T. v8	6.20	31.11 (15.55)[j]
T. v9	4.70	47.77 (23.88)[d]
T. v10	5.40	40.00 (20.00)[f]
Control	9.00	-

Conclusion:-

Trichoderma viride is an effective fungal biocontrol agent against the soilborne plant pathogens due to its various modes of actions viz., parastism, antibiosis, competition and lysis. In this study *Trichoderma viride* isolate 4 was found to be effective in controlling the Greengram root rot pathogen *Macrophomina phaseolina*. This can be mass multiplied and applied in the soil along with the sand or Farm Yard Manure for the effective management of root rot disease in Greengram in an ecofriendly manner.

References:-

1. Deshmukh, A.J., Sabalpara, A.N and Bambharolia, R.P. (2020) Efficacy of fungicide treatments on seed-borne diseases of Greengram. International Journal of Economic Plants. 7(3):138-143.
2. Dev, S.C., and Singh, B. (2013). Integrated management of major diseases of mungbean seed treatment and foliar application of insecticide, fungicide and biological. Crop Protection, pp. 47, 55–60.
3. Elad, Y., Chet, R.(1983). Improved selective media for isolation of Trichoderma spp. and Fusarium spp. Phytoparasitica, 11, 55–58.
4. Gajera, H., Bambharolia, R., Patel, S., Khatrani, T., &Goalkiya, B. (2012). Antagonism of Trichoderma spp. Against *Macrophomina phaseolina*: evaluation of coiling and cell wall degrading enzymatic activities. Journal of Plant Pathology and Microbiology, pp. 3, 149.
5. Karibasappa, C.S., Bharati, N and Rao, C.S. (2020) Variability of *Macrophomina phaseolina*, the causal organism of sesame root rot and its management. Int.J. Chem. Stud. 8(2): 1791-1797.
6. Kshyap, A.H., Manzar, N., Ahmad, F and Tilagam,J. (2022). First report of root rot in Greengram (*Vigna radiata*) caused by *Ectophoma multirostrata* in India. Plant Disease 106 (8): 1-4.
7. Kumara, P., Pakeerathan, K and Deepani, P.(2021). Assessment of yield loss in Greengram. Biology and life science Forum. 3(1):22
8. Liu, Y., He, P., Munir, S., Ahmed, A., Wu, Y., Yang, Y., Lu, J., Wang, J., Yang, J., Pan, X., Tian, Y and He, Y. (2022) Potential biocontrol efficacy of Trichoderma species against Oomycete pathogens. Front. Microbiol. 13:974024.
9. Padmini, R. (2014). Studies on integrated management of wilt and root rot complex of chickpea (Cicer arietinum L.) caused by *Fusarium* spp. and *Rhizoctonia solani*. PhD. Thesis, MPUAT, Udaipur (Raj.).
10. Pratt, R.G., McLaughlin, M.R., Pederson, G.A., and Rowe, D.E. (1998) Pathogenicity of *Macrophomina phaseolina* to mature plant tissues of alfalfa and white clover. Plant Disease, 82, 1033-1038.
11. Rangaswami, G. (1993). Diseases of crop plants in India. Prentice hall of India Pvt. Ltd., New Delhi, India, p. 520.
12. Zivanov, S.T., Jetvic, R., Laeosevic, M., Zivanov,D., Pap, S.M and Zupunski, V. (2017) Efficacy of *Trichoderma* against common fungal pathogen.Ratar. Povrt. 54(3):104-109.