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Research Article

Pharmacognosy for efficient natural therapy

Potential of Microbes Containing Formulations to Alter Growth and Phytochemicals of Medicinal Aromatic Plant- *Foeniculum Vulgare*

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Abstract: Sustainable plant production with the integration of eco-friendly agricultural practices, low chemical inputs, minimal deleterious effects on human health and low cost methods, is the need of the moment. In this direction, scientific/industrial community, continuously exploring novel and reliable methods. Plant growth promoting microbes are proving more promising to achieve eco-friendly and sustainable agricultural outcomes. In the present study, individuals of *Foeniculum vulgare*, a valuable aromatic plant species with potential medicinal value, were inoculated with different plant growth promoting microorganisms: (1) Arbuscular mycorrhizal like fungi (*Sebacina vermifera*) (2) Phosphate solubilizing bacteria (*Pseudomonas fluorescens*) (3) Azotobacter (*Azotobacter chroococcum*). Response of individual microorganism species was evaluated with reference to the emergence, plant growth and yield of essential oil along with qualitative effects on essential oil. Comparatively, significant response of *Sebacina vermifera,* in the stimulation of emergence of seeds, growth of plant and yield enhancement of essential oil was observed. An enhanced synthesis of anethole (major chemical constituent) was also recorded. Moreover, the enhancement in growth of plants was dependent on the extent of colonization percentage. A periodic study of growth parameters indicates plant's health and vitality influenced by *Sebacina vermifera*. The enhanced essential oil of seeds along with enhanced synthesis of anethole was in agreement with the assumption that *Sebacina vermifera* trigger defensive responses and hence improve phytochemical production. A mechanistic insight is also illustrated. In conclusion *Sebacina vermifera* possesses immense potentials in the pursuit of agro-ecological attributes of medicinal plant cultivation and crop production. It exerts excellent growth effects and enhances phytochemical production in medicinal plants.

Keywords: Anethole, Azotobacter, Essential oil, Fennel, Phosphate solubilizing bacteria, *Sebacina vermifera*

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1. INTRODUCTION

Fennel, *Foeniculum vulgare* Mill. belonging to family Umbelliferae (Apiaceae), is a hardy annual, biennial or perennial herb. Fennel is considered native to the Mediterranean areas and widely cultivated throughout the temperate and tropical regions of Northern, Eastern and Western hemispheres, specifically in Asia, North America, and Europe.[1-3] Further, it is a high value aromatic and medicinal herb used in various traditional systems of medicine like in Ayurveda, Unani, Siddha, Iranian and Chinese medicinal system.[4] Fennel is being used by human since antiquity for its traditional, contemporary applications and treatments of different ailments including insomnia, constipation, conjunctivitis, abdominal pain, cough, cold, hair growth, emesis, diarrhoea, stomach-ache, flatulence, arthritis, fever, indigestion also in addition it was used as diuretic, emmenagogue, galactagogue and anticancer agent.[3] Fennel is well known for its flavour and aroma. Various food preparation use fennel's different part as ingredients of different preparations including pickles, salted meat, fish, soups, sauces, salad, herbal tea, alcoholic beverages, bakeries, and many polyherbal formulations.[3,5,6] A number of biological studies involving extracts and isolated compounds of fennel has been evaluated for anti aging, antiallergic, anticarcinogenic, anti colitic, anti hirsutism, anti-inflammatory, antimicrobial, antimutagenic, antimycobacterial, antinociceptive, antioxidant, antipyretic, antispasmodic, antistress, antitumor, antiulcerogenic, antiviral, apoptotic, chemopreventive, cytoprotective, cytotoxic, hepatoprotective, hypoglycaemic, memory-enhancing, ocular hypotensive, oestrogenic, vascular effects.[3] The aromatic nature of seeds of fennel is due to the presence of essential oil (EO) which is a rich source of anethole, limonene, fenchone, estragole and camphene among them the anethole is the most important constituent with determinant role in quality of the EO of seeds.[7] The EO from aromatic plants has usually been isolated by either steam distillation or hydrodistillation.[1] The EO of fennel plays an important role in confectionary, perfumery, cosmetic and pharmaceutical industries.[8,9] EO composition depends upon internal factors like genetic, various growth stages and external factors like environmental and agricultural practices.[10,11] In our previous investigation, we conducted the first approach of arbuscular mycorrhiza (AM) like fungi (*Sebacina vermifera*) colonization with coriander. We found that *S. vermifera* extended the multifaceted benefits to plants including triggering growth enhancements and improvement in yield of EO. Furthermore, *S. vermifera* altered the composition of EO in a positive manner. *S. vermifera* enhanced the major chemical components including monoterpene alcohol (linalool) and monoterpene hydrocarbon (α-pinene and β-pinene) of coriander seeds.[12] In general, pure culture of arbuscular mycorrhizal fungi (AMF) not cultivable outside the host, but *S. vermifera* is a potential AM-like fungi which is cultivable under lab conditions.[13] AMF colonized plants may show increased biomass, faster growth rate & maturity, resistance to biotic and abiotic stresses, and more effective photosynthesis, enhanced yield and better composition of EO.[14-15] Mycorrhizal colonization influences the plant growth and alters the level of secondary metabolites which may depend on extent of root colonization by AMF.[16] Azotobacter and phosphate solubilizing bacteria are non symbiotic free living bacteria capable of fixing nutrients, produce phytohormones and synthesize antibiotics provide defense to plant against diseases.[17-18] Though there is enormous data on the yield and growth increase in medicinal and aromatic plants, comparatively only a few attempts have been made to establish the role of microorganisms in enhancing the productivity and growth of EO in fennel. Keeping the immense medicinal value of fennel in mind, the present study was carried out to find out whether the colonization of selected plant growth microorganisms could ameliorate emergence, growth, physiological activities, yield attributes and production of EO of fennel. In this regard, pot and field experiments were conducted. The effects of selected plant growth microorganisms were evaluated by emergence studies, growth studies and phytochemical studies.

2. MATERIALS AND METHODS

2.1 Microbes based formulations

Three types of microorganisms were applied in experimentation; (1) AM-like fungi (*S. vermifera*) (2) Azotobacter (*A. chroococcum)* (3) PSB (*P. fluorescens*). Here, *S. vermifera* was gifted by Prof. Virendra Swarup Bisaria (Department of Biochemical Engineering and Biotechnology, Indian Institute of Technology-Delhi, India). The culture was maintained on slants containing modified Kaefer-agar medium at a pH 6.5 and 30 ± 1 °C for 10 days and stored at 4 °C.[13] The fungal culture was grown in 500 mL Erlenmeyer flasks containing 100 mL of Modified Kaefer liquid medium, at 200 rpm using gyratory shaker.[19,20] The 100 mL of 8 day old culture was taken and 1 mL carboxy methyl cellulose (CMC) was added to it. 25 mL of CMC added culture was taken and added to 75 g of sterilized talcum powder.[21,22] This formulation was further used for experimentation. Azotobacter (*A. chroococcum*) and Phosphate Solubilizing Bacteria (*P. fluorescens*) were marketed biofertilizers, supplied by Ganesh Agro Service Centre, belonging to the local market of Moga, Punjab, India.

2.2 *Preparation of plant material*

Seeds of fennel were collected from Punjab Agriculture University, Ludhiana, during the rabi season (October-march). Seeds were pre-treated with 1% Savlon (Johnson and Johnson, USA), followed by 70% alcohol for 1 min. pre-treated seeds then washed thrice with sterile double-distilled water (SDDW). After this seeds of fennel were treated with 0.01% Bavistin (Saraswati Agro Chemicals Pvt. Ltd. India) for 2 min and washed 4-5 times with SDDW. Finally seeds were soaked in distilled water for 24 hour.[12]

2.3 Experimental design

Experimentation was carried under two different conditions *viz.* greenhouse condition and field condition. The physical properties of soil along with its chemical reserves are given in Table 1. The soil used in pot under greenhouse conditions was sterilized at 121 °C, three times for 1 h in 24 h intervals. The following treatments were used: (1) CON - without any microbes (2) AZOTO - *A. chroococcum* (3) PSB - *P. fluorescens* (4) SV - *S. vermifera.* Treatment scheme of all groups was the same throughout the experiment. Seeds were treated with formulation mixed with water as a paste (1 kg L^{-1}) and applied on the surface of seed (sufficient for 10 kg of seeds). After drying under shade seeds were sown. Plantlets were treated by dipping the roots in the paste and sown immediately. Soil treatment was carried by induction of 1 kg

formulation in 19 kg of vermicompost and mixed with soil in a ratio of 60:40 (soil: treated vermicompost). Whereas in field, treated vermicompost was spreaded as 60 kg per acre the same mixture was applied to established plants by drenching after every 8 week followed by irrigation.[12]

Table 1 Physical and chemical properties of soil

Sample	pH	Electrical conductivity (mmho/cm)	C (%)	P (kg/ha)	K (kg/ha)	Fe (kg/ha)	Zn (kg/ha)	Cu (kg/ha)	Mn (kg/ha)	N (%)
Pot	6.7	0.63	0.66	42.25	370.65	13.54	4.89	3.51	9.59	0.08
Field	7.4	0.59	0.48	32.37	311.35	12.26	4.10	1.53	7.36	0.06

Note: C-Carbon, P-Phosphorus, K-Potash, Fe-Ferrous, Zn-Zinc, Cu-Copper, Mn- Manganese, N-Nitrogen

2.4 *Pot studies*

The pot study was conducted under greenhouse condition facilitated with controlled environmental conditions *viz.* temperature (25 ± 2 °C), light intensity (1,000 Lux), relative humidity (70 %) with 16 h light and 8 h dark period. For the emergency response, total 100 seeds for each treatment group were taken. Each pot contains pre-treated soil sown with pre-treated seed, 2 cm below the surface and irrigation was carried out daily to avoid drought conditions. Day of sowing was considered a zeroth day. The emergence of shoot tips at the surface of soil was considered as germinated seed. Germinated seeds were counted after every 24 h until no more seed germinated.[12] For growth response the young and healthy 25 days old plants of similar size and of same physiological stage were taken. After treatment plantlets were planted on hard substratum of pre-treated soil in pots followed by proper irrigation. For each treatment a total of 100 plants were taken. Day of plantation was considered as zeroth day. The plants were randomly selected and uprooted after the interval of 30 days for analysis of plant growth parameters, first harvesting was done 30 days after transplantation (DAT) followed by 60 DAT, 90 DAT and final harvesting was done on 120 DAT. Before uprooting, pots were moistened enough so that the whole plants were easily uprooted. Uprooted plants were washed under streams of water. Experimentation was based on complete randomized design (CRD). Sample size from each replicate was 6 plants.[12]

2.5 Field studies

The field study was conducted in agricultural fields under natural conditions. For the emergency response, total 100 seeds for each treatment group were taken. Seed beds were made fine porous by ploughing and planking followed by irrigation. Pre-treated seeds were sown in pre-treated soil, 2 cm below the surface and irrigation was carried out daily. Day of sowing was considered a zeroth day. The emergence of seed radicals at the surface of soil was considered as germinated seed. Germinated seeds were counted after every 24 h until no more seed germinated. For growth response the young and healthy 25 days old plants of similar size and of same physiological stage were taken. After treatment plantlets were planted in pre-treated soil under field conditions followed by proper irrigation. For each treatment a total of 100 plants were taken. Day of plantation was considered as zeroth day. The plants were randomly selected and uprooted after the interval of 30 days for analysis of plant growth parameters, first harvesting was done on 30 DAT (Day after transplantation) followed by 60 DAT, 90 DAT and final harvesting was done on 120 DAT. Before uprooting, the field was moistened enough so that the whole plants were easily uprooted. Uprooted plants were washed under streams of water. Sample size from each replicate was 6 individual plants.[12] All field experiments were done using randomized complete block design (RCBD). A complete field topology and sowing scheme is given in Fig. 1, 2.

Emergence response for both pot and field trials was evaluated using following parameters:

a) Germination percentage[23]
b) Mean germination time[24,25]
c) Germination index[26]
d) T_{50} of germination[27-29]
e) Seed vigour[30]
f) Vigour index[31]
g) The uprooted plants from both pots and field were observed for the following parameters:
a) Total plant length (root+shoot)
b) Total fresh weight of plant (root+shoot)
c) Total dry weight of the plant (root+shoot)

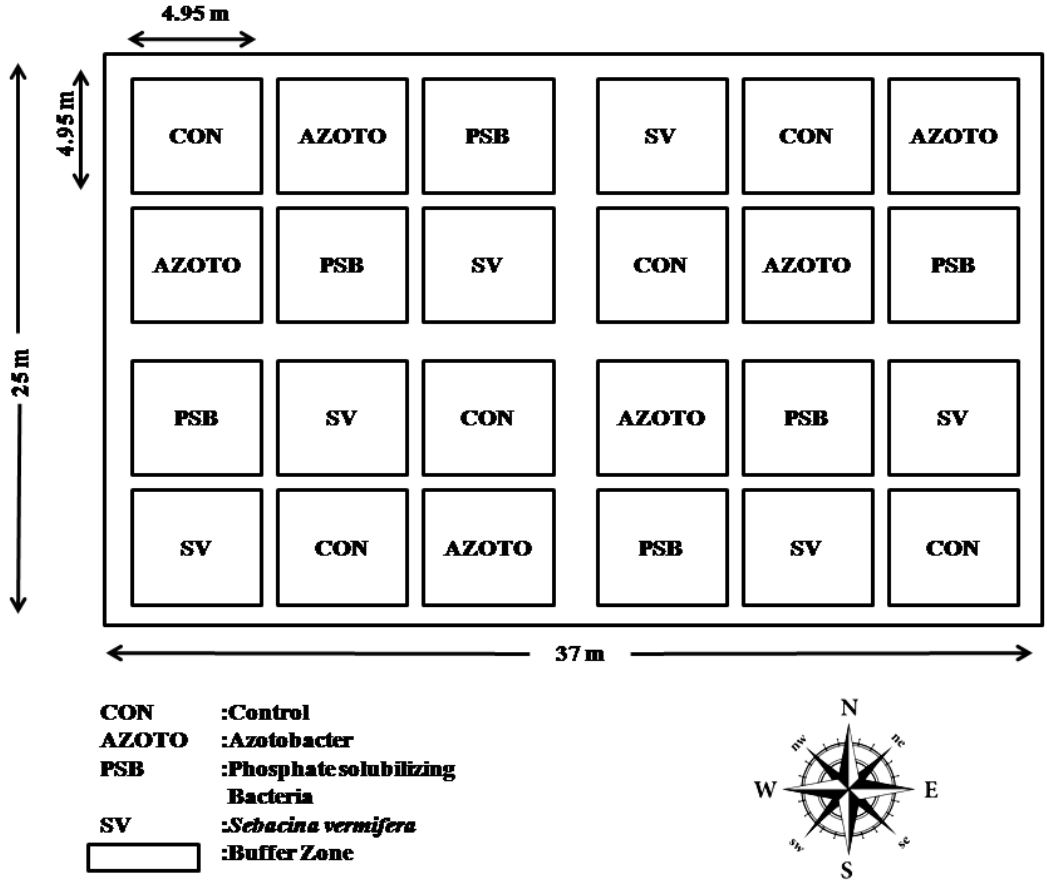

Fig 1 Topology of field experiments (Randomized complete block design)

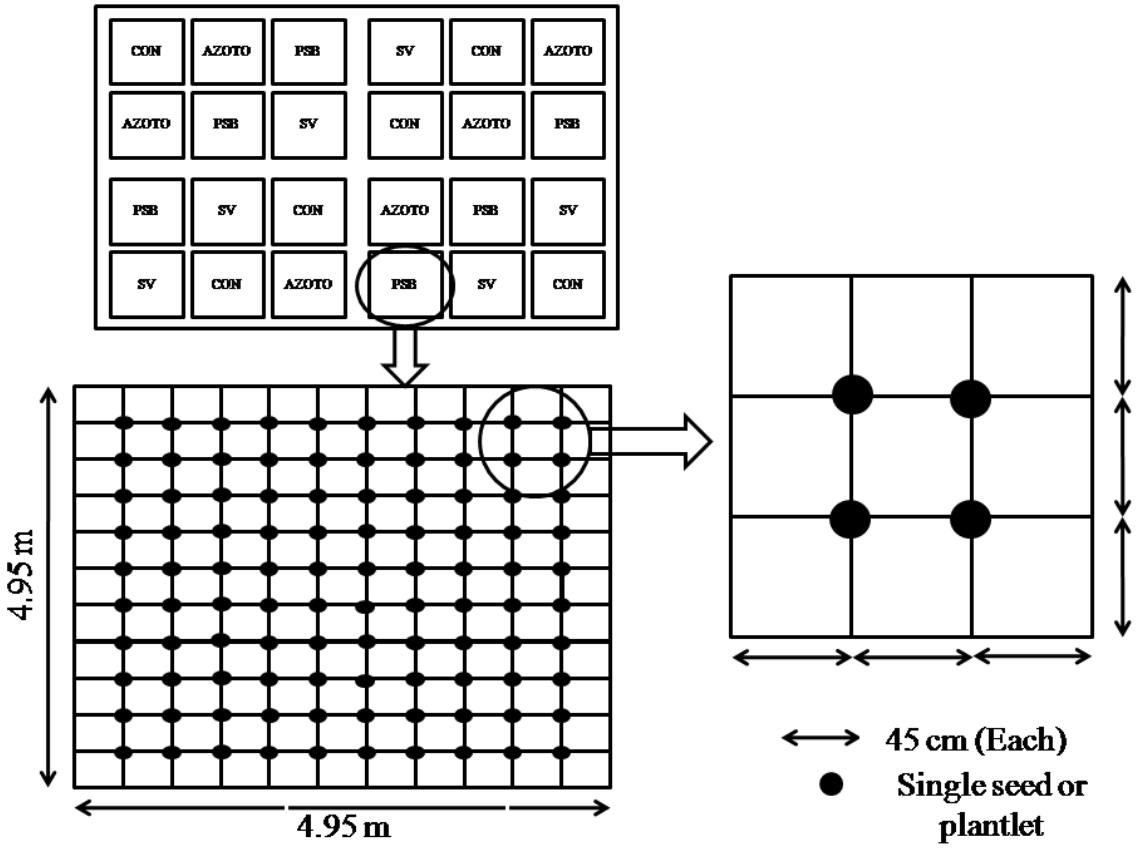

Fig 2 Seeds and plantlets sowing scheme

2.6 Root colonization studies

Sample roots of SV treated plants were collected at 30 DAT, 60 DAT, 90DAT and 120DAT. After proper washing, roots were cut into 1cm segments. All segments were boiled with 10% potassium hydroxide for 15 min. followed by neutralization with 1 N hydrochloric acid for 3-4 min. After proper washing segments were stained with 0.5% trypan blue overnight and mounted in a lactoglycerol solution, using the slightly modified method given by Phillips and Hayman (1970).[32] Segments were selected randomly and examined under light microscope. Percentage of colonization was assessed using following formula: [33]

$$\text{Percentage colonization} = \frac{\text{Number of root segments colonized}}{\text{Number of total segments examined}} \times 100$$

2.7 Phytochemical studies

2.7.1 Oil extraction

The sample seeds were grounded first to a course powder and subjected to hydrodistillation using a Clevenger apparatus for 4 hours. The collected EO was dried over anhydrous sodium sulphate and stored at 4 °C until further analysis.

2.7.2 Gas chromatography-mass spectrometry

The GC-MS analyses were performed on a gas chromatograph TRACE ™ 1300 GC equipped with TSQ™ 8000 Evo MS with electron impact ionization. A TG-5MS capillary column (30 m × 0.25 mm, 0.25 µm film thickness) was used. The column temperature was programmed to rise from 50 to 280 °C at a rate of 10 °C min^{-1}. The carrier gas was Helium with a flow rate of 1 mL min^{-1}. Mass range was 50-550 m/z, respectively. The injected volume was 1 µL and the total run time was approximately 30 min. Identification of the oil constituents was based on the comparison of their retention indexes relative to C8-C22 n-alkanes with those in the literature or with those of authentic compounds available in laboratory. Further identification was made by matching their recorded spectra with those stored in the National Institute of Standards and Technology (NIST) mass spectral library of the GC-MS data system. The percentage peak area of corresponding component was taken as concentration without using correction factors.[34]

3. STATISTICAL ANALYSIS

All experiments were conducted in six replicates. Sample size from each replicate was 6. In phytochemical studies, GC-MS was performed with 3 replicate (total three samples from each group). The data was expressed as mean ± standard deviation (SD). Means from emergence responses, yield of EO and composition of various components of EO were compared by using the one way-analysis of variance (ANOVA) followed by Tukey's post hoc test. Means from growth response were compared by using two way-ANOVA followed by Bonferroni post test. Individual means were deemed to be significant at $p<0.05$. All statistical analysis was performed using the GraphPad Prism v6 software package.

4. RESULTS

4.1 Root colonization

Both pot and field trials showed a positive interaction with *S. vermifera.* Microscopic study revealed normal healthy fungal hyphae. Under both conditions the percentage of association was found lowest at 30 DAT, whereas on subsequent harvesting i.e. 60 DAT and 90 DAT it was found in increasing fashion. The highest percentage of colonization was observed on 120 DAT under both conditions Table 2.

Table 2 Colonization percentage of *Sebacina vermifera* with fennel roots at different time periods in pot and field trials

Harvesting	Pot (Mean±SD)	Field (Mean±SD)
30 DAT	25.21±0.89	12.12±0.62
60 DAT	42.34±1.46	28.31±1.12
90 DAT	68.61±2.23	46.06±1.82
120 DAT	88.91±3.12	82.23±2.43

Root colonization (%) of Sebacina vermifera treated plants: data expressed as mean ± SD of six replicates; DAT- Day after transplantation.

4.2 Pot studies

4.2.1 Emergence response

Emergence studies on fennel seeds were observed for 23 days from the day of sowing of seeds. Different analytical traits *viz.* germination percentage, mean germination time, germination index, T_{50} of germination, seedling vigour and vigour index were investigated carefully. All traits revealed that different treatments exerted significant effects on emergence of seeds under pot trials (Table 3). Results of emergence studies under pot conditions have been presented in Table 3. Data shows that all treatments viz. SV, PSB and AZOTO outperformed CON with respect to all the measured emergence parameters. Among the tested plant growth promoting microbes, SV showed best results followed by PSB and AZOTO whereas SV underperformed here. As compared to CON, germination percentage, germination index, seedling vigour and vigour index for SV were improved by 25%, 9%, 50.7% and 88.9%, respectively. For PSB, these were improved by 20%, 7%, 44.9% and 74%, respectively while for AZOTO, these factors were improved by 17.7%, 4%, 40.9% and 65.9%, respectively. On the other hand, mean germination time was reduced by 10.9%, 8.8% and 1%, respectively for SV, PSB and AZOTO. Similarly, T_{50} germination was reduced by 17.8%, 10.7% and 1% for SV, PSB and AZOTO, respectively (Table 3).

Table 3 Effects of various treatments on fennel seeds under pot trials				
Parameters	**CON**	**AZOTO**	**PSB**	**SV**
Germination percentage (%)	79.00±3.00	93.00±2.00^{a}	95.00±3.00^{a}	99.00±3.00^{a,b}
Mean germination time (days)	13.73±0.64	13.59±0.45	12.51±0.49^{a,b}	12.23±0.60^{a,b}
Germination index	25.85±1.48	26.93±1.35	27.73±1.40	28.24±1.30^{a}
T_{50} germination (Days)	13.81±0.50	13.64±0.40	12.32±0.35^{a,b}	11.35±0.30^{a,b,c}
Seedling vigour (mm)	81.60±3.00	115.00±3.00^{a}	118.30±2.00^{a}	123.00±3.00^{a,b,c}
Vigour index	6446.40±204	10695.00±250^{a}	11238.50±280^{a,b}	12177.00±296^{a,b,c}

Emergence response of seeds to various treatments in pot trials: data expressed as mean ± SD of six replicates. Superscripts with different letters (a-c) within the same row represent significance level as p^{a}<0.05 v/s CON; p^{b}<0.05 v/s AZOTO; p^{c}<0.05 PSB; CON- Normal control; AZOTO- Azotobacter; PSB- Phosphate solubilizing bacteria; SV- S. vermifera

4.2.2 Growth response

Effect of tested plant growth promoting microbes on the growth profile of fennel was also evaluated to understand their role in physiological development of fennel plants. Growth profiles were measured in terms of total plant length (root+shoot), total fresh weight (root+shoot) and total dry weight (root+shoot). All the treatments as mentioned in the materials and methods were applied respectively to soil and plants. Statistical significance of observed effects was also established by using statistical techniques. Finally, it was found that all the treatments resulted in significant growth enhancement in pot trials. During pot studies, all the three treatments (SV, AZOTO and PSB) showed positive effect when compared with control (CON) experiments. SV was found to have the most prominent effect on plant height improvement which was followed by PSB and AZOTO, respectively (Fig. 3, 4a, b). However, when relative growth rates (for plant height) were compared, all SV, PSB and AZOTO showed significantly better results when compared with CON experiments. Growth rates (plant height) of SV treated plants were found to be maximum among all treatment groups at 30, 60, 90 and 120 DAT. Similar trends were observed when plant growth was measured in terms of total fresh weight (Fig. 4c, d) and total dry weight (Fig. 4e, f). As compared to CON, SV treatment resulted in a maximum of 211.8%, 227% and 555% improvement in total plant length, fresh weight and dry weight, respectively. In case of PSB the maximum improvement was 138%, 142% and 345% in total plant length, fresh weight and dry weight, respectively and AZOTO improved the total plant length, fresh weight and dry weight by 67%, 73% and 175%, respectively. In pot conditions, maximum growth rate for plant length was observed in between 30 - 90 days whereas it declined after 90 days for plant fresh weight and dry weight (Fig. 4).

Table 4 Effects of various treatments on fennel seeds under field trials				
Parameters	**CON**	**AZOTO**	**PSB**	**SV**
Germination percentage (%)	39.00±1.00	40.00±2.00	80.00±3.00^{a,b}	99.00±2.00^{a,b,c}
Mean germination time (days)	14.15±0.40	13.53±0.60	13.04±0.40^{a}	12.35±0.50^{a,b}
Germination index	22.82±1.80	23.01±1.30	26.27±1.40^{a,b}	28.11±1.20^{a,b}
T_{50} germination (Days)	15.50±0.55	14.14±0.50^{a}	13.50±0.60^{a}	12.92±0.46^{a,b}
Seedling vigour (mm)	30.00±2.00	63.00±2.00^{a}	81.60±3.00^{a,b}	114.00±4.00^{a,b,c}
Vigour index	1170.00±150	2520.00±198^{a}	6528.00±210^{a,b}	11286.00±250^{a,b,c}

Emergence response of seeds to various treatments in field trials: data expressed as mean ± SD of six replicates. Superscripts with different letters (a-c) within the same row represent significance level as p^{a}<0.05 v/s CON; p^{b}<0.05 v/s AZOTO; p^{c}<0.05 PSB; CON- Normal control; AZOTO- Azotobacter; PSB- Phosphate solubilizing bacteria; SV- S. vermifera

Fig 3 Plants under pot trials at 120 DAT (a) Control (b) *Azotobacter* (c) Phosphate solubilizing bacteria (d) *S. vermifera*

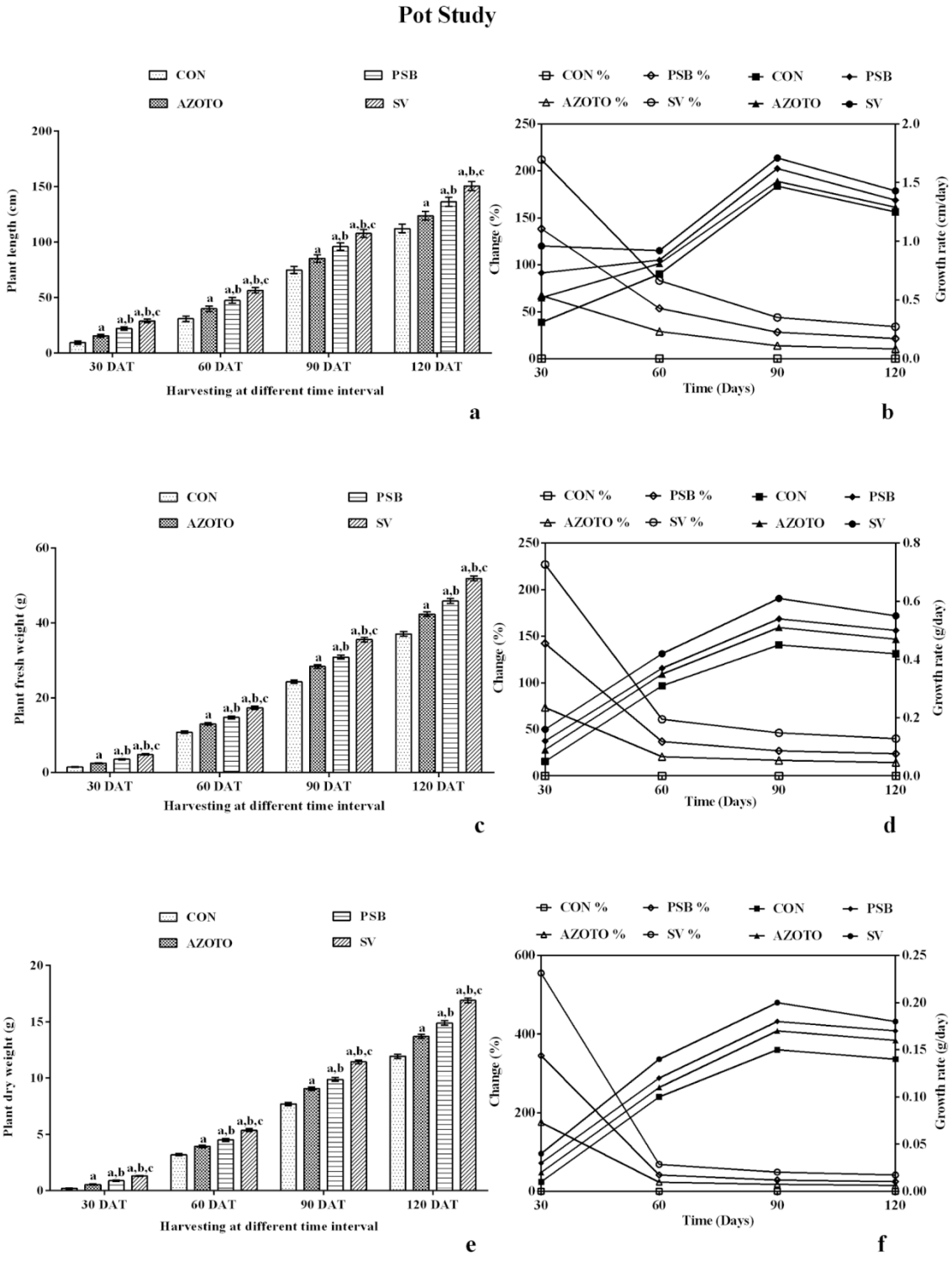

Fig 4 Effect of treatments on (a, b) Total length of plants (c, d) Total fresh weight of plants (e, f) Total dry weight of plants under pot conditions; data expressed as mean ± SD of six replicates. Superscripts with different letters (a-c) within the same harvesting group represent significance level as p^a<0.05 v/s CON; p^b<0.05 v/s AZOTO; p^c<0.05 v/s PSB; CON- Normal control; AZOTO- *Azotobacter*; PSB- Phosphate solubilizing bacteria; SV- *S. vermifera*

4.3 Field studies

4.3.1 Emergence response

The relative effect of all three plant growth promoting microbes (SV, AZOTO and PSB) during field study were in alignment to what we observed during pot studies. Among all the four tested groups (CON, SV, AZOTO and PSB) under field conditions, all three treatment groups viz. SV, PSB and AZOTO significantly altered the emergence profile of all the traits. Treatment with SV resulted in maximum effect followed by PSB and AZOTO. Here, germination percentage, germination index, seedling vigour and vigour index for SV were improved by 153.8%, 23%, 280% and 864.6%, respectively when compared with CON. Similarly, with PSB, these factors were improved by 105%, 15%, 172% and 457.9%, respectively. With AZOTO, respective improvements of mentioned parameters were 2.56%, 0.83%, 110% and 115%. On the other hand, mean germination time was reduced by 12.7%, 7.8% and 4% for SV, PSB and

AZOTO, respectively. Similarly, T_{50} germination was reduced by 16.6% 12.9% and 8.7% for SV, PSB and AZOTO, respectively in comparison to CON (Table 4).

4.3.2 Growth response

Once we got encouraging results at pot scale, we extended this study to field scale to validate the applicability of lab scale data in actual field conditions. Similar growth responses were observed with experiments done in pot conditions and data was correlated well with pot scale experimental data. Again, all the three treatments (SV, AZOTO and PSB) showed significant positive effect when compared with CON experiments. However, best results were observed in experiments with SV, which was followed by PSB and AZOTO, respectively (Fig. 5, 6).

Fig 5 Plants under field trials at 60 DAT (a) Control (b) *Azotobacter* (c) Phosphate solubilizing bacteria (d) *S. vermifera*

Trends were similar to pot responses when plant growth was measured in terms of total length of plant (Fig, 6 a, b), total fresh weight (Fig. 6c, d) and total dry weight (Fig. 6e, f). As compared to control, SV treatment resulted in a maximum of 203.8%, 2212% and 600% improvement in total plant length, fresh weight and dry weight, respectively. PSB improved total plant length, fresh weight and dry weight by 134.9%, 139%, 380%, respectively and AZOTO improved total plant length, fresh weight and dry weight by 65.7%, 67%, 180%, respectively. In field conditions also, maximum growth rate in plant length was observed during the first 60-90 days whereas it was declined after 90 days for plant fresh weight and dry weight (Fig. 6).

4.3.3 Yield of essential oils

It was found that yield of essential oil was effectively altered by different treatments viz. AZOTO, PSB and SV. Although all treatments enhanced the yield of EO but SV treatments enhanced the yield prominently. Enhanced yield of EO by SV was found maximum (pot 2.8%; field 2.5%), whereas the enhanced yield by PSB was found as 2.4% in both pot & field relative to CON. Enhanced yield by AZOTO was 2.4% & 2.3% in pot and field, respectively relative to CON. The Yield of CON group was found minimum (pot 1.9% & field 1.8%) in comparison to all other groups. Under both conditions (pot & field) all treatments significantly enhanced yield when compared to CON whereas enhanced yield by SV was found significantly higher when compared to PSB and AZOTO. PSB and AZOTO enhanced yield of EO equally under pot conditions whereas yield enhancement by PSB was more than AZOTO under field condition (Fig. 7)

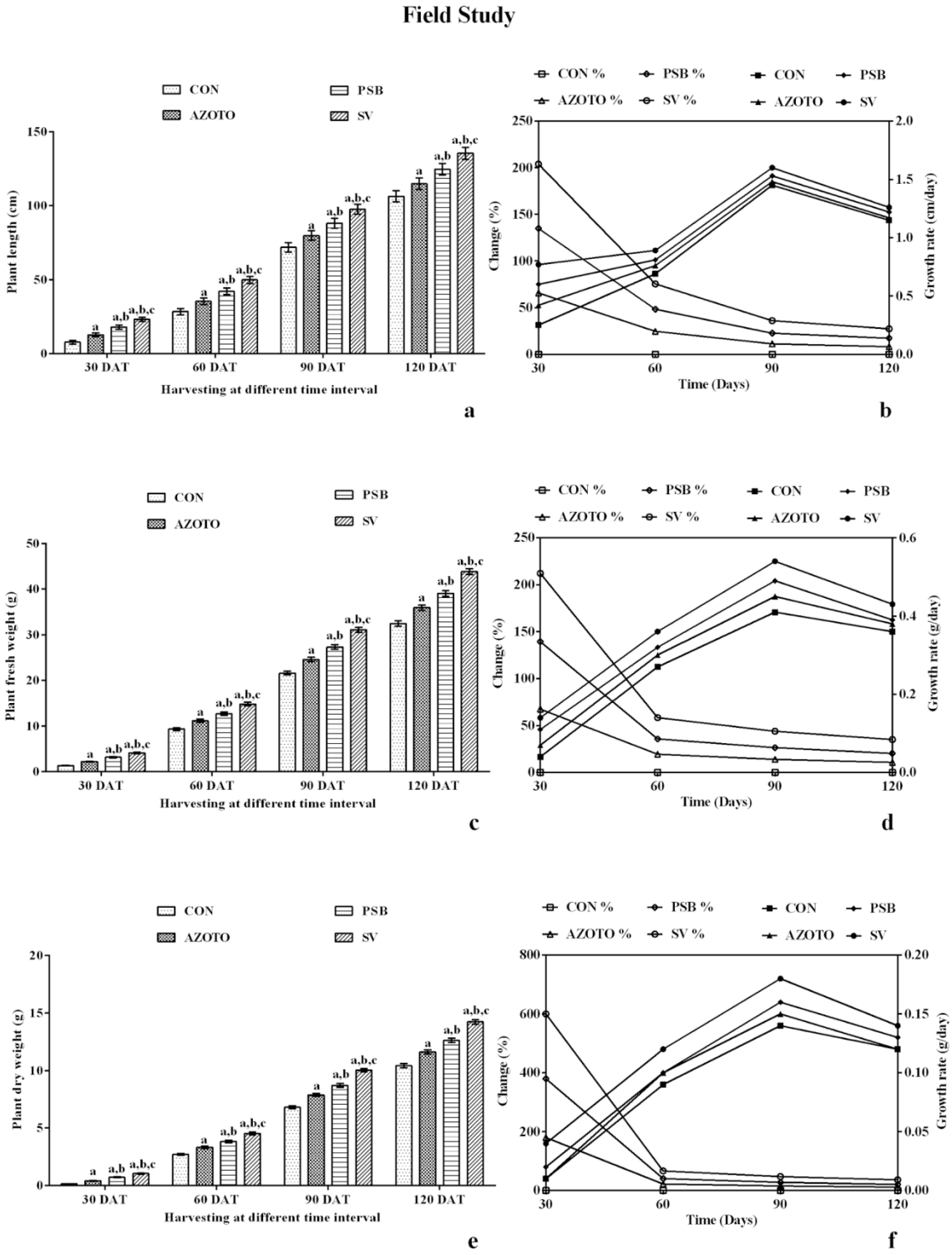

Fig 6 Effect of treatments on (a, b) Total length of plants (c, d) Total fresh weight of plants (e, f) Total dry weight of plants under field conditions; data expressed as mean ± SD of six replicates. Superscripts with different letters (a-c) within the same harvesting group represent significance level as p^a<0.05 v/s CON; p^b<0.05 v/s AZOTO; p^c<0.05 v/s PSB; CON- Normal control; AZOTO- *Azotobacter*; PSB- Phosphate solubilizing bacteria; SV- *S. vermifera*

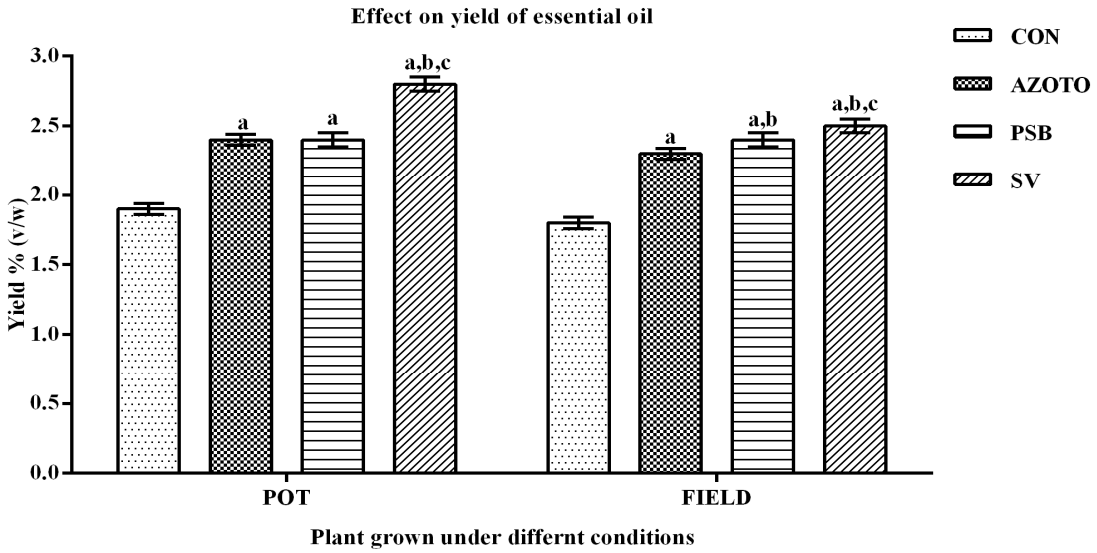

Fig 7 Effect of treatments on yield of EO of plant seeds under pot and field conditions: data expressed as mean ± SD of six replicates. Superscripts with different letters (a-c) within the same condition group represent significance level as p^a<0.05 v/s CON; p^b<0.05 v/s AZOTO; p^c<0.05 v/s PSB; CON- Normal control; AZOTO- *Azotobacter*; PSB- Phosphate solubilizing bacteria; SV- *S. vermifera*

4.3.4 Chemical composition of EO of fennel under pot condition

The effect of different treatments in pots under greenhouse conditions, on composition of the EO of fennel is given in Table 5. GC-MS analysis of EO resulted in the identification of 16 constituents, representing 99.99-100% of the oils. Significant changes (p<0.05) were observed in the chemical profiles of EO. Nevertheless, anethole was the major constituent (46.5-59%) followed by limonene (11-23%), fenchone (11.8-14.5%), estragole (6.9-9%), α-pinene (2.8-6%) relative to the composition of EO. In contrast, the relative percentage of individual components varied significantly (p<0.05) with different treatments. Anethole was the major compound of EO comparative to all treatments and reached a maximum of 59% in SV treated groups. PSB and AZOTO also recorded with the Anethole 54% and 51%, respectively, whereas CON group was recorded with 46.5% only. Fenchone was also recorded with an enhancement. But AZOTO enhanced the fenchone to maximum 14.5% whereas SV and PSB recorded with 12% and 11.8%, respectively. CON was recorded with the lowest percentage 10%. It was observed that treated groups uptriggerd the selective components which were anethole, fenchone, estragole, fenchyl acetate, *p*-anisaldehyde and down triggered some components which were α-pinene, camphene, α-phellandrene, α-myrcene, o-cymene and limonene. It was also observed that β-pinene was selectively synthesized by only CON group further β-thujene & ϒ-terpinene by PSB group whereas, linalool and camphor were synthesized by treated groups only and found absent in CON group.

Table 5 Effect of different treatments on composition of various EO components of fennel seeds grown in pots under greenhouse conditions

Components	CON	AZOTO	PSB	SV
α-Pinene	6.43±0.13	2.89±0.22^a	3.58±0.45^{a,b}	3.10±0.14^{a,c}
Camphene	1.00±0.17	0.44±0.02^a	0.49±0.08^a	0.52±0.08^a
α-Phellandrene	0.87±0.02	0.61±0.02^a	0.74±0.06^{a,b}	0.48±0.07^{a,b,c}
β-pinene	0.38±0.02	-	-	-
α-Myrcene	2.10±0.19	0.59±0.03^a	0.78±0.16^a	0.79±0.09^a
β-Thujene	-	-	0.33±0.06	-
o-Cymene	0.58±0.03	0.68±0.02^a	0.64±0.08	0.41±0.07^{a,b,c}
Limonene	23.38±0.45	13.66±0.50^a	13.05±0.55^a	11.02±0.28^{a,b,c}
ϒ-Terpinene	-	-	0.39±0.06	-
Fenchone	10.23±0.04	14.53±0.68^a	11.88±0.63^{a,b}	12.24±0.97^{a,b}
Linalool	-	0.25±0.02	0.30±0.05^b	0.27±0.01
Camphor	-	0.72±0.02	0.46±0.04^b	0.43±0.04^b
Estragole	6.97±0.02	9.08±0.66^a	9.18±0.38^a	8.56±0.26^a
Fenchyl acetate	0.59±0.04	1.65±0.05^a	1.44±0.14^{a,b}	1.09±0.04^{a,b,c}
p-Anisaldehyde	0.93±0.04	3.57±0.03^a	2.71±0.20^{a,b}	2.03±0.02^{a,b,c}
Anethole	46.54±0.58	51.32±0.92^a	54.01±1.69^{a,b}	59.06±1.05^{a,b,c}

Data expressed as mean ± SD of three replicates: Superscripts with different letters (a-c) within the same row represent significance level as p^a<0.05 v/s CON; p^b<0.05 v/s AZOTO; p^c<0.05 v/s PSB; CON- Normal control; AZOTO- Azotobacter; PSB- Phosphate solubilizing bacteria; SV- S. vermifera

4.3.5 *Chemical composition of EO of fennel under field condition*

The effect of different treatments under field conditions, on composition of the EO of fennel is given in Table 6.GC-MSanalysis of EO resulted in the identification of 16 constituents, representing 99.99-100% of the oils. It was observed that under field conditions individual components were found slightly different than pot conditions. Significant changes (p<0.05) were observed in the chemical profiles of EO. Like pot condition here also anethole was the major constituent (43-57.8%) followed by limonene (9.7-22%), fenchone (8.5-12.7%), estragole (7-8.9%), α-pinene (3-6%) relative to the composition of EO. In contrast, the relative percentage of individual components varied significantly (p<0.05) with different treatments. Anethole was the major compound of EO comparative to all treatments and reached a maximum at 57.8% in the SV treated group. PSB and AZOTO also recorded with the anethole 53% and 47.8%, respectively, whereas CON group was recorded with 43% only. Fenchone was also recorded with an enhancement. But AZOTO enhanced the fenchone to maximum 12.7% whereas SV and PSB recorded 11.6% and 10.5%, respectively. CON was recorded with lowest percentage 8.5%. It was observed that like pot condition here also treated groups uptriggerd the selective components which were anethole, fenchone, estragole, fenchyl acetate, *p*-anisaldehyde, α-phellandrene o-cymene and down triggered some components which were α-pinene, camphene, α-myrcene, and limonene. It was also observed that β-pinene was selectively synthesized by only CON group further β-thujene & Y-terpinene by PSB group whereas, linalool and camphor were synthesized by treated groups only and found absent in CON group.

Table 6 Effect of different treatments on composition of various EO components of fennel seeds grown under field conditions

Components	CON	AZOTO	PSB	SV
α-Pinene	6.23±0.11	3.67±0.24[a]	3.24±0.43[a,b]	3.20±0.16[a,b]
Camphene	2.88±0.19	1.42±0.04[a]	1.46±0.10[a]	1.51±0.06[a]
α-Phellandrene	0.84±0.04	1.53±0.04[a]	1.24±0.04[a,b]	0.52±0.09[a,b,c]
β-pinene	0.34±0.04	-	-	-
α-Myrcene	3.98±0.17	1.52±0.05[a]	1.21±0.14[a,b]	1.71±0.11[a,c]
β-Thujene	-	-	1.25±0.08	-
o-Cymene	1.56±0.01	1.64±0.04	1.12±0.06[a,b]	0.35±0.09[a,b,c]
Limonene	22.21±0.47	11.57±0.48[a]	11.36±0.57[a]	9.72±0.26[a,b,c]
Y-Terpinene	-	-	1.31±0.04	-
Fenchone	8.52±0.06	12.76±0.66[a]	10.53±0.65[a,b]	11.60±0.95[a,b]
Linalool	-	1.26±0.04	1.25±0.03	0.24±0.03[b,c]
Camphor	-	1.71±0.05	1.41±0.06[b]	0.43±0.02[b,c]
Estragole	7.20±0.04	8.93±0.68[a]	7.60±0.36[b]	8.08±0.28[a,b]
Fenchyl acetate	1.53±0.06	2.61±0.03[a]	1.46±0.16[b]	1.04±0.02[a,b,c]
p-Anisaldehyde	1.60±0.02	3.49±0.05[a]	2.36±0.18[a,b]	3.71±0.04[a,b,c]
Anethole	43.11±0.60	47.88±0.90[a]	53.20±1.71[a,b]	57.89±1.03[a,b,c]

Data expressed as mean ± SD of three replicates: Superscripts with different letters (a-c) within the same row represent significance level as p^a<0.05 v/s CON; p^b<0.05 v/s AZOTO; p^c<0.05 v/s PSB; CON- Normal control; AZOTO- Azotobacter; PSB- Phosphate solubilizing bacteria; SV- S. vermifera

5. DISCUSSION

Microscopic study after staining technique revealed colonization in the form of normal and healthy fungal hyphae spread through the root surface to epidermal and cortical cell layers. This confirmed the presence of *S. vermifera* inside the inoculated fennel roots. This is in agreement with previous findings with fennel roots.[35] A number of observations also support the potential of *S. vermifera* to colonize with a wide range of plant species i.e. *Nicotiana attenuata*,[36]*Thymus vulgaris*,[37] Brassicaceae plants,[38]*Panicum virgatum*,[39,40]*Oryza sativa.*[41] Azotobacter and PSB both enhanced the emergence trait of fennel in a positive way under both pot and field trials. Similar observations were made with *A. chroococcum* treated *Dodonaea viscose* [42]and *P. fluorescens* treated *Vicia faba*.[43] Our observation was in agreement with studies reported the germination enhancing capabilities of *A. chroococcum*[44] and *P. fluorescens*.[45-48] Although, underlying mechanisms are still unknown, but it is conceivable that he enhancement in emergence traits could be accredited by the phytohormones and other complex mixture of biological active compounds and growth promoting metabolites that are modulated by *A. chroococcum*,[49-51]*P. fluorescens*.[52,53] Here *S. vermifera* exerted a prominently high influence on the emergence trait of fennel seeds. Our observations were in close agreement with previous observations that *S. vermifera* enhanced the germination of *P. virgatum* plant in petri plates and also with the stimulation of the seed germination, increase growth & stalk elongation along with earlier flowering, more flower yield and greater maturation of seed capsules in *Nicotiana attenuata*.[36,54] Contrary to this AMF can alter emergence traits negatively while has the tendency to still improve growth on later growth stages.[55] Also preliminary investigations suggested that AMF has a negative effect on seed germination of *Striga hermonthica*.[56]A recent study involving the seeds of *Cynorkis purpurea* sown with *S. vermifera*, also favour the stimulatory behaviour of AMF to emergence traits.[57] It is speculated that the effect of *S. vermifera* is directly bound to seed species and peculiarities of applications and experimental designs. Demonstration by Maighal et al. (2016)[58] support our observation and suggested that two factors "Mycorrhiza" and "seed species" are responsible for seed viability and thus germination. The inhibition of germination appears to be due response of competition between viable fungal spores and seed for available oxygen.[59] Along with this Lendzemo et al. (2007)[60] suggested that inhibition of seeds after AMF colonization may

be a consequence of formation of unwanted metabolites, inhibitions of required metabolites and negative mycorrhizosphere effects. Overall emergence traits were excellent in pot trials in comparison to field trials. It is likely external harsh conditions prevented the threshold metabolic process and proliferation of embryonic tissues thus affected the emergence traits. While, favourable and controlled conditions lead the better emergence traits under greenhouse conditions. The morphological indices i.e. total plant length, total fresh weight and total dry weight shows that microorganism treated fennel plants exhibit excellent growth performance as compared to control. Similar findings, representing the growth enhancing features of *A. chroococcum,*[61,62] *P. fluorescens* [63,64] and *S. vermifera*.[41,54] A considerable increase in length of plant was recorded with *S. vermifera* inoculated *F. vulgare* plants [35]. Similarly, the *S. vermifera* treated *T. vulgaris* resulted in substantial increase in fresh weight and dry weight [37]. *S. vermifera* treated *Mentha piperita* and Brassicaceae plants also recorded with enhanced Plant length, enhanced fresh weight, dry weight, enhanced root system and number of nodes.[38,65] Indeed higher expression of genes,[66] and phytohormonal involvements[67] are responsible for development. Further, *A. chroococcum, Pseudomonas* species and AMF (*P. indica*- closely related to *S. vermifera*) has been assessed for phytohormonal modulations.[67-71] Moreover, the growth-promoting effects could also be due to up regulation of nutrient uptake especially nitrogen and phosphorus from the soil, this could affect biomass allocation & accumulation, pronounced growth of plant and good quality and enhanced yield of seeds.[72] Although, *A. chroococcum* has potential to fix nitrogen to soil[73,74] and *Pseudomonas* genera is well known as phosphate solubilizing bacteria[75] but *S. vermifera* has capabilities to fixing and efflux large no. of macro- and micro-nutrients including nitrogen and phosphorus.[40,54] Further, elemental analysis of C, H, N from dry matter of plants confirms the enhanced nutrients uptake and mass accumulation (Data is not shown here). Notably *S. vermifera* treated plants significantly accumulated essential elements which indeed contributed for the enhanced biomass when compared to all other treatments. It is tempting to speculate that the better and pronounced growth effects of *S. vermifera* are due to integrated efforts of exploration of large volumes of soil by fungal hyphae and fixation of essential macro- and micro-nutrients. Treated plants tended to increase the EO to a certain percent. In support of our investigations there are some studies which suggested that *A. chroococcum*,[76-81] *P. fluorescens* [82-84] and *S. vermifera* [35,37] could stimulate the synthesis of EO and its composition. Although the exact mechanism behind the enhancement of EO is still uncovered however increases EO may be consequence of defensive response [83, 85] and could be associated with morphological traits,[87,88] genes of biosynthetic pathway[88-90]and P availability.[90-94] It is likely the higher enhancement in *S. vermifera* treated plants is associated with integration of aforesaid factors and higher availability of N and P which may have stimulated universal precursors of terpenoids i.e. isopentenyl pyrophosphate (IPP) and dimethylallyl pyrophosphate (DMAPP) synthesis better than other microorganisms and hence enhanced the yield of EO of fennel seeds. Microbial treatments of plants augmented with major chemical constituents. It is worth to mention that the elevated synthesis of major chemical constituents corresponds perfectly with the results of increased anethole in *F. vulgare* with *A. chroococcum*[78] and increased (+) pulegone and (-) menthone in *M. piperita*with *P. fluorescens.*[84] Likewise plants treated with *S. vermifera* synthesize higher concentration of anethole in *F. vulgare*[35] analogous effects have been observed with *T. vulgaris* with increased content of Thymol[36]. Similarly *S. vermifera* enhanced the production of podophyllotoxin and its 6-methoxy derivative in *Linum album*,[95-97] Additionally, precursor and elicitor of fungal origins are also in prevalence to enhance secondary metabolites in plant cell culture.[98-100] Enhancement in commercially important secondary metabolites *i.e.* artemisinin and withaferin-A were already achieved efficiently in cell culture technique facilitated with elicitor of fungal origins.[101,102] Although the underlying mechanism(s) behind triggering some particular component in aromatic plants is still to elucidate but it is speculated that the availability of integrated nutrients, early and late gene expressions and phytohormonal stimulus excavating some specific biosynthetic pathway and hence responsible for enhancement in some specific class of monoterpene in EO. Baldi et al. (2008) [96] suggested that *S. vermifera* could activate the defense pathway and hence activate the enhanced secondary metabolite production. Baldi et al. (2010)[13] also demonstrated that elicitors of fungal origin have potential to target the biosynthetic pathway and to enhance the secondary metabolite in cell culture. Further it was also demonstrated that *S. vermifera* enhanced the phenylalanine ammonia-lyase (PAL) activity which is a key role in the phenylpropanoid pathway to synthesize lignin.[96] Notably, the overall growth and biochemical responses were slightly elevated under pot trials in comparison to field trials. It is likely the soil of pot was taken from the field where regular practices were performed so the soil was with good texture, rich in nutrients along, augmented biological and microbial process and experiment was performed under control environmental conditions, which leads the plants to perform better whereas, the field trial was performed in barren land, deprived of biological and microbial processes, lesser nutrients especially NPK (Table 1) and harsh environmental conditions leads to the detrimental effects on the plant performance. Extrapolating all results achieved after microbial colonization study it is summarized that although Azotobacter and PSB could contribute towards the plant performance but *S. vermifera* can be excellent contributing agent toward overall positive changes in fennel plant including enhanced emergence, physiological fitness, enhanced yield of seed and hence enhanced yield of EO subsequently a better composition of secondary metabolites. A mechanistic insight of collective performances of *S. vermifera* is illustrated in Fig. 8.

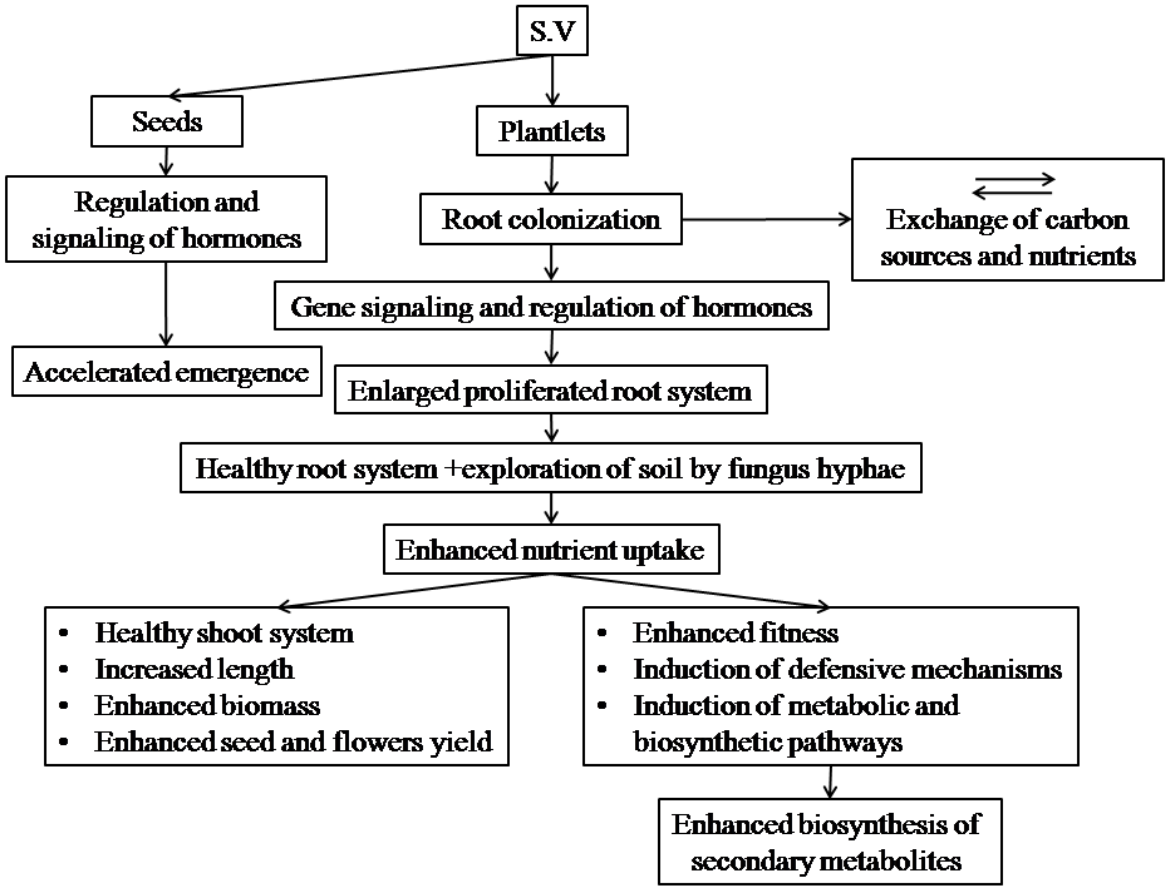

Fig 8 A mechanistic insight of collective performances of *S. vermifera*

6. CONCLUSION

In conclusion, *S. vermifera* extends all favourable effects on plant performance by contributing a wide range of beneficial nutrients therefore can improve the nutritional values of fennel as well as other nutraceuticals species. In addition to this, increased yield of seeds and EO could gain economical and commercial interest. Also our findings reveal that application of *S. vermifera* can be a cost effective and ecofriendly approach towards the fertilization. This could be a better alternative to conventional chemical -based fertilizers. Molecular level studies on unraveling the contribution of multifaceted beneficial fungi and colonization potential to a wide spectrum of plant species would be the futuristic exploration.

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9. AUTHORS CONTRIBUTION STATEMENT

The corresponding author, Dr. Ashish Baldi designed the work, supervised the experimentation, reviewed and edited the manuscript. Mr. Kamal Jeet performed the experiment, collected data, analyzed and prepared the manuscript.

10. CONFLICT OF INTEREST

Conflict of interest declared none.

11. REFERENCES

1. Gulfraz M, Mehmood S, Minhas N, Jabeen N, Kausar R, Jabeen K, Arshad G. Composition and antimicrobial properties of essential oil of *Foeniculum vulgare*. Afr J Biotechnol. 2008;7(24):4364-8. https://www.ajol.info/index.php/ajb/article/view/59591
2. Barros L, Carvalho AM, Ferreira ICFR. The nutritional composition of fennel (*Foeniculum vulgare*): Shoots, leaves, stems and inflorescences. LWT-Food Sci and Technol. 2010 Jun 1;43(5):814-8. doi: 10.1016/j.lwt.2010.01.010
3. Badgujar SB, Patel VV, Bandivdekar AH. *Foeniculum vulgare* Mill: A review of its botany, phytochemistry, pharmacology, contemporary application and toxicology. BioMed Research International. 2014. doi: 10.1155/2014/842674
4. Rahimi R, Ardekani MRS. Medicinal properties of *Foeniculum vulgare* Mill. in traditional Iranian medicine and modern phytotherapy. Chin J Integr Med. 2013 Jan 1;19(1):73-9. doi:10.1007/s11655-013-1327-0

5. Diaz-Maroto MC, Hidalgo IJD, Sanchez-Palomo E, Perez-Coello MS. Volatile components and key odorants of fennel (*Foeniculum vulgare* Mill.) and Thyme (*Thymus vulgaris* L.) Oil extracts obtained by simultaneous distillation–extraction and supercritical fluid extraction. J Agric Food Chem. 2005 Jun 29;53(13):5385-9. doi: 10.1021/jf050340+
6. Hashmi N, Khan MMA, Moinuddin, Idrees M, Khan ZH, Ali A, Varshney L. Depolymerized carrageenan ameliorates growth, physiological attributes, essential oil yield and active constituents of *Foeniculum vulgare* Mill. Carbohydrate polymers. 2012 Sep 1;90(1):407-12. doi: 10.1016/j.carbpol.2012.05.058
7. Khan MMA, Azam ZM, Samiullah S. Changes in the essential oil constituents of fennel (*Foeniculum vulgare*) as influenced by soil and foliar levels of N and P. Can J Plant Sci. 1999 Oct 1;79(4):587-91. doi: 10.4141/P98-090
8. Abdallah N, El-Gengaihy S, Sedrak E. The effect of fertilizer treatments on yield of seed and volatile oil of fennel (*Foeniculum vulgare* Mill.). Die Pharmazie. 1978 Sep;33(9): 607-8. https://europepmc.org/article/med/733880
9. Piccaglia R, Marotti M. Characterization of some italian types of wild fennel (*Foeniculum vulgare* Mill.) J Agric Food Chem. 2001 Jan 15;49(1):239-44. doi: 10.1021/jf000636+
10. Telci I, Toncer OG, Sahbaz N. Yield, Essential oil content and composition of *Coriandrum sativum* cultivars (var. *vulgare* Alef. and var. *microcarpum* DC.) grown in two different locations. J Essent Oil Res. 2006 Mar 1;18(2):189-93. doi: 10.1080/10412905.2006.9699063
11. Fuente EB, Gil A, Lenardis AE, Pereira ML, Suarez SA, Ghersa CM, Grass MY.Response of winter crops differing in grain yield and essential oil production to some agronomic practices and environmental gradient in the Rolling Pampa. Argentina. Agr Ecosyst Environ. 2003 Oct 1;99(1-3):159-69. doi: 10.1016/S0167-8809(03)00131-2
12. Jeet K, Malaviya A, Baldi A. Productivity enhancement of *coriandrum sativum* using plant biologicals. Int J Pharm Pharm Sci. 2020 Mar 31;12(5):60-72. doi:10.22159/ijpps.2020v12i5.37374.
13. Baldi A, Farkya S, Jain A, Gupta N, Mehra R, Datta V, Srivastava AK, Bisaria VS. Enhanced production of podophyllotoxins by co-culture of transformed *Linum album* cells with plant growth-promoting fungi. Pure Appl Chem. 2010 Jan 5; 82(1):227-41. doi: 10.1351/PAC-CON-09-02-09
14. Khaosaad T, Vierheilig H, Nell M, Zitterl-Eglseer K, Novak J. Arbuscular mycorrhiza alter the concentration of essential oils in oregano (*Origanum* sp., Lamiaceae). Mycorrhiza. 2006 Sep 1; 16(6):443-6. doi: 10.1007/s00572-006-0062-9
15. Smith SE, Read DJ. Mycorrhizal symbiosis.New York: Academic press; 2008.
16. Zubek S, Stojakowska A, Anielska T, Turnau K. Arbuscular mycorrhizal fungi alter thymol derivative contents of *Inula ensifolia* L. Mycorrhiza. 2010 Oct 1;20(7):497-504. doi: 10.1007/s00572-010-0306-6
17. Bona E, Cantamessa S, Massa N, Manassero P, Marsano F, Copetta A, Lingua G, D'Agostino G, Gamalero E, Berta G. (2016) Arbuscular mycorrhizal fungi and plant growth-promoting *Pseudomonads* improve yield, quality and nutritional value of tomato: A field study. Mycorrhiza. 2017 Jan 1;27(1):1-11. doi: 10.1007/s00572-016-0727-y
18. Saharan BS, Nehra V. Plant growth promoting rhizobacteria: A critical review. Life Sci Med Res. 2011 Jan 1;21(1):30. https://pdfs.semanticscholar.org/4bd8/7791af3a7e8cb1cd165e22bd6b67b47c7aca.pdf
19. Prasad R, Pham HG, Kumari R, Singh A, Yadav V, Sachdev M, Garg AP, Peskan T, Hehl S, Sherameti I, Oelmuller R, Varma A. Sebacinaceae: Culturable mycorrhiza-like endosymbiotic fungi and their interaction with non-transformed and transformed roots. In: Declerck S, Strullu DG, Fortin JA, editors. *In vitro* culture of mycorrhizas, soil biology. Berlin Heidelberg: Springer; 2005.p.291-312. doi:10.1007/3-540-27331-X_16 (2005)
20. Kumar V, Sahai V, Bisaria VS. High-density spore production of *Piriformospora indica*, a plant growth-promoting endophyte, by optimization of nutritional and cultural parameters. Bioresour Technol. 2011 Feb 1;102(3):3169-75. doi: 10.1016/j.biortech.2010.10.116
21. Kumar V, Sarma MVRK, Saharan K Rashmi Srivastava, Kumar L, Sahai Vikram, Bisaria VS, Sharma AK. Effect of formulated root endophytic fungus *Piriformospora indica* and plant growth promoting rhizobacteria fluorescent pseudomonads R62 and R81 on *Vigna mungo*. World J Microbiol Biotechnol. 2012 Feb 1;28(2):595-603. doi: 10.1007/s11274-011-0852-x
22. Jeet K, Baldi A. Development of inorganic carrier based bioformulation of *sebacina vermifera* and its evaluation on *Trigonella foenumgraecum.* Int J Pharma Bio Sci. 2020 April 1;11(2):69-82. DOI: 10.22376/ijpbs.2020.11.2.p69-82
23. ISTA. International rules for seed testing. Seed Sci Technol.1985;13:299-513.
24. Ellis RH, Roberts EH. The quantification of ageing and survival in orthodox seeds. Seed Sci Technol.1981 9(2):373-409. https://agris.fao.org/agris-search/search.do?recordID=XE8182678
25. Dezfuli PM, Sharif-Zadeh F, Janmohammadi M. Influence of priming techniques on seed germination behavior of maize inbred lines (*Zea mays* L.) ARPN J Agric Biol Sci. 2008 May; 3(3):22-25. https://www.semanticscholar.org/paper/INFLUENCE-OF-PRIMING-TECHNIQUES-ON-SEED-GERMINATION-Dezfuli-Sharif-zadeh/8d82a23e154c8df9b1199ed380f2b725cf9494a8
26. AOSA. Seed vigor testing handbook. contribution no. 32 to the handbook on Seed Testing. 1983.
27. Coolbear P, Francis A, Grierson D. The effect of low temperature pre-sowing treatment under the germination performance and membrane integrity of artificially aged tomato seeds. J Exp Bot. 1984 Nov 1; 35(11):1609-17. doi: 10.1093/jxb/35.11.1609
28. Farooq M, Basra SMA, Ahmad N, Hafeez K. Thermal hardening: A new seed vigor enhancement tool in rice. J Integr Plant Biol. 2005 Feb;47(2):187-93. doi: 10.1111/j.1744-7909.2005.00031.x
29. Farooq M, Basra SMA, Hafeez-ue-Rehman, Mehmood T. Germination and early seedling growth as affected

by pre-sowing ethanol seed treatments in fine rice. Int J Agr Biol 2006; 8:19-22. http://www.fspublishers.org/published_papers/40693_..pdf

30. Srinivasan K, Saxena S. Effect of dormancy breaking treatments on seed quality during storage of four acacia species. Indian J Forest 2007; 30:233-40. https://www.researchgate.net/publication/284385250_Effect_of_dormancy_breaking_treatments_on_seed_quality_during_storage_of_four_acacia_species
31. Abdul-Baki AA, Anderson JD. Vigour determination in soybean seed by multiple criteria. Crop Sci. 1973; 13(6):630-3. doi: 10.2135/cropsci1973.0011183X001300060013x
32. Phillip JM, Hayman DS. Improved procedures for clearing roots and staining parasitic and VAM fungi for rapid assessment of infection. T Brit Mycol Soc. 1970 Aug 1;55(1):158-161. doi: 10.1016/S0007-1536(70)80110-3
33. Giovannetti M, Mosse B. An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. New Phytol. 1980 Mar 1;84:489-500. doi: 10.1111/j.1469-8137.1980.tb04556.x
34. Tarraf W, Ruta C, Tagarelli A, De Cillis F, De Mastro G. Influence of arbuscular mycorrhizae on plant growth, essential oil production and phosphorus uptake of *Salvia officinalis* L. Ind Crops Prod. 2017 Aug 1;102:144-53. doi: 10.1016/j.indcrop.2017.03.010
35. Dolatabadi HK, Goltapeh EM, Jaimand K, Rohani N, Varma A. Effects of *Piriformospora indica* and *Sebacina vermifera* on growth and yield of essential oil in Fennel (*Foeniculum vulgare*) under greenhouse conditions. J Basic Microbiol 2011 Feb;51(1):33-9. doi: 10.1002/jobm.201000214
36. Barazani O, Benderoth M, Groten K, Kuhlemeier C, Baldwin IT. *Piriformospora indica* and *Sebacina vermifera* increase growth performance at the expense of herbivore resistance in *Nicotiana attenuata*. Oecologia. 2005 Dec 1;146(2):234-43. doi: 10.1007/s00442-005-0193-2
37. Dolatabadi HK, Goltapeh EM, Moieni A, Jaimand K, Sardrood BP, Varma A. Effect of *Piriformospora indica* and *Sebacina vermifera* on plant growth and essential oil yield in *Thymus vulgarisin vitro* and *in vivo* experiments. Symbiosis. 2011 Jan 1;53(1):29-35. doi: 10.1007/s13199-010-0104-0
38. Dolatabadi HK, Goltapeh EM. Effect of inoculation with *Piriformospora indica* and *Sebacina vermifera* on growth of selected brassicaceae plants under greenhouse conditions. J Hort Res. 2013 Dec 1;21(2):115-124. doi:10.2478/johr-2013-0030
39. Ghimire SR, Craven KD. Enhancement of switchgrass (*Panicum virgatum* L.) biomass production under drought conditions by the ectomycorrhizal fungus *Sebacina vermifera*. Appl Environ Microbiol. 2011Oct 1;77:7063-7. doi: 10.1128/AEM.05225-11
40. Ray P, Ishiga T, Decker SR, Turner GB, Craven KD. A novel delivery system for the root symbiotic fungus, *Sebacina vermifera*, and consequent biomass enhancement of low lignin COMT switchgrass lines. BioEnergy Research. 2015 Sep 1;8(3):922-33. doi: 10.1007/s12155-015-9636-8
41. Pirdashti H, Yaghoubian Y, Goltapeh E, Hosseini S. Effect of mycorrhiza-like endophyte (*Sebacina vermifera*) on growth, yield and nutrition of rice (*Oryza sativa* L.) under salt stress. J Agric Technol 2012; 8:1651-61. http://www.ijat-aatsea.com/pdf/v8_n5_12_July/12_IJAT_2012_8(5)_Hemmatollah_Pirdashti__T___Mycology.pdf
42. Yousefi S, Kartoolinejad D, Bahmani M, Naghdi R. Effect of *Azospirillum lipoferum* and *Azotobacter chroococcum* on germination and early growth of hopbush shrub (*Dodonaea viscosa* L.) under salinity stress. J Sustainable Forest. 2017 Feb 17; 36(2):107-20. doi: 10.1080/10549811.2016.1256220
43. Demissie S, Muleta D, Berecha G. Effect of phosphate solubilizing bacteria on seed germination and seedling growth of faba bean (*Vicia faba* L.). Int J Agric Res. 2013; 8(3):123-36. doi: 10.3923/ijar.2013.123.136
44. Gohokar RT, Saroodi D, Dethe RP, Ul Din Shah M. Impact of *Azotobacter chroococcum* and *Bacillus polymyxa* on Bamboo Seed Germination, Plant Growth and Soil Nitrogen and Phosphorus. SKUAST Journal of Research 2014; 16 (2):119-29. http://www.indianjournals.com/ijor.aspx?target=ijor:skuastjr&volume=16&issue=2&article=006
45. Nandakumar R, Babua S, Viswanathan R, Raguchandera T, Samiyappan R. Induction of systemic resistance in rice against sheath blight disease by Pseudomonas fluorescens. Soil Biol Biochem. 2001 Apr 1; 33(4-5):603-12. doi: 10.1016/S0038-0717(00)00202-9
46. Abo-Elyousr KAM, El-Hendawy HH. Integration of *Pseudomonas fluorescens* and acibenzolar-S-methyl to control bacterial spot disease of tomato. Crop Prot. 2008 Jul 1;27(7):1118-24. doi: 10.1016/j.cropro.2008.01.011
47. Fu Q, Liu C, Ding N, Lin Y, Guo B. Ameliorative effects of inoculation with the plant growth-promoting rhizobacterium *Pseudomonas* sp. DW1 on growth of eggplant (*Solanum melongena* L.) seedlings under salt stress. Agr Water Manage . 2010 Nov 15;97(12):1994-2000. doi: 10.1016/j.agwat.2010.02.003
48. Srivastava R, Khalid A, Singh US, Sharma AK. Evaluation of arbuscular mycorrhizal fungus, *fluorescent Pseudomonas* and *Trichoderma harzianum* formulation against *Fusarium oxysporum* f. sp. *lycopersici* for the management of tomato wilt. Biol Control 2010 Apr 1;53(1):24-31. doi: 10.1016/j.biocontrol.2009.11.012
49. Rubenchik IL, Starkey RL. *Azotobacter* and its use in agriculture. Soil Science. 1964 Oct 1; 98(4):280 https://journals.lww.com/soilsci/citation/1964/10000/azotobacter_and_its_use_in_agriculture.19.aspx
50. Mishustine N. The importance of non-symbiotic nitrogen-fixing microorganisms in agriculture. Plant Soil 1970 Jun 1;32:545-54. doi: 10.1007/BF01372895
51. Brown ME. Seed and root bacterization. Annu Rev Phytopathol 1974 Sep;12(1):181-197. https://www.annualreviews.org/doi/pdf/10.1146/annurev.py.12.090174.001145
52. Gholami A, Shahsavani S, Nezarat S. The effect of plant growth promoting rhizobacteria (PGPR) on germination, seedling growth and yield of Maize. International Journal of Biological, Biomolecular, Agricultural, Food and Biotechnological Engineering

2009 Jan 26;3(1):9-14. http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.192.9348&rep=rep1&type=pdf
53. Ardebili ZO, Ardebili NO, Hamdi SMM. Physiological effects of *Pseudomonas fluorescens* CHA0 on tomatoes (*Lycopersicon esculentum* Mill.) plants and its possible impact on *Fusarium oxysporum* f. sp. *lycopersici*. Aust J Crop Sci 2011; 5(12):1631-38 https://pdfs.semanticscholar.org/b4cf/51b3e609d4195b62ac9afe845de2b3dbd567.pdf
54. Ghimire SR, Charlton ND, Craven KD. The mycorrhizal fungus, *Sebacina vermifera*, enhances seed germination and biomass production in switchgrass (*Panicum virgatum* L). Bioenergy Res. 2009 Jun 1; 2(1):51-8. doi: 10.1007/s12155-009-9033-2
55. Varga S. Effects of arbuscular mycorrhizal fungi and maternal plant sex on seed germination and early plant establishment. Am J Bot. 2015 Mar; 102(3):358-66. doi: 10.3732/ajb.1400361
56. Lendzemo VW. The tripartite interaction between sorghum, *Striga hermonthica*, and arbuscular mycorrhizal fungi. Wageningen University; 2004. p. 112. Ph.D. Thesis
57. Rafter M, Yokoya K, Schofield EJ, Zettler LW, Sarasan V. Non-specific symbiotic germination of *Cynorkis purpurea* (Thouars) Kriezl., a habitat-specific terrestrial orchid from the Central Highlands of Madagascar. Mycorrhiza 2016 Aug 1; 26(6):541-52. doi: 10.1007/s00572-016-0691-6
58. Maighal M, Salem M, Kohler J, Rillig MC. Arbuscular mycorrhizal fungi negatively affect soil seed bank viability. Ecol Evol. 2016 Nov; 6(21):7683-9. doi: 10.1002/ece3.2491
59. Harper SHT, Lynch JM. Effects of fungi on barley seed germination. Microbiology. 1981 Jan 1;122(1):55-60. doi: 10.1099/00221287-122-1-55
60. Lendzemo VW, Kuyper TW, Matusova R, Bouwmeester HJ, Ast AV. Colonization by arbuscular mycorrhizal fungi of sorghum leads to reduced germination and subsequent attachment and emergence of *Striga hermonthica*. Plant Signal Behav. 2007 Jan 1; 2(1):58-62. doi:10.4161/psb.2.1.3884
61. Anantha naik T, Earanna N, Suresh CK. Influence of *Azotobacter chroococcum* strains on growth and biomass of *Adathoda vasica* Nees. Karnataka J Agric Sci. 2007;20(3):613-5. http://14.139.155.167/test5/index.php/kjas/article/viewFile/937/930
62. Chaudhary D, Narula N, Sindhu SS, Behl RK. Plant growth stimulation of wheat (*Triticum aestivum* L.) by inoculation of salinity tolerant *Azotobacter* strains. Physiol Mol Biol Plants. 2013 Oct 1;19(4):515-9. doi:10.1007/s12298-013-0178-2
63. Alemu F, Alemu T. *Pseudomonas fluorescens* isolates used as a plant growth promoter of Faba Bean (*Vicia faba*) *in vitro* as well as *in vivo* study in Ethiopia. Am J Life Sci. 2015 Mar 24;3(2):100-8. doi: 10.11648/j.ajls.20150302.17
64. Otieno N, Lally RD, Kiwanuka S, Lloyd A, Ryan D, Germaine KJ, Dowling DN. Plant growth promotion induced by phosphate solubilizing endophytic *Pseudomonas* isolates. Front Microbiol. 2015 Jul 22; 6:745. doi: 10.3389/fmicb.2015.00745
65. Dolatabadi HK, Goltapeh EM, Moieni A, Varma A. Evaluation of different densities of auxin and endophytic fungi (*Piriformospora indica* and *Sebacina vermifera*) on *Mentha piperita* and *Thymus vulgaris* growth. Afr J Biotechnol. 2012; 11(7):1644-50. doi: 10.5897/AJB10.1336
66. Waller F, Mukherjee K, Deshmukh SD, Achatz B, Sharma M, Schäfer P, Kogel KH. Systemic and local modulation of plant responses by *Piriformospora indica* and related Sebacinales species. J Plant Physiol. 2008 Jan 15;165(1):60-70. doi: 10.1016/j.jplph.2007.05.017
67. Vadassery J, Ritter C, Venus Y, Camehl I, Varma A, Shahollari B, Novák O, Strnad M, Ludwig-Müller J, Oelmüller R. The role of auxins and cytokinins in the mutualistic interaction between *Arabidopsis* and *Piriformospora indica*. Mol Plant Microbe Interact. 2008 Oct; 21(10):1371-83. doi: 10.1094/MPMI-21-10-1371
68. Xie H, Pastrnak JJ, Glick BR. Isolation and characterization of mutants of plant growth promoting rhizobacteria *Pseudomonas putida*, GR12-2 that over produced indole acetic acid. Curr Microbiol. 1996 Feb 1;32(2):67-71.doi: 10.1007/s002849900012
69. Lee YC, Johnson JM, Chien CT, Sun C, Cai DG, Lou BG, Oelmüller R, Yeh KW. Growth promotion of Chinese cabbage and *Arabidopsis* by Piriformospora indicais not stimulated by mycelium-synthesized auxin. Mol Plant Microbe Interact. 2011 Apr;24(4):421-31. doi: 10.1094/MPMI-05-10-0110
70. Karthikeyan A, Sakthivel KM. Efficacy of *Azotobacter chroococcum* in rooting and growth of *Eucalyptus camaldulensis* stem cuttings. Res J Microbiol. 2011 Jul 1; 6:618-24. doi:10.3923/jm.2011.618.624
71. Dong SQ, Tian ZH, Chen PJ, Kumar RS, Shen CH, Cai DG, Oelmüller R, Yeh KW. The maturation zone is an important target of *Piriformospora indica* in Chinese cabbage roots. J Exp Bot. 2013 Nov 1;64 (14):4529-40. doi:10.1093/jxb/ert265
72. Li Y, Hou L, Song B, Yang L, Li L. Effects of increased nitrogen and phosphorus deposition on offspring performance of two dominant species in a temperate steppe ecosystem. Sci Rep. 2017 Jan 19;7 (1):40951. doi: 10.1038/srep40951.
73. Lakshminarayana K. Influence of *Azotobacter* on nitrogen nutrition of plants and crop productivity. Proc Indian natn Sci Acad B. 1993;59:303-8. https://www.insa.nic.in/writereaddata/UpLoadedFiles/PINSA/Vol59B_1993_3and4_Art15.pdf
74. Kizilkaya R. Nitrogen fixation capacity of *Azotobacter* spp. strains isolated from soils in different ecosystems and the relationship between them and the microbiological properties of soils. J Environ Biol 2009 Jan 1;30(1):73-82. http://www.jeb.co.in/journal_issues/200901_jan09_spl/paper_12.pdf
75. Rodrıguez H, Fraga R. Phosphate solubilizing bacteria and their role in plant growth promotion. Biotechnol Adv. 1999 Oct 1;17(4-5):319-39. doi: 10.1016/S0734-9750(99)00014-2
76. Maheshwari SK, Gangrade SK, Trivedi KC. Comparative response of palmarosa to *Azotobacter* and nitrogen under rainfall and irrigated swords. Indian Perfume. 1991;35(2):308-311
77. Lewis AL, Domingues LO, Munoz OS. Effect of time and method of *Azotobacter chroococcum* application on

the cultivation of garlic (*Allium sativum* L.) cv. Vietnamita. Int Am Soc Tropical Hort.1995;39:27-32

78. Mahfouz SA, Sharaf-Eldin MA. Effect of mineral vs. biofertilizer on growth, yield, and essential oil content of fennel (*Foeniculum vulgare* Mill.). Int Agrophysics. 2007 Jan 1;21(4):361-6. http://www.international-agrophysics.org/Effect-of-mineral-vs-biofertilizer-on-growth-yield-and-essential-oil-content-of-fennel,106568,0,2.html
79. Ordookhani K, Sharafzadeh S, Zare M. Influence of PGPR on growth, essential oil and nutrients uptake of sweet basil. Adv Environ Biol. 2011 Jan 1; 5(4):672-7. http://scinet.dost.gov.ph/union/Downloads/672-677_225062.pdf
80. Shirkhodaie M, Darzi MT, Hadi MHS. Influence of vermicompost and biostimulant on the growth and biomass of coriander (*Coriandrum sativum* L.). International Journal of Advanced Biological and Biomedical Research. 2014; 2:706-14. http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.883.2960&rep=rep1&type=pdf
81. Darzi MT, Shirkhodaie M, Hadi MRHS. Effects of vermicompost and nitrogen fixing bacteria on seed yield, yield components of seed and essential oil content of Coriander (*Coriandrum sativum*). Journal of Medicinal Plants and By-products. 2015 Apr 1; 4(1):103-9. https://journals.areeo.ac.ir/article_108897_e149c62460c152c2d784baa3539e99d1.pdf
82. Hemavathi, Navi V, Sivakumar VS, Suresh CK, Earanna N. Effect of *Glomus fasciculatum* and plant growth promoting rhizobacteria on growth and yield of *Ocimum basilicum*. Karnataka J Agric Sci. 2006;19:17-20. http://14.139.155.167/test5/index.php/kjas/article/viewFile/587/577
83. Banchio E, Bogino PC, Zygadlo J, Giordano W. Plant growth promoting rhizobacteria improve growth and essential oil yield in *Origanum majorana* L. Biochem Syst Ecol. 2008 Oct 1;36(10):766-71. doi: 10.1016/j.bse.2008.08.006
84. Santoro MV, Zygadlo J, Giordano W, Banchio E. Volatile organic compounds from rhizobacteria increase biosynthesis of essential oils and growth parameters in peppermint (*Mentha piperita*). Plant Physiol Biochem. 2011Oct 1;49(10):1177-82. doi:10.1016/j.plaphy.2011.07.016
85. Copetta A, Lingua G, Berta G. Effects of three AM fungi on growth, distribution of glandular hairs, and essential oil production in *Ocimum basilicum* L. var. *Genovese.* Mycorrhiza. 2006 Oct 1;16(7): 485-94. doi: 10.1007/s00572-006-0065-6
86. Kapoor R, Chaudhary V, Bhatnagar AK (2007) Effects of arbuscular mycorrhiza and phosphorus application on artemisinin concentration in *Artemisia annua* L. Mycorrhiza 17:581-587. doi: 10.1007/s00572-007-0135-4
87. Mandal S, Upadhyay S, Wajid S, Ram M, Jain DC, Singh VP, Abdin MZ, Kapoor R. Arbuscular mycorrhiza increase artemisinin accumulation in *Artemisia annua* by higher expression of key biosynthesis genes via enhanced jasmonic acid levels. Mycorrhiza. 2015 Jul 1;25(5):345-57. doi: 10.1007/s00572-014-0614-3
88. Floß DS, Hause B, Lange PR, Kuester H, Strack D, Walter MH. Knock-down of the MEP pathway isogene 1-deoxy-d-xylulose 5-phosphate synthase 2 inhibits formation of arbuscular mycorrhiza-induced apocarotenoids, and abolishes normal expression of mycorrhiza-specific plant marker genes. Plant J. 2008 Oct; 56(1):86-100. doi: 10.1111/j.1365-313X.2008.03575.x
89. Mandal S, Upadhyay S, Singh VP, Kapoor R. Enhanced production of steviol glycosides in mycorrhizal plants: a concerted effect of arbuscular mycorrhizal symbiosis on transcription of biosynthetic genes. Plant Physiol Biochem. 2015 Apr 1;89:100-6. doi: 10.1016/j.plaphy.2015.02.010
90. Torelli A, Trotta A, Acerbi L, Arcidiacono G, Berta G, Branca C. IAA and ZR content in leek (*Allium porrum* L.), as influenced by P nutrition and arbuscular mycorrhizae, in relation to plant development. Plant Soil. 2000 Sep1; 226(1):29-35. doi: 10.1023/A:1026430019738
91. Kapoor R, Giri B, Mukerji KG. *Glomus macrocarpum*: A potential bioinoculant to improve essential oil quality and concentration in Dill (*Anethum graveolens* L.) and Carum (*Trachyspermum ammi* (Linn.) Sprague). World J Microbiol Biotechnol. 2002 Jul 1;18(5):459-63. doi: 10.1023/A:1015522100497
92. Strack D, Fester T, Hause B, Schliemann W, Walter MH. Review Paper: Arbuscular mycorrhiza: biological, chemical, and molecular aspects. J Chem Ecol. 2003 Sep 1;29(9):1955-79. doi: 10.1023/A:1025695032113
93. Krishna H, Singh SK, Sharma RR, Khawale RN, Grover M, Patel VB. Biochemical changes in micropropagated grape (*Vitis vinifera* L.) plantlets due to arbuscular-mycorrhizal fungi (AMF) inoculation during ex vitro acclimatization. Sci Hort. 2005 Nov1;106(4):554-67. doi: 10.1016/j.scienta.2005.05.009
94. Sailo GL, Bagyaraj DJ. Influence of different AM-fungi on the growth, nutrition and forskolin content of *Coleus forskohlii*. Mycol Res. 2005Jul;109(7):795-8. doi: 10.1017/S0953756205002832
95. Baldi A. Production of anticancer drug podophyllotoxin by plant cell cultivation of linum album (Doctoral dissertation).
96. Baldi A, Jain A, Gupta N, Srivastava AK, Bisaria VS. Co-culture of arbuscular mycorrhiza-like fungi (*Piriformospora indica* and *Sebacina vermifera*) with plant cells of *Linum album* for enhanced production of podophyllotoxins: A first report. Biotechnol Lett. 2008 Sep1;30(9):1671-7. doi: 10.1007/s10529-008-9736-z
97. Farkya S, Baldi A, Kumar V, Datta V, Mehra R, Gupta N, Jain A, Srivastava AK, Bisaria VS. Impact of symbiotic fungi on production of secondary metabolites by plant cell culture. AsPac J Mol Biol Biotechnol 2010;18:51-3. https://pdfs.semanticscholar.org/dfbe/fdc7a67f40f19bbea05d179f504379e7527e.pdf
98. Bisaria VS, Baldi A, Kumar V, Gupta N, Jain A, Farkya S, Srivastava AK. Interaction of phyto promotional fungi and plant cells on synthesis of plant-derived metabolites. J Biotechnol 2008 Oct;136:S11. doi: 10.1016/j.jbiotec.2008.07.1804
99. Baldi A, Jain A, Gupta N, Srivastava AK, Bisaria VS. Co-culture of *Linum album* cells and *Piriformospora indica* for improved production of phytopharmaceuticals. In: Varma A, Kharkwal AC,

editor. Symbiotic Fungi fungi, . Berlin Heidelberg:Springer; 2009.p.361-72. https://link.springer.com/chapter/10.1007/978-3-540-95894-9_22

100. Baldi A, Srivastava AK, Bisaria VS. Fungal elicitors for enhanced production of secondary metabolites in plant cell suspension cultures. In: Varma A, Kharkwal AC, editor. Symbiotic fungi. Berlin Heidelberg: Springer; 2009.p.373-80. https://link.springer.com/chapter/10.1007/978-3-540-95894-9_23

101. Baldi A, Dixit VK. Yield enhancement strategies for artemisinin production by suspension cultures of *Artemisia annua*. Bioresour Technol. 2008 Jul 1;99(11):4609-14. doi 10.1016/j.biortech.2007.06.061

102. Baldi A, Singh D, Dixit VK. Dual elicitation for improved production of withaferin A by cell suspension cultures of *Withania somnifera.* Appl Biochem Biotechnol. 2008 Dec 1; 151(2-3):556-64. doi: 10.1007/s12010-008-8231-2