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EFFICACY OF INDIGENOUS ENTOMOPATHOGENIC FUNGI IN THE MANAGEMENT OF SUCKING PESTS OF CUCUMBER

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ABSTRACT

Entomopathogenic fungi (EPF) are promising microorganisms for biologically regulating insect pests. Isolation of entomopathogenic fungi from the soil is ideal as they naturally inhabit soils. We have isolated fungi from the soils of various regions of Tamil Nadu and some of the isolated fungal species demonstrated entomopathogenic properties. For the isolation of EPF, soil baiting using *Galleria mellonella* larva was performed. Out of the fifty-five soil samples collected, a total of thirty-seven fungal isolates were obtained; out of which, seventeen were identified as fungi with entomopathogenic ability. EPF belonging to the genus *Aspergillus* was highly isolated followed by four isolates of *Beauveria bassiana*, one isolate each of *Purpureocillium lilacinum*, *Clonostachys rosea*, *Talaromyces pinophilus*, *Penicillium simplicissimum*, and *Fusarium solani*. Furthermore, the pathogenicity test confirmed the virulence of isolated EPF with 3 *Beauveria bassiana* isolates and *Penicillium lilacinum* showing 100% mortality in the tested insect. All the isolates of *B. bassiana* were tested under laboratory conditions for their efficacy against *A. gossypii* and 97.67% of mortality was recorded by isolate TNAU SGG 1. The best isolate (TNAU SGG 1) was employed in the management of sucking pests of cucumber under the IPM programme and EPF employed in the IPM strategy proved to be successful in controlling sucking pests of cucumber and in providing sustainable yield.

***Keywords*:** Entomopathogenic fungi, *Galleria mellonella*, Isolation, Pathogenicity, Integrated pest management, *Beauveria bassiana*, *Aphis gossypii*

Introduction

The rapid rise in the global population is increasing demand for food production. The major problem with increasing food production is that insects are becoming detrimental to the crops. Losses to crops due to insect pests were 23% (Dhaliwal *et al*. 2015). Though several chemicals have been developed to combat pests, they are not without drawbacks such as resistance, resurgence, residues in edible food, risk to natural enemies, and so on. To surmount these, other methods of pest control must be followed. One such method is utilizing Entomopathogens to bring pest populations under the economic threshold level. Entomopathogens are the disease-causing microorganisms in insects, mites, and ticks. They include bacteria, viruses, fungi, and nematodes. The utilization of these organisms in integrated pest management is also termed "microbial control of arthropod pests." Among the world's total pesticide market, 1.3% is contributed by microbial pesticides (Hajek and Leger 1994). The present study is focused on entomopathogenic fungi.

Entomopathogenic fungi contribute more to insect diseases (60%) compared to other entomopathogens (Vega *et al*. 2009). Elie Metchnikoff was the first to conduct field trials with EPF in 1888. EPF are present in soil ecosystems all over the world, and they infect various insects (Behie and Bidochka 2014; Litwin *et al*. 2020). Six classes make up this group of fungi. Oomycetes, Chytridiomycota, Microsporidia, Entomophtoromycota, Basidiomycota, and Ascomycota are the classes. From all these classes,

nearly 750 species of EPF from 85 genera are known (Norjmaa *et al*., 2019). The most known EPFs are *Metarhizium*, *Beauveria*, *Paecilomyces*, and *Verticillium*. They infect insects through contact, unlike other entomopathogens which need to be orally ingested. These fungi are not infective to plants, animals, and humans. They are spore formers and are distinguished from other saprophytic fungi by their mode of action. They adhere to the insect, penetrate the insect cuticle, produce hyphae in the insect body, and finally cause the death of the insect.

The EPF infects insects naturally and their cadavers can be collected to isolate the fungi. However, diverse ranges of EPF are present in the soil. Hence, isolation from soil provides us with various EPFs which can be effective against pests. Isolation of EPF from soil can be done by serial dilution method and baiting method.

Cucumber is a subtropical crop that flourishes in warm climates with ample moisture. Under protected cultivation, cucumber faces the menace of several insect pests, including greenhouse whitefly [*Trialeurodes vaporarium* (Westwood)], cotton whitefly [*Bemisia tabaci* (Gennadius)], melon aphid [*Aphis gossypii* Glover], western flower thrips [*Frankliniella occidentalis* (Pergande)], caterpillars, cucumber beetles, leaf miners, onion thrips [*Thrips tabaci* (Linderman)], plant bugs and two-spotted spider mite [*Tetranychus urticae* Koch]. Aphids are responsible for transmitting cucumber mosaic virus, significantly reducing cucumber yield by up to 60% (Thackray *et al*., 2004). Viruses transmitted by whiteflies also contribute to yield losses ranging from 10 to 20% (Abrahamian and Abou-Jawdah, 2014). For cucumbers, damage caused by red spider mites leads to yield loss when the leaf damage index reaches 1.9 or when 30% of the leaf area is affected (Hussey and Parr, 1963).

Tamil Nadu is a South Indian state with a dry sub-humid to semi-arid climate. The state is completely dependent upon monsoons for water sources, so monsoon failures lead to water scarcity and drought. Since EPF need enough moisture for their growth in the soil, the isolation of fungi from the soils of Tamil Nadu is uncertain. Hence, the present study was conducted to isolate and identify the fungi, observe the infectivity rate of isolated fungi, and exploit their potential in managing sucking pests of cucumber under protected conditions.

Materials and Methods

Soil sampling

Fifty-five Soil samples were collected from various places in Tamil Nadu (Fig 1). Litter was removed from soil surface and soil was dug to 15 cm depth for soil sampling. 1 Kg of soil was collected randomly from five places in each plot in a sealed polythene bag, stored at -20ºC, and was carried to the laboratory. Soil samples were air-dried and sieved to remove roots, clods, etc., and used for baiting.

Rearing Wax moth

Greater Wax moth (*Galleria mellonella*) pupae were obtained from Insectary, Tamil Nadu Agricultural University, Coimbatore (11.0142° N 76.9358° E) and reared in plastic boxes for soil baiting by providing an artificial diet. Different artificial diets were used by various scientists for rearing wax moth larvae. Slight modifications were made to the existing diet (Gitanjali 2021) and an artificial diet was prepared. The constituents of the diet are wheat flour (350 gm), corn flour (200 gm), milk powder (120 gm), yeast (60 gm), honey (120 ml), glycerine (150 ml), wheat bran (60 gm), bees wax (60 gm), vitamin E capsule (2), streptomycin sulphate (0.5 gm). Honey and glycerine were heated to melt beeswax, added to the remaining ingredients, and mixed well to prepare an artificial diet. It was stored at 4ºC in the refrigerator for further use. Adults were provided with a 30% honey solution.

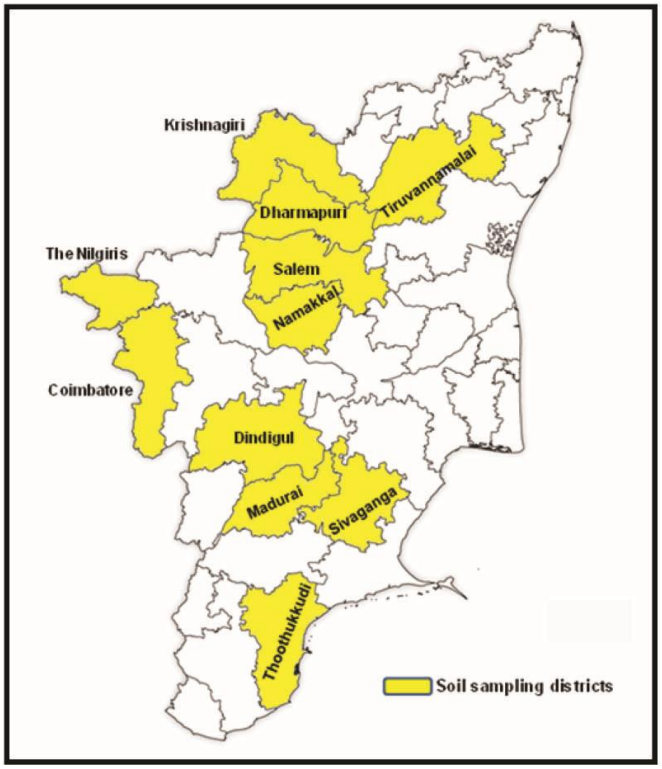

Fig. 1 : Soil sampling regions

Soil baiting with *Galleria mellonella*

Fourth instar larvae were used for the soil baiting method. First, the larvae were heat-treated to prevent webbing in the soil. Water was heated to 56ºC in the water bath, and then larvae were treated for 10-15 seconds in the hot water, placed on a dry tissue paper, and then left in dark condition for 3 hours. Soil was placed in boxes and sprayed with distilled water to provide required moisture for the growth of EPF and then 4 heat-treated larvae were placed in these boxes. These boxes were incubated at 25 ± 5 ºC for 21 days. After 21 days, the larvae infected with fungi were sterilised in 1% sodium hypochlorite for 2 minutes, followed by washing them in distilled water thrice. The sterilised larva was placed on SDAY media in a Petri plate for further growth.

Morphological and Molecular identification

Morphological identification was done by observing macroscopic characteristics of fungi such as colony colour–front and reverse side, colony texture, growth pattern, and microscopic characters such as the shape of the spore.

Fungal DNA extraction

Genomic DNA extraction from fungal mycelium was done by using the CTAB (Cetyltrimethyl-ammonium bromide) method (Zhang *et al*., 2010). 10 mg of fungal mycelium was scraped from 15-day-old fungal plates and crushed in mortar and pestle using 1 ml of CTAB buffer. Crushed mycelium was transferred to an Eppendorf tube and incubated in a water bath @ 65 ºC for 1 hour. Then centrifuged @12000 rpm for 10 minutes at 4 ºC , the supernatant was collected, and an equal volume of Phenol: Chloroform: Isoamyl alcohol (25:24:1) was added and vortexed. It was centrifuged @ 12000 rpm for 10 minutes at 4 ºC. Phase separation was observed, the supernatant was collected and an equal volume of ice-cold isopropanol was added and incubated overnight @ -30 ºC. The next day, it was centrifuged @13000 rpm for 15 minutes at 4 ºC and the DNA pellet was washed in 70% ethanol, air dried, and dissolved in 50 µl of nuclease-free water.

Amplification and Sequencing

ITS1-5.8S-ITS2 region of extracted rDNA was PCR (Polymerase chain reaction) amplified and sequenced using primers ITS1 (5´-TCCGTAGGTGAACCTGCGG-3´) and ITS4 (5´-TCCTCCGCTTATTGATATGC-3´). PCR mixture (30 µl) consisted of 15 µl of universal master mix, 12 µl of molecular water, 1 µl of primers ITS1 and ITS4, and 1 µl of extracted DNA. PCR amplification was done with an initial denaturation at 95 ºC for 10 minutes followed by 35 cycles of denaturation at 95 ºC for 1 minute, annealing at 52 ºC for 30 seconds, and final elongation at 72 ºC for 2 minutes (Gandarilla-Pacheco *et al* 2021). After amplification, the amplicons were run through 1% Agarose gel and Ethidium bromide and bands were observed under UV in the Gel documentation unit. Amplified PCR products were processed at Syngenome (OPC) Private Limited, Coimbatore, and sequences were obtained. Obtained sequences were compared with sequences in the NCBI (National Centre for Biotechnology Information) database using the Basic Local Alignment Search Tool (BLAST). Sequences were then submitted to Genbank (NCBI) and accession numbers were obtained (Table 2). MEGA 11.0 software was used for the construction of a phylogenetic tree by using the Neighbor-joining tree statistical method and Tamura – 3 model (Tamura *et al*., 2007) with 1000 bootstrap replications (Fig 2).

Infectivity rate

A preliminary pathogenicity test (Koch's Postulates) was conducted to confirm the infectivity rate of isolated entomopathogenic fungi. The spore suspension was prepared by scraping the fungi into sterile distilled water and the spore count was adjusted to 1 x 10^8 using Haemocytometer. The fourth instar larva of wax moth was dipped in spore suspension, air dried, released in a box, and incubated @ 25 ± 5 ºC for 11 days. 15 larvae were kept in each of 4 replicates. Larvae were inspected every day for 15 days.

Efficacy of *Beauveria bassiana* isolates against *Aphis gossypii*

Four isolates of *Beauveria bassiana* (TNAU SGG 1, TNAU OTC 1, TNAU DKT 1, and TNAU IDP 1) were tested for their efficacy against *Aphis gossypii* in laboratory conditions at four different concentrations (1×10^8, 1×10^7, 1×10^6 and 1×10^5 spores/ml). The bioassay was conducted using the leaf dip method and was replicated four times. Mortality percent, LC_{50}, and LT_{50} were calculated.

Entomopathogenic fungi against sucking pests of cucumber under protected conditions

Two field trials were conducted from September to November 2022 and May to July 2023 in a farmers' field, in Kuppepalayam village, Coimbatore district, Tamil Nadu. Integrated pest management (IPM) module incorporating silver mulching, yellow and blue sticky traps (25/ha), Azadirachtin 1500 ppm @ 1% at 15 DAS, an isolate of *Beauveria bassiana* (TNAU SGG 1) 1×10^8 spores/ml @ 7 ml/L at 35 and 70 DAS and spinosad 45 SC @ 0.3 ml/L at 60 DAS was compared with farmers practices in managing sucking pests of cucumber and cost economics was calculated based on yield.

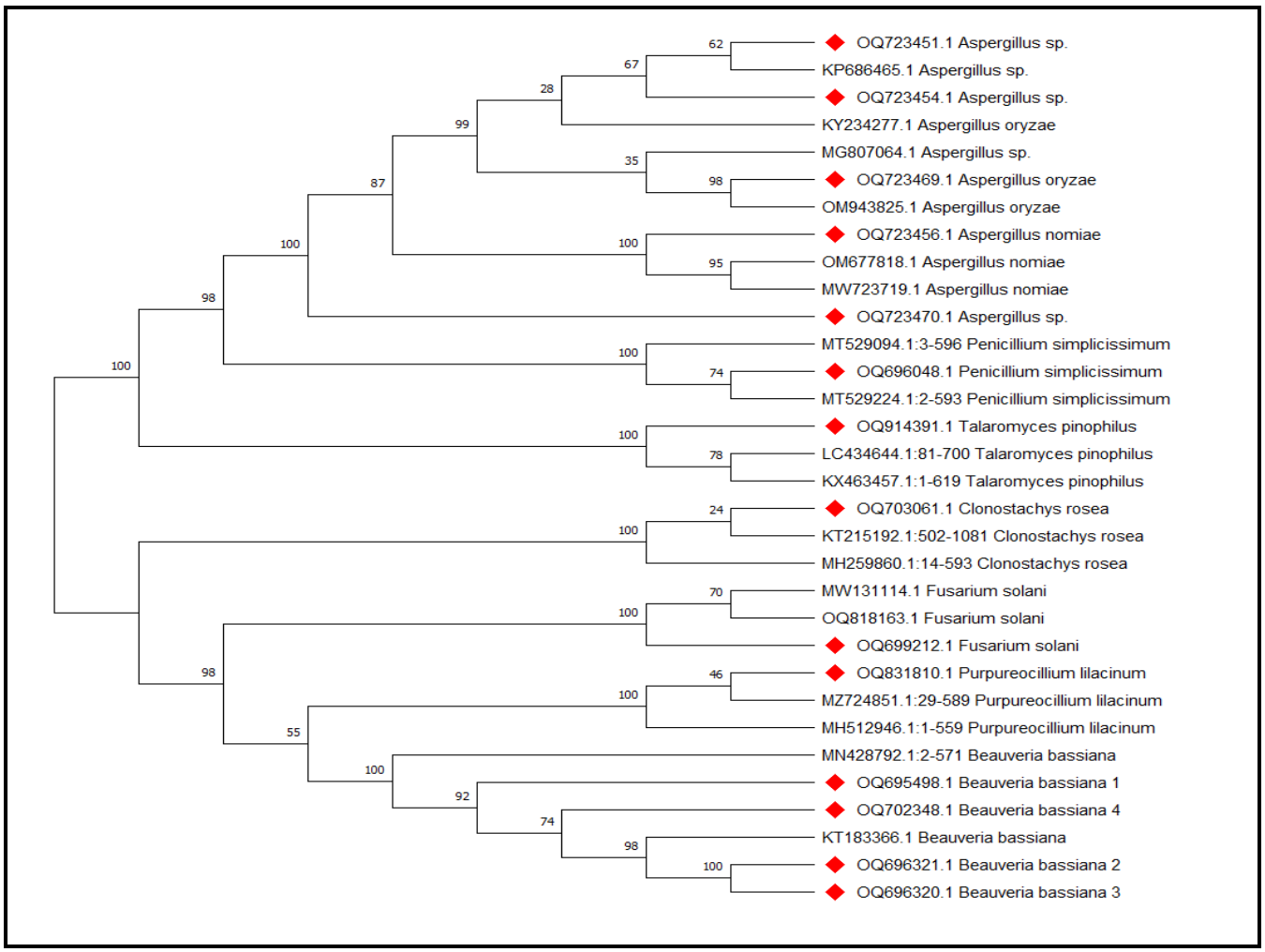

Fig. 2 : Phylogenetic tree comparing the ITS sequences of isolated Entomopathogenic fungi (in red rhombus) with other ITS sequences from NCBI database

Data analysis

Data on Percent mortality was corrected using Abbott's formula (Abbott 1925) and Concentration mortality (LC_{50}) and time mortality (LT_{50}) were calculated using probit analysis (Finney 1971). Data from lab studies was subjected to arcsine transformation and data from field studies was subjected to square root transformation.

Results

Fungi isolated from the soils of Tamil Nadu

Morphological identification of EPF was done based on the taxonomic keys (Barnett and Hunter 1999; Humber 2012) (Table 1). Species confirmation was achieved through sequence comparison with the NCBI database.

Table 1 : Morphological characters of isolated entomopathogenic fungi

Fungal species	Growth pattern	Colony colour		Texture	Shape of spore
		Front	Back		
Beauveria bassiana 1	Disperse	White	Pale yellow	Powdery	Round
Beauveria bassiana 2	Disperse	White	Pale yellow	Powdery	Round
Beauveria bassiana 3	Disperse	White	Pale yellow	Cottony	Round
Beauveria bassiana 4	Disperse	White	White	Cottony	Round
Clonostachys rosea	Disperse	White to Purple	Light yellow	Cottony	Round
Purpureocillium lilacinum	Disperse	Purplish grey	White	Cottony	Oval
Talaromyces pinophilus	Disperse	White	White	Cottony	Round

Penicillium simplicissimum	Disperse	Dark Green	Yellow	Powdery	Rod-shaped
Fusarium solani	Disperse	White	Pale white	Cottony	Ellipsoid
Aspergillus nomiae	Disperse	Creamy	Brown	Powder	Round
Aspergillus sp.	Disperse	White	Brown	Cottony	Round
Aspergillus oryzae	Disperse	White	White	Powdery	Round

Thirty-seven isolates of fungi have been isolated from a total of fifty-five soil samples, out of which seventeen were entomopathogenic fungi and the remaining twenty were saprophytic fungi, identified by morphological and molecular confirmation. Among the isolated fungi, the genus *Aspergillus* was the richest (21) and has 4 species, *A. nomius, A. flavus, A. oryzae*, and *Aspergillus* sp. It is followed by *Beauveria* bassiana (4), *Fusarium solani* (3) and 1 isolate each of *Clonostachys rosea*, *Talaromyces pinophilus*, *Purpureocilium lilacinum*, and *Penicillium simplicissimum* and 5 unidentified fungal isolates. EPFs among these fungi are 4 isolates of *Beauveria bassiana*, 1 isolate of *Penicillium simplicissimum*, *Clonostachys rosea*, *Talaromyces pinophilus*, *Purpureocillium lilacinum*, 1 isolate of *Fusarium solani*, 1 isolate of *Aspergillus nomiae*, 6 isolates of *Aspergillus* sp and 1 isolate of *Aspergillus oryzae*.

Table 2 : Accession numbers of isolated entomopathogenic fungi

Fungi	Accession number	Location	Isolate	Geographical coordinates
Beauveria bassiana 1	OQ702348	Sivaganga	TNAU SGG 1 (Bb)	9.8433 N 78.4809 E
Beauveria bassiana 2	OQ695498	Ooty (Nilgiris)	TNAU OTC1 (Bb)	11.4102 N 76.6950 E
Beauveria bassiana 3	OQ696321	Denkanikottai (Krishnagiri)	TNAU DKT 1 (Bb)	12.5270 N 77.7899 E
Beauveria bassiana 4	OQ696320	Idayapatti (Madurai)	TNAU IDP 1 (Bb)	9.9384 N 78.2792 E
Clonostachys rosea	OQ703061	Ooty (Nilgiris)	TNAU OTD 1 (Cr)	11.4102 N 76.6950 E
Purpureocillium lilacinum	OQ831810	Kondayampalayam (Coimbatore)	TNAU KDP 1 (Pl)	11.5188 N 77.4343 E
Talaromyces pinophilus	OQ914391	Ooty (Nilgiris)	TNAU OTY 1 (Tp)	11.4102 N 76.6950 E
Penicillium simplicissimum	OQ696048	Ooty (Nilgiris)	TNAU OTE 1 (Ps)	11.4102 N 76.6950 E
Fusarium solani	OQ699212	Yercaud (Salem)	YCD 1 (Fs)	11.7748 N 78.2097 E
Aspergillus nomiae	OQ723456	Ooty (Nilgiris)	OTB 1 (An)	11.4102 N 76.6950 E
Aspergillus sp	OQ723454	Yercaud (Salem)	YCD 3 (As)	11.7748 N 78.2097 E
Aspergillus sp	OQ723470	Dindigul	DDG 1 (As)	10.3624 N 77.9695 E
Aspergillus sp	OQ832049	Ooty (Nilgiris)	OTB 1 (As)	11.4102 N 76.6950 E
Aspergillus oryzae	OQ723469	Dindigul	DDL 1 (Ao)	10.3624 N 77.9695 E

Infectivity rate

Pathogenicity test (Koch's postulates) results were summarized in Table 3. *Beauveria bassiana* isolates-TNAU SGG 1, TNAU OTC 1, TNAU DKT 1 and *Penicillium simplicissimum* isolate showed 100% mortality, followed by *Purpureocillium lilacinum* isolate and *Clonostachys rosea* isolate (93.75%). Isolates of genus *Aspergillus* showed mortality ranging from 31.25-87.5% but were not cultured further due to their detrimental health effects on humans.

Table 3 : Pathogenicity test on *Galleria mellonella* larvae (Koch's postulates)

S.No.	Fungal species	No. of Isolates	Days to death	Mortality* (%)
1	*Beauveria bassiana*	4	5-7	68.75-100
2	*Penicillium simplicissimum*	1	7	100
3	*Clonostachys rosea*	1	6	93.75
4	*Purpureocillium lilacinum*	1	6	93.75
5	*Talaromyces pinophilus*	1	7	75
6	*Fusarium solani*	1	6	56.25
7	*Aspergillus* sp	5	7-9	31.25-81.25
8	*Aspergillus nomiae*	1	6	87.5
9	*Aspergillus oryzae*	1	7	56.25

*Mean of mortality of 4 replications i.e., 60 larvae

Efficacy of *Beauveria bassiana* isolates against *Aphis gossypii*

Among all the four isolates of *B. bassiana* used, TNAU SGG 1 showed the highest mortality (96.67%) at 1×10^8 spores/ ml with LT_{50} of 4.44 days (Table 4) and LC_{50} of 0.14×10^5 spores/ ml (Table 5). It was followed by isolate TNAU OTC 1 with 85% mortality at the highest concentration. The lowest performance was recorded by isolate TNAU IDP 1 with 70% mortality at higher concentration with LT_{50} of 5.92 days. This proves that all the isolates of *B. bassiana* showed pathogenicity towards *A. gossypii* with isolate TNAU SGG 1 recording best performance.

Table 4 : Efficacy of *Beauveria bassiana* isolates against *Aphis gossypii*

Concentration (Spores/ ml)	TNAU SGG 1		TNAU OTC 1		TNAU DKT 1		TNAU IDP 1	
	Mortality (%)*	LT_{50} (Days)	Mortality (%)*	LT_{50} (Days)	Mortality (%)*	LT_{50} (Days)	Mortality (%)*	LT_{50} (Days)
1×10^5	66.67	6.86	63.33	7.28	61.67	7.90	53.33	8.88
1×10^6	73.33	6.09	66.67	6.79	70.00	6.96	56.67	8.45
1×10^7	81.67	5.32	75.00	5.93	73.33	6.73	66.67	7.26
1×10^8	96.67	4.44	85.00	5.72	78.33	5.92	70.00	7.02
Control	0.00	-	0.00	-	0.00	-	0.00	-

*Values are the mean of four replications

Table 5 : Median lethal concentration (LC_{50}) of *Beauveria bassiana* isolates

Fungal isolate	LC_{50} (Spores/ ml)
TNAU SGG 1	0.14×10^5
TNAU OTC 1	0.05×10^5
TNAU DKT 1	0.008×10^5
TNAU IDP 1	0.37×10^5

Entomopathogenic fungi against sucking pests of cucumber under protected conditions

The assessment of insect pest populations was performed until the first picking. Results indicated that farmers' practices recorded the lowest pest population at the time of picking (9.88 aphids/3 leaves/plant, 4.26 thrips/3 leaves/plant, and 6.92 red spider mites/2 cm^2 leaf/plant) (Table 6). IPM module was significant with farmer's practice with a pest population of 10.13 aphids/3 leaves/plant, 5.07 thrips/3 leaves/plant, and 7.08 red spider mites/2 cm^2 leaf/plant during the first trial and yield of 44.46 t/ha and 42.08 t/ha was obtained from Farmer's practice and IPM treatments. During the second trial, pest count of 8.33 aphids/3 leaves/plant, 7.52 red spider mites/2 cm^2 leaf/plant, and 4.62 whitefly/3 leaves/plant were recorded from FP treatment followed by IPM module (8.48 aphids/3 leaves/plant, 7.72 red spider mites/2 cm^2 leaf/plant, and 5.04 whitefly/3 leaves/plant). Farmer's practice recorded a yield of 35.23 t/ha and IPM module obtained 33.98 t/ha of fruit yield. A benefit-cost ratio of 2.94 and 3.22 was obtained during the first and second trials respectively for the IPM module which is higher than those of Farmer's practices (2.88 and 2.98).

Table 6 : Influence of IPM on sucking pests of cucumber

	Trial I			Trial II		
	Aphids	Thrips	Spider mite	Aphids	Spider mite	whitefly
IPM	10.13 (3.26)^a	5.07 (2.36)^{ab}	7.08 (2.75)^a	8.48 (3.00)^{ab}	7.72 (2.87)^a	5.04 (2.35)^{ab}
FP	9.88 (3.22)^a	4.26 (2.18)^a	6.92 (2.72)^a	8.33 (2.97)^a	7.52 (2.83)^a	4.62 (2.26)^a
Control	13.82 (3.78)^b	10.76 (3.36)^b	13.69 (3.77)^b	10.59 (3.33)^b	12.22 (3.57)^b	12.88 (3.66)^b

IPM – Integrated Pest Management, FP – Farmers' practice

Figures in parenthesis are $\sqrt{x + 0.5}$ transformed values.

Means followed by same superscript are not significantly different at the 5% level by DMRT

Discussion

The occurrence of entomopathogenic fungi in the soils of various agroecosystems is not fully studied, even though they are essential natural adversaries of insect pests (Sun and Liu 2008). Since soil is a primitive site for isolating entomopathogenic fungi, they are extensively distributed in a variety of soil types (Hajek 1997). Despite being considered agents for the biological control of insect pests for over a century, there is not much proof that backs the usage of fungi (Sujeetha and Sahayaraj 2014). The use of *Galleria mellonella* larvae brought about a significant detection of entomopathogenic fungi, indicating that the *Galleria mellonella* bait approach is a highly effective method for detecting entomopathogenic fungi in soil samples (Keller *et al*. 2003). Vivekanandhan *et al*. (2020) isolated forty-six strains of entomopathogenic fungi from the soils of the Eastern Ghats of South Indian Forest. Twenty *Beauveria* isolates have been identified by Senthilraja *et al*. (2010) from the Tamil Nadu groundnut soils. 54.17% of the entomopathogenic fungi were recovered from sixty-five Ethiopian soil samples by Gebremarium *et al*. (2021). Ahmed *et al*. (2022) isolated *Beauveria bassiana* from the soils of Egypt. According to Mukherjee *et al*. (2020), out of the forty fungal isolates isolated from the soils of North 24 Paraganas, a district in West Bengal, twenty isolates were determined to be pathogenic against *Aphis gossypii*. In the present study, *Beauveria bassiana*, *Penicillium simplicissimum*, *Clonostachys rosea*, *Purpureocillium lilacinum*, and *Talaromyces pinophilus* showed similar colony colour, texture, and shape of spores as reported by earlier workers (Visagie *et al*. 2014; Yilmaz *et al*. 2014; Anwar *et al*. 2018; Sun *et al*. 2021).

In the present study, entomopathogenic fungi detected using *Galleria mellonella* larva were *Aspergillus*, *Beauveria bassiana*, *Purpureocillium lilacinum*, *Clonostachys rosea*, *Penicillium simplicissimum*, *Talaromyces pinophilus* and *Fusarium solani*. These results were supported by Yakubu *et al.* (2022) who noted that *Aspergillus* sp has more occurrence (22.25%) followed by *Beauveria bassiana* and other unidentified fungi in the soils of Nigeria.

According to Tamta *et al.* (2022), *Clonostachys rosea* was 96.67% lethal to mango hopper. *Purpureocillium lilacinum* displayed 100% mortality against *Galleria mellonella* when Demicri *et al.* (2019) investigated its pathogenicity. When compared to *Penicillium simplicissimum* in our study, which displayed 100% mortality against *Galleria mellonella,* investigations of Baydar *et al*. (2016) and Rosa *et al.* (2022) showed that the pathogenicity of *Penicillium* sp against other insects was rather low.

Mortality studies describe the virulence and effectiveness of isolated fungi (Ignoffo *et al*., 1976). Four isolates of *B. bassiana* were tested for their efficiency in causing mortality of *A. gossypii*, and it was reported that isolate TNAU SGG 1 was the best performer with 96.67% mortality at the highest concentration followed by isolate TNAU OTC 1 (85%). Similar studies on the effectiveness of *B. bassiana* against *A. gossypii* were conducted by Jandricic *et al*., 2014; Erol *et al*., 2023.

Pest control was obtained both from farmers' practices and the IPM module during both trials. The findings underscore the observation that, while the FP yields the highest pest control and crop yield, the IPM module also delivers comparable pest control and yield results, alongside the added advantage of recording the highest cost-benefit ratio and reduced pesticide load in the field compared to farmer's practices. Considering the primary consumption of cucumbers in their raw form, the IPM treatment emerges as the optimal and sustainable choice for cucumber cultivation. Similar reports on the success of IPM were reported by Khooshe-Bast *et al*. (2016); Kumar *et al*. (2020); Mohsin *et al*. (2020); El Sayed and Ibrahim (2020) where IPM including entomopathogenic fungi reduced the pest population in cucumber.

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