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In vitro evaluation of different carrier-based *Trichoderma asperellum* granules against *Fusarium* wilt and growth enhancement of brinjal

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Abstract

Fusarium wilt caused by *Fusarium oxysporum* f. sp. *melongenae* is an important soil-borne disease limiting brinjal production. The present study was undertaken to evaluate the efficacy of different carrier-based granular formulations of *Trichoderma asperellum* against *Fusarium* wilt and to assess their effect on plant growth under pot culture conditions. Seven carrier materials, viz., talc, perlite, gypsum, vermicompost, farmyard manure (FYM), charcoal and vermiculite, were evaluated using a completely randomized design with three replications. The *T. asperellum* biomass was incorporated into sterilized carriers with 1Per cent CMC and applied around the root zone of brinjal seedlings grown in pathogen-infested soil. Significant variation was observed among the carrier formulations in disease suppression and plant growth. Talc- and vermicompost-based granules were most effective, recording the lowest disease incidence of 11.11Per cent, with 83.33Per cent reduction over control, respectively. Talc-based granules also recorded the highest plant height (28.37 cm), shoot length (24.53 cm) and root length (12.83 cm), followed by vermicompost and FYM. In contrast, gypsum-based granules showed comparatively poor disease suppression, while the untreated control recorded the highest disease incidence (77.77Per cent) and lowest plant growth. The study demonstrated that carrier material strongly influences the performance of *T. asperellum*, and talc and vermicompost are promising carriers for developing effective granular formulations for the management of *Fusarium* wilt of brinjal.

Keywords: *Fusarium* wilt, *T. asperellum*, carrier, granules, disease incidence, growth parameter

Introduction

Brinjal (*Solanum melongena* L.) is an important vegetable crop belonging to the family Solanaceae and is widely cultivated in tropical, subtropical and warm temperate regions. The crop has considerable economic and nutritional importance and is valued for its dietary fibre, vitamins, minerals and bioactive compounds (Sihachakr *et al.*, 1994; Caguiat and Hautea, 2014) [16, 5]. However, its productivity is affected by several biotic stresses, among which soil borne diseases are particularly important. *Fusarium* wilt is one of the major diseases of brinjal and is predominantly associated with *Fusarium oxysporum* f. sp. *melongenae* (Lolpuri, 2002) [9]. The pathogen is soil-borne and can survive for several years in the absence of a susceptible host. The disease is characterized by stunting, chlorosis, vascular browning, progressive wilting and eventual death of infected plants. The persistent nature of the pathogen makes its management difficult and often results in considerable yield losses. Management of *Fusarium* wilt has traditionally relied on chemical fungicides; however, their excessive and indiscriminate use may lead to environmental contamination, development of fungicide resistance and disruption of beneficial soil microorganisms (Akrami and Yousefi, 2015) [1]. Consequently, biological control has emerged as an environmentally sustainable alternative for managing soil-borne pathogens. Among the various microbial antagonists, *Trichoderma* spp. are considered highly effective because of their broad-spectrum antagonistic activity against several phytopathogenic fungi (Saravanakumar *et al.*, 2016) [14]. *Trichoderma asperellum* possesses biocontrol potential through mechanisms such as mycoparasitism, antibiosis, competition for nutrients and space, production of hydrolytic enzymes and induction of host resistance (Segarra *et al.*, 2013) [15].

Its rapid growth, adaptability to different substrates and ability to colonize the rhizosphere make it a promising biological control agent for the management of soil-borne diseases.

The efficacy of *Trichoderma* as a biocontrol agent is greatly influenced by the formulation and carrier material used for its delivery. Suitable carrier materials provide nutrients and a favourable microenvironment, thereby improving the survival, multiplication, sporulation and establishment of the antagonist (Das *et al.*, 2021) [6]. Materials such as talc, farmyard manure, vermicompost, neem cake, gypsum and other agricultural substrates have been utilized for the preparation of *Trichoderma*-based formulations. Recent advances in bioformulation technology have further improved the shelf life and effectiveness of carrier-based formulations (Sudha *et al.*, 2024) [17]. Granular formulations offer advantages in handling, storage and application and may provide an effective means of delivering *T. asperellum*. Conidia are particularly suitable for formulation because of their stability, ease of production and tolerance to desiccation (Amsellem *et al.*, 1999) [2]. Therefore, selection of a suitable carrier is essential for developing an effective *T. asperellum* granular formulation.

In vitro evaluation provides a rapid and controlled method for comparing the antagonistic efficacy of different *T. asperellum* granular formulations against *F. oxysporum* f. sp. *melongenae*. Such preliminary screening helps identify promising carrier formulations based on their ability to inhibit pathogen growth before further evaluation under pot and field conditions. Considering the economic importance of brinjal, the persistent nature of *Fusarium* wilt and the need for environmentally safe disease management strategies, the present investigation was undertaken to evaluate the *in vitro* efficacy of different granular formulations of *Trichoderma asperellum* against *Fusarium oxysporum* f. sp. *melongenae*, the causal pathogen of *Fusarium* wilt of brinjal.

Material and Method

Isolation, purification and identification of *Fusarium spp*

Brinjal tissues showing typical *Fusarium* wilt symptoms were surface sterilized with 0.1Per cent sodium hypochlorite and placed on PDA plates. After incubation at $25 \pm 2^\circ\text{C}$, developing white to pink colonies were purified by hyphal tip and single-spore isolation. The pathogen was identified based on cultural and morphological characteristics, including colony appearance, macroconidia, microconidia and chlamydospores.

Isolation, purification and identification of *Trichoderma spp*

Rhizospheric soil samples were serially diluted up to 10^{-9} and plated on PDA for fungal isolation. *Trichoderma* isolates were purified by single-spore isolation (Johnston and Booth, 1983) and identified based on cultural and morphological characteristics. The identified *T. asperellum* was maintained on PDA slants at 4°C and mass multiplied in PDB for seven days at room temperature.

Preparation of *Trichoderma asperellum* granules using different carriers

Different carrier materials were evaluated for cost-effective mass multiplication of *Trichoderma asperellum*. The fungus was grown in PDB for 12 days, and 50 ml biomass

suspension was mixed with 100 g sterile carriers along with 1Per cent CMC. The mixtures were thoroughly mixed, shade dried for two days, and stored in polythene bags at $30 \pm 2^\circ\text{C}$.

In vitro efficacy of *Trichoderma asperellum* granules against *Fusarium* wilt of Brinjal

The efficacy of *Trichoderma asperellum* granules against *Fusarium oxysporum* f. sp. *melongenae* was evaluated under pot culture conditions. The pathogen was mass multiplied in sterilized Potato Dextrose Broth (PDB) and incubated at $25 \pm 2^\circ\text{C}$ for 10-15 days. The culture was mixed with sterilized potting soil at 5Per cent (w/w), and the soil was incubated for one month under suitable moisture and temperature conditions to develop sick soil. Healthy, uniform brinjal seedlings were transplanted into the sick soil, and *T. asperellum* granules were applied around the root zone at the prescribed dose. The treatments were maintained under suitable conditions and evaluated for their efficacy in suppressing *Fusarium* wilt and promoting plant growth.

Experimental Detailed

- **Design:** CRD
- **Replication:** Three
- **Treatment:** Eight

Treatment Detailed

Treatment	Carrier Materials
T ₁	Talc + <i>T. asperellum</i> (2:1w/v)
T ₂	Perlite + <i>T. asperellum</i> (2:1w/v)
T ₃	Gypsum + <i>T. asperellum</i> (2:1w/v)
T ₄	Vermicompost + <i>T. asperellum</i> (2:1w/v)
T ₅	FYM + <i>T. asperellum</i> (2:1w/v)
T ₆	Charcoal + <i>T. asperellum</i> (2:1w/v)
T ₇	Vermiculite + <i>T. asperellum</i> (2:1w/v)
T ₈	Control

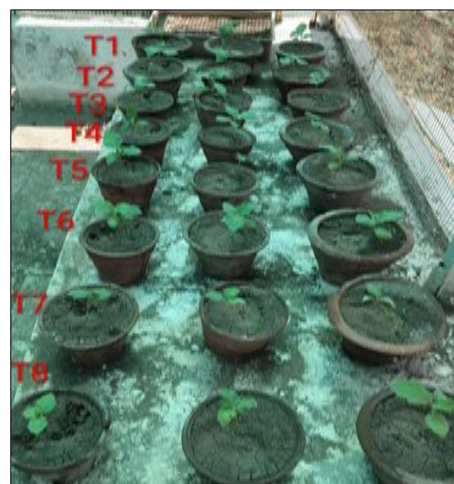


Plate 1: Efficacy of different carrier based *T. asperellum* granules in management of wilt of Brinjal in pot culture.

Result and Discussion

Effect of different carrier based granules of *T. asperellum* on the disease incidence and percent reduction of *Fusarium* wilt of brinjal (Pot culture)

The efficacy of different carrier-based granules of *Trichoderma asperellum* against *Fusarium* wilt of brinjal was evaluated under pot conditions, and the results are presented in Table 1. A considerable variation in disease

incidence was observed among the different carrier formulations, indicating that the choice of carrier material substantially influenced the efficacy of *T. asperellum*. Disease incidence ranged from 11.11 to 77.77Per cent, with the untreated control (T₈) recording the maximum incidence of 77.77Per cent. Among the bioagent treatments, talc-based granules (T₁) and vermicompost-based granules (T₄) recorded the minimum disease incidence of 11.11Per cent each, followed by FYM (T₅) with 22.22Per cent and vermiculite (T₇) with 33.33Per cent. Whereas gypsum-based granules (T₃) were comparatively less effective, recording 66.67Per cent disease incidence. The superior performance of talc-based granules may be attributed to the favourable physical properties of talc, which facilitate better survival, stability and distribution of *Trichoderma* propagules in the soil. Vermicompost also proved to be a highly suitable

carrier, possibly due to its organic matter and nutrient-rich characteristics, which may support the establishment, multiplication and rhizosphere colonization of *T. asperellum*. FYM and vermiculite also exhibited appreciable disease-suppressive potential and were statistically at par with each other, indicating their suitability as alternative carrier materials.

The present findings are consistent with Mishra *et al.* (2020) ^[11], Ramesh and Singh (2017) ^[12], Jaydev *et al.* (2015) ^[8], Bhaumik *et al.* (2021) ^[4] and Das *et al.* (2023) ^[6], who demonstrated the effectiveness of suitable carrier materials in enhancing the efficacy and persistence of *Trichoderma* formulations against *Fusarium* wilt. In the present study, talc and vermicompost-based *T. asperellum* granules were most effective and at par, followed by FYM and vermiculite, while gypsum was least effective.

Table 1: Effect of different carrier based granule of *T. asperellum* on the incidence and percent reduction of wilt of brinjal

Tr. No.	Carrier material	Percent Disease Incidence (%)	Reduction over control (%)
T ₁	Talc	11.11 (11.75)	83.33 (74.99)
T ₂	Perlite	44.44 (41.74)	44.44 (41.73)
T ₃	Gypsum	66.67 (59.78)	22.22 (18.23)
T ₄	Vermicompost	11.11 (11.75)	88.89 (78.23)
T ₅	FYM	22.22 (23.50)	66.67 (59.98)
T ₆	Charcoal	55.56 (48.23)	28.55 (26.74)
T ₇	Vermiculite	33.33 (35.25)	55.55 (48.22)
T ₈	Control	77.77 (66.27)	0.00
S. Em. ±		8.85	11.88
C.D. at 1%		26.78	35.92

(Figures in the parenthesis indicates are sine transformed value), *Mean of three replication

Effect of different carrier based granules of *T. asperellum* on the growth parameter

The data presented in Table 2. revealed significant variation in the growth of brinjal plants treated with different carrier-based formulations of *Trichoderma asperellum* under *Fusarium oxysporum* f. sp. *melongenae* infection. The talc-based formulation (T₁) recorded the maximum plant height (28.37 cm), shoot length (24.53 cm) and root length (12.83 cm), followed by vermicompost (T₄) and FYM (T₅). In contrast, the untreated control (T₈) recorded the minimum plant height (18.47 cm), shoot length (14.90 cm) and root length (6.53 cm). Among the other treatments, vermiculite and perlite showed moderate performance, whereas charcoal and gypsum were comparatively less effective. These results

indicate that carrier material influences the establishment and activity of *T. asperellum* and consequently plant growth under wilt stress.

The present findings was supported by Rao *et al.* (2021) ^[13], reported that enhanced plant height and root development in brinjal treated with *Trichoderma* fortified vermicompost under *Fusarium* wilt conditions. Mishra *et al.* (2020) ^[11] also demonstrated the growth-promoting potential of *T. asperellum* and reported higher viability of the antagonist in talc-based formulations. Similar observations were reported by Bahadur *et al.* (2022) ^[3], Faruq *et al.* (2014) ^[7], Mathivanan *et al.* (2023) ^[10] and who highlighted the effectiveness of talc-based *Trichoderma* formulations in improving plant growth under *Fusarium* wilt conditions.

Table 2: Effect of different carrier based granules of *T. asperellum* on the growth parameter.

Tr. No.	Carrier material	Plant height (cm)	Shoot length (cm)	Root length (cm)
T ₁	Talc	28.37	24.53	12.83
T ₂	Perlite	23.80	19.87	8.77
T ₃	Gypsum	19.87	16.53	6.73
T ₄	Vermicompost	27.60	24.27	11.50
T ₅	FYM	26.17	23.70	10.70
T ₆	Charcoal	22.57	18.67	7.17
T ₇	Vermiculite	24.20	20.83	9.20
T ₈	Control	18.47	14.90	6.53
S. Em. ±		0.2	0.29	0.40
CD at 1%		0.86	1.22	1.65

*Mean of three replication

Conclusion

The present investigation demonstrated that the efficacy of *Trichoderma asperellum* against *Fusarium oxysporum* f. sp. *melongenae* was markedly influenced by the carrier material used for granule formulation. Significant variation was

observed among the different carrier-based formulations in terms of *Fusarium* wilt incidence and plant growth under pot culture conditions. Among the evaluated treatments, talc and vermicompost-based granules were the most effective, each recording the lowest disease incidence of 11.11 Per

cent and the highest reduction over control of 83.33 Per cent. These treatments were followed by FYM and vermiculite, while gypsum was the least effective formulation. The carrier-based formulations also positively influenced the growth of brinjal plants under *Fusarium* wilt stress. Talc-based *T. asperellum* granules recorded the maximum plant height (28.37 cm), shoot length (24.53 cm) and root length (12.83 cm), followed by vermicompost and FYM, whereas the untreated control recorded the lowest growth. Overall, the study indicates that carrier selection is

an important factor in improving the biocontrol efficiency and plant growth-promoting potential of *T. asperellum*. Among the tested formulations, talc and vermicompost emerged as the most promising carriers for managing *Fusarium* wilt of brinjal. These formulations therefore have potential for development as effective, economical and environmentally compatible bioformulations for integrated management of *Fusarium* wilt, although further validation under field conditions is required.

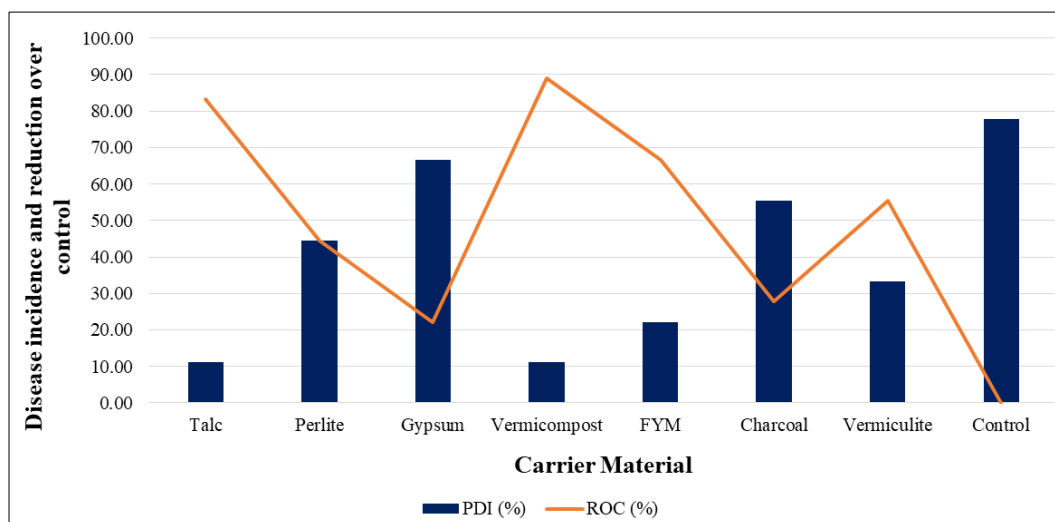


Fig 1: Impact of different carrier based granules of *T. asperellum* on the Percent Disease Incidence and Reduction over control of wilt of brinjal (Pot culture)

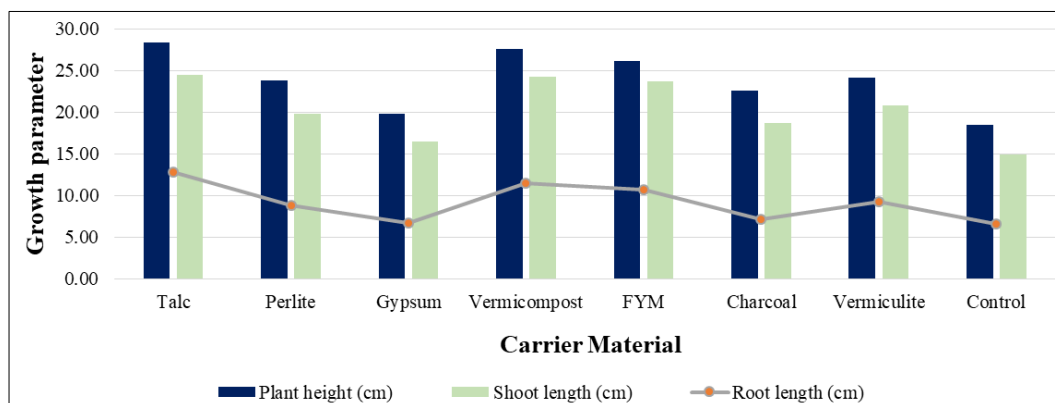


Fig 2: Impact of different carrier based granules of *T. asperellum* on growth parameter of Brinjal (Pot culture)

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