



Compatibility of *Trichoderma asperellum* with Some Selected Fungicides and Insecticides

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ABSTRACT

An investigation was conducted to assess the compatibility of widely utilized fungicides and insecticides, applied at recommended dosages, with *Trichoderma asperellum* under in vitro conditions. A total of ten fungicides and nine insecticides were individually evaluated for their compatibility.. *Trichoderma asperellum* was compatible with fungicides sulphur and copper oxychloride, where the percentage inhibition was 18.55 and 31.55, respectively. The biocontrol agent showed moderate compatibility with fungicides, potassium phosphonate, mancozeb and propineb. All the systemic fungicides tested were highly incompatible with *Trichoderma asperellum* and exhibited cent percent inhibition. As compared to fungicides, insecticides were relatively more compatible with *Trichoderma asperellum*. Insecticides acephate, flubendiamide, thiamethoxam and spiromesifen did not suppress the *in vitro* growth of *Trichoderma*. Imidacloprid and cartap hydrochloride inhibited the growth of *Trichoderma* partially and expressed moderate compatibility. High level of incompatibility of *Trichoderma* was observed with insecticides quinalphos and dimethoate. The *in vitro* study indicated the possibility of using these compatible chemicals in integrated pest management along with the biocontrol agent, *Trichoderma asperellum*.; further field level investigations are needed for further confirmation.

Key Words: Compatibility, Fungicides, Insecticides, *Trichoderma*, biocontrol agent.

INTRODUCTION

Plant diseases are said to be an important factor in reducing food production. Plant diseases are estimated to cause losses ranging from 10% to 40% (Tyskiewicz *et al*, 2022). Though chemical pesticides are the most commonly adopted method for disease control, it poses various threats to environment (Ghorbanpour *et al*, 2018). The non judicious use of pesticides will also lead to resistance development in pathogens (Bora *et al* 2024). Integrated Disease Management (IDM) strategy is the most sustainable and reliable approach for plant disease management. IDM is the integration of various disease management practices such as chemical management, cultural management, biological management etc. for the management of diseases. Biological control agents (BCAs) form an integral part of IDM. *Trichoderma* species are widely utilized as fungal biocontrol agents against plant pathogens. Various

Trichoderma species exhibit antagonistic properties toward phytopathogenic fungi and nematodes, primarily through the production of enzymes and antibiotics. Apart from its effect as a biocontrol agent, it also acts as a biostimulant (Lopez-Bucio *et al*, 2022)

The integration of biological antagonists with synthetic chemical treatments can mitigate the risk of resistance development while lowering the required application of chemical pesticides (Ons *et al*, 2020). Evaluating the compatibility of prospective biocontrol agents with fungicides, insecticides, and fertilizers is essential for developing a sustainable and environmentally friendly disease management strategy. Understanding how fungicides and insecticides affect both the pathogens and the antagonists can help the selection of appropriate fungicides and resistant antagonists through in vitro compatibility studies. Additionally, findings of certain

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Table 1. Compatibility of *Trichoderma asperellum* with different fungicides.

Sr. No.	Treatment	Dose of chemical	Radial growth of fungus(cm) [‡]	Inhibition (%)
T1	Mancozeb 75WP	0.30%	3.82 ^f	57.55 ^b
T2	Mancozeb 75WP	0.40%	3.48 ^f	61.33 ^b
T3	Propineb 50WP	0.25%	4.45 ^e	50.55 ^c
T4	Tebuconazole 250EC	0.15%	0.00 ^g	100.00 ^a
T5	Propiconazole 25EC	0.10%	0.00 ^g	100.00 ^a
T6	Sulphur 80WP	0.20%	7.33 ^b	18.55 ^f
T7	Copper Oxychloride 50WP	0.30%	6.16 ^c	31.55 ^e
T8	Hexaconazole 5EC	0.20%	0.00 ^g	100.00 ^a
T9	Trifloxystrobin 25%+Tebuconazole 50% (75 WG)	0.05%	0.00 ^g	100.00 ^a
T10	Potassium Phosphonate 40%	0.30%	4.88 ^d	45.77 ^d
T11	Carbendazim 50WP	0.10%	0.00 ^g	100.00 ^a
T12	Carbendazim 50WP	0.20%	0.00 ^g	100.00 ^a
T13	Control	-	9.00 ^a	0.00 ^g
	CD(0.05)		0.345	3.835

*Mean of 5 replications

researchers indicate a synergistic effect of *Trichoderma* with fungicides when applied in combination for the management of soil-borne diseases (Wojtkowiak-Gebarowska and Pietr, 2006).

In this context, the study sought to investigate the potential of combining *Trichoderma asperellum* with fungicides and insecticides under in vitro conditions.

MATERIALS AND METHODS

The experiment was conducted during 2019-2020 at the laboratory of Krishi Vigyan Kendra, Pattambi, Kerala. Pure culture of antagonistic microorganism *Trichoderma asperellum* maintained at KVK Pattambi was used for the study. The efficacy of the isolate against major plant pathogens is already proven. The *Trichoderma asperellum* culture was subcultured and maintained on Potato Dextrose Agar (PDA) plates for further studies. (Potato 250g, Dextrose 20g, Agar 20g, Water 1 l).

An *in vitro* study was conducted to assess the compatibility of ten fungicides and nine insecticides with the biocontrol agent, *Trichoderma asperellum* employing the poisoned food technique. The recommended concentrations of each chemical, as detailed in Tables 1 and 2, were incorporated into molten sterile potato dextrose agar (PDA) before being poured into separate sterile petri dishes. Control plates were prepared using PDA without any added chemicals. For each treatment, five plates were maintained. Mycelial discs, 0.5 cm in diameter, were aseptically obtained from a seven-day-old actively growing fungal culture using a sterile cork borer and positioned at the center of the treated PDA plates. The inoculated dishes were incubated at room temperature ($28 \pm 2^\circ\text{C}$), and measurements of radial colony growth (in cm) were taken after five days, at which point the control plate recorded full growth of *Trichoderma asperellum*.

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Table 2. Compatibility of *Trichoderma asperellum* with different insecticides.

Sr. No.	Treatment	Dose of chemical	Radial growth of fungus(cm ‡)	Inhibition (%)
T1	Quinalphos 25%EC	0.20%	2.28 ^c	74.66 ^a
T2	Dimethoate 30%EC	0.20%	3.84 ^d	57.33 ^b
T3	Acephate 75%SP	0.16%	9.00 ^a	0.00 ^c
T4	Imidacloprid 200SL	0.03%	6.92 ^b	23.11 ^d
T5	Thiamethoxam 25% WG	0.02%	9.00 ^a	0.00 ^c
T6	Cartap Hydrochloride 50 SP	0.20%	6.12 ^c	32.00 ^c
T7	Flubendiamide 20%WDG	0.025%	9.00 ^a	0.00 ^c
T8	Flubendiamide 480SC	0.01%	9.00 ^a	0.00 ^c
T9	Spiromesifen 240SC	0.07%	8.66 ^a	3.77 ^c
T10	Control	-	9.00 ^a	0.00 ^c
	CD(0.05)		0.641	7.125

*Mean of 5 replications

The radial mycelial growth was evaluated by measuring its diameter. Colony growth on control plates, which were not treated with any chemicals, was compared to the growth on plates treated with different concentrations of the test substances, and the differences were quantified as percent inhibition. The percent inhibition of *Trichoderma* was determined based on the colony's growth diameter using the following formula:

$$I = \frac{[C - T]}{C} \times 100$$

where I represents percent inhibition, C is the radial growth of *Trichoderma asperellum* in the control plates, and T is the radial growth of *Trichoderma asperellum* in the treated plates. The data were statistically analyzed using analysis of variance for a Completely Randomized Design (CRD), employing the statistical software 'WASP 2.0' for the *in vitro* study results.

RESULTS AND DISCUSSION

Inhibition on mycelial growth of *Trichoderma asperellum* by fungicides

The radial growth of the fungus on PDA media as well as the percentage inhibition compared to untreated control are given in Table 1 and Plate 1.

All the fungicides exhibited some degree of inhibition on the growth of *Trichoderma asperellum*. Contact fungicides exhibited only partial inhibition against *Trichoderma asperellum*. Lowest inhibition was noticed in sulphur 80WP (18.55%). Copper oxy chloride exhibited 31.55 % inhibition at 0.3%. Potassium phosphonate (0.3%) exhibited 45.77 % inhibition whereas highest inhibition among contact fungicides were observed in mancozeb 75 WP. The moderate compatibility of copper oxy chloride and potassium phosphonate has a great practical significance as these fungicides are recommended for the management of quick wilt in black pepper and *Trichoderma* spp. are widely used as a prophylactic measure to combat the disease and is highly efficient for its management. The present results indicated the possibility of integration of both these components for sustainable disease management. The work conducted by Susheela and Thomas (2010) confirms the result as they observed zero percent inhibition of copper oxychloride (0.25%) against *Trichoderma harzianum* in *in vitro* studies. Their field trials also proved compatibility of copper oxychloride against *Trichoderma harzianum*. However, Shahida *et al* (2010) reported 67% inhibition by copper oxychloride on *Trichoderma*

viride and had studied the effect of potassium phosphonate on the growth of *Trichoderma viride* and reported complete compatibility. But in our study 45% inhibition was noticed. Theertha *et al* (2017) reported an inhibition of 52.7% by mancozeb 75 WP on the mycelia growth of *Trichoderma asperellum* even at a lower concentration of 800ppm.

The systemic fungicides fully inhibited the *in vitro* growth of *Trichoderma asperellum*. Cent percent inhibition was recorded in Tebuconazole (0.15%), Propiconazole (0.1%), Hexaconazole (0.2%), Trifloxystrobin+Tebuconazole (0.05%) and Carbendazim (0.1% and 0.2%). This result suggested that the biocontrol agent *Trichoderma* is incompatible with these fungicides. Similar *in vitro* incompatibility results with *Trichoderma* spp. were reported by several other workers. Shashikumar *et al* (2019) reported 94% inhibition on the *in vitro* growth of *Trichoderma viride* with carbendazim at 0.1%. Research conducted by Arunasri *et al* (2011) and Madhavi *et al* (2011) on the compatibility of *Trichoderma* species with propiconazole revealed that the biocontrol agent was incompatible with the fungicide, as it resulted in 100% inhibition of mycelial growth at the tested concentration. The inhibition of mycelia growth of *Trichoderma* by various insecticides, fungicides and fertilizers used in cardamom was studied by Dhanya *et al* (2017) and observed 90% inhibition by hexaconazole. The incompatibility of hexaconazole with *Trichoderma* was also reported by Soumik *et al* (2010).

The difference in inhibition levels reported by various workers may be due to the difference in strains used. Some strains of *Trichoderma* exhibit better compatibility with fungicides and can be incorporated in IDM strategies (Dutta and Chatterjee, 2004). Yang *et al.* (2005) reported that *Trichoderma* can degrade xenobiotic compounds and persist in environments with fungicide residues. This ability may be helping them to overcome the toxicity of even broad-spectrum fungicides like copper oxy chloride and limit the inhibition to 31%. Further field level investigations are needed to study the possibility of incorporating *Trichoderma* sp. in IDM strategies along with fungicides like sulphur and

copper oxychloride which shows moderate compatibility.

Inhibition on mycelial growth of *Trichoderma* by insecticides

Insecticides exhibited lower toxicity to *Trichoderma asperellum* when compared to fungicides (Table 2, Plate 2).

The results revealed that five insecticides out of nine were completely compatible to the growth of *Trichoderma asperellum* at the recommended dosages under *in-vitro* condition. The percent inhibition of Spiromesifen was statistically at par with that of Flubendiamide, Thiamethoxam and Acephate indicating the high compatibility of these insecticides with *Trichoderma asperellum*. Imidacloprid and cartap hydrochloride are moderately compatible with *Trichoderma asperellum*.

Quinalphos 25% EC exhibited highest level of toxicity to *Trichoderma asperellum* followed by Dimethoate. These results were consistent with the findings of Soumik *et al* (2010), which showed that the insecticide quinalphos exhibited toxicity at a low concentration of 10 ppm, indicating substantial incompatibility with *Trichoderma harzianum*. In contrast, research conducted by Madhavi *et al* (2008) reported that *T. harzianum* and *T. viride* demonstrated high compatibility with imidacloprid. Dhanya *et al* (2017) studied the *in vitro* inhibition of insecticides against *Trichoderma viride* and reported the compatible nature of imidacloprid 17.8 SL and Flubendiamide 39.35 SC with *T. viride*. Their study also revealed the incompatibility of quinalphos with *T. viride*. Rangathswamy *et al* (2011) found that Thiamethoxam demonstrated strong compatibility with *Trichoderma* sp. In a separate study, Bheemaraya *et al* (2012) noted that imidacloprid and dimethoate were compatible with *T. harzianum*, while quinalphos exhibited incompatibility. The study by Theertha *et al* (2017) also reveals the incompatibility of quinalphos and dimethoate with *T. asperellum*. The present study also supports these findings.

The study revealed the levels compatibility of various fungicides and

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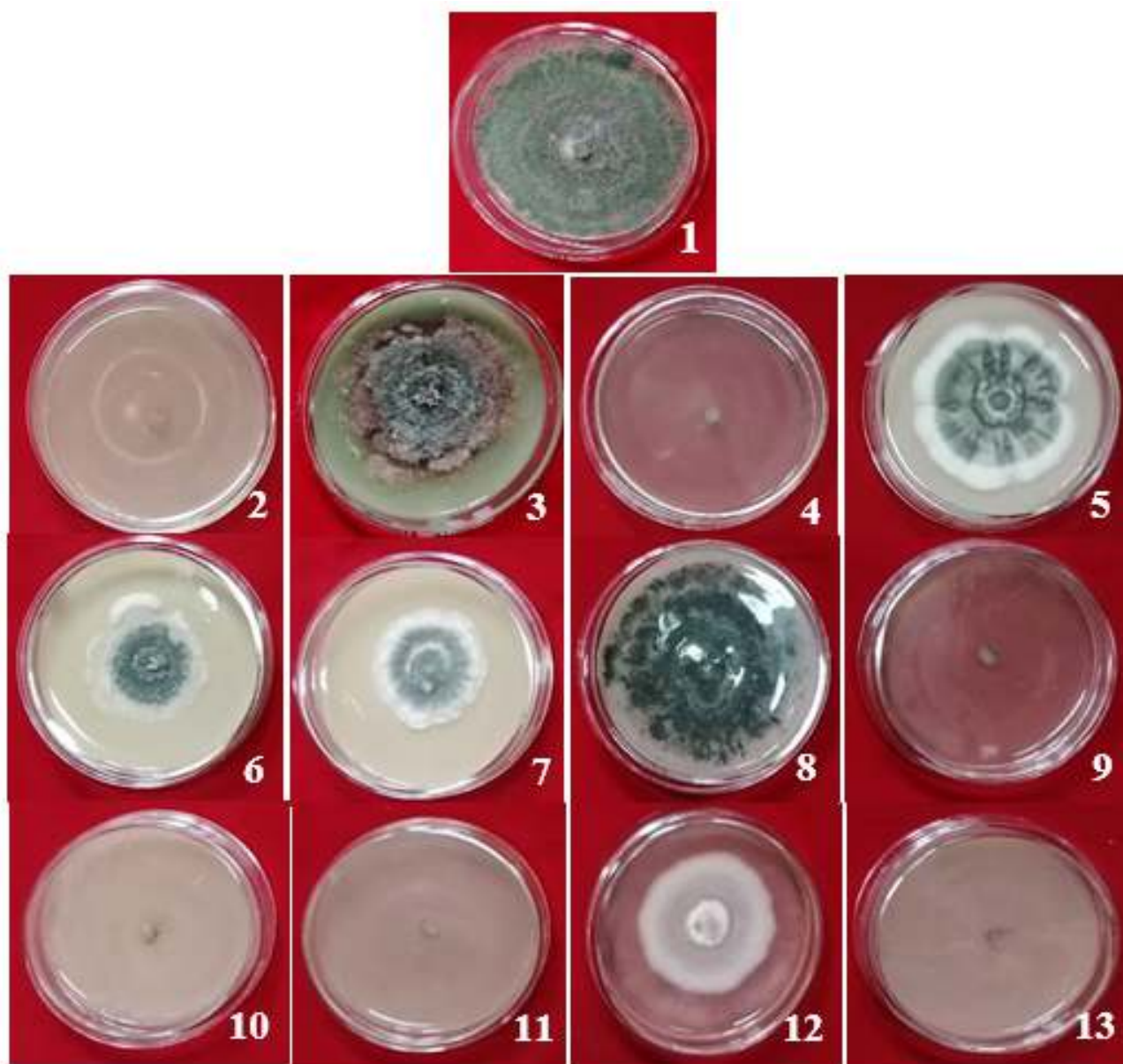


Plate 1 (1-13) *In vitro* effect of fungicides on growth and sporulation of *Trichoderma* 1-Control. 2-Propiconazole 25EC (0.10%). 3-Copper Oxychloride 50WP (0.30%). 4-Hexaconazole 5EC (0.20%). 5-Propineb 50WP (0.25%). 6-Mancozeb 75WP (0.40%). 7-Mancozeb 75WP (0.30%). 8-Sulphur 80WP (0.20%). 9-Trifloxystrobin 25% + Tebuconazole 50% (0.05%). 10-Carbendazim 50WP (0.10%). 11-Carbendazim 50WP (0.20%). 12-Potassium Phosphonate 40% (0.30%). 13-Tebuconazole 250EC (0.15%)

insecticides at their recommended doses against KAU isolate of *Trichoderma asperellum*. This information will aid in selecting the appropriate combinations of fungicides or insecticides to use alongside the biocontrol agent, *Trichoderma*, in integrated pest management strategies. Additionally, the findings from the *in vitro* study should be validated through *in vivo* research in

field conditions.

CONCLUSION

Integrating pesticides with biocontrol agents represents a more sustainable approach to disease management. The combined use of chemical treatments and biocontrol agents can enhance the duration of effective disease control

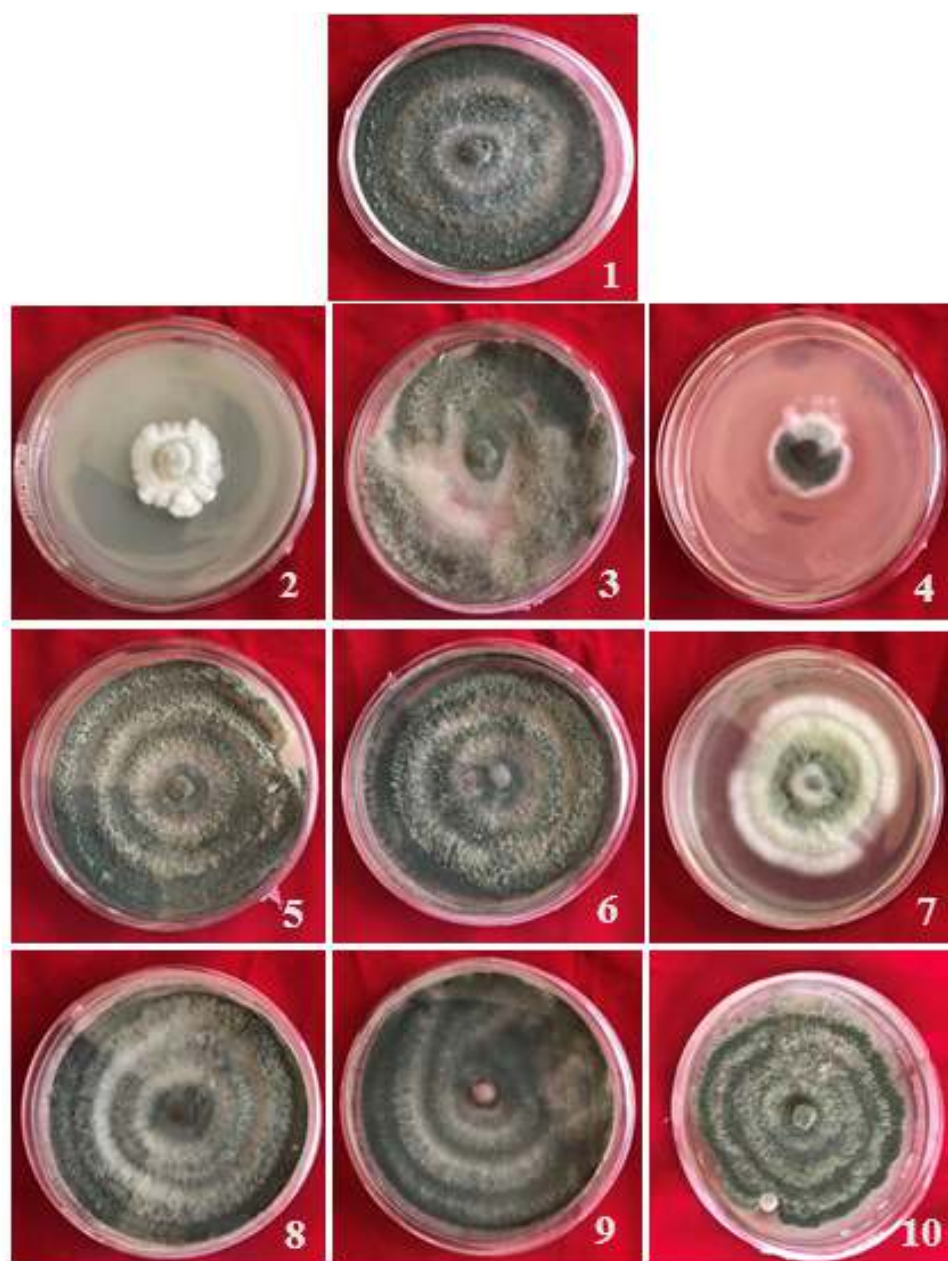


Plate 2 (1-10) Effect of selected insecticides on *in vitro* growth and sporulation of *Trichoderma asperellum*. 1-Control. 2-Quinalphos 25%EC(0.20%). 3-Spiromesifen 240SC(0.07%). 4-Dimethoate 30%EC(0.20%). 5-Flubendiamide Fame(0.01%). 6-Flubendiamide Takumi(0.025%). 7-Cartap Hydrochloride(0.20%). 8-Acephate 75%SP(0.16%). 9-Thiamethoxam 25%WG(0.02%). 10-Imidacloprid 200SL(0.03%).

while also lowering crop protection costs. Therefore, assessing the compatibility of biocontrol agents with agrochemicals is essential when deciding on management strategies. This study demonstrated the compatibility of the KAU isolate of *Trichoderma asperellum* with various agrochemicals, particularly fungicides and

insecticides. Among the tested fungicides, Sulphur 80 WP and Copper oxychloride were found to be moderately compatible with *Trichoderma asperellum* and safe to be applied together in the system. Almost 50% of the mycelial growth of *Trichoderma asperellum* was inhibited by Potassium phosphonate, Mancozeb

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and Propineb. All the systemic fungicides tested, viz., Carbendazim 50 WP and triazole fungicides were 100 per cent incompatible with *Trichoderma asperellum*. Most of the insecticides tested were compatible and safe to be used with *Trichoderma asperellum*. Among the insecticides tested, Quinalphos and Dimethoate were incompatible whereas Imidacloprid and Cartap hydrochloride were moderately compatible with *Trichoderma asperellum*. To reach definitive conclusions, it is essential to study the effects of agrochemicals on *Trichoderma asperellum* in field conditions.

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