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Physiological and Biochemical Attributes of an Endophyte *Stenotrophomonas maltophila,* AVSW 1 Isolated from Chilli on PGP of Tomato

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Abstract

This study aims to understand the role of *Stenotrophomonas maltophilia* AVSW 1, a chilli root endophytic bacteria, in promoting plant growth and fungal antagonism against *Fusarium oxysporum* in tomato. Ability of AVSW1 in terms of fungal antagonism, SEM analysis of root colonization, growth optimization and enhancement of the production of Indole-3-aceticacid, Ammonia and siderophore, and phosphate solubilisation followed by *in vitro* plant growth promotion of tomato using seed bacterization were evaluated. using GC-MS and HPLC analysis of volatile compounds and secondary metabolites of AVSW1was also studied. AVSW1 showed 26.3µg/ml of Ammonia production, 19.33 µg of IAA production, 60.67 psu of Siderophore and 91.67ppm of phosphate solubilisation under optimised growth conditions(35^{0}C, pH7,1% NaCl,1% Fructose, 1% Peptone and 60 h incubation). Growth parameters like root length, shoot height, no. of leaves and lateral roots, biomass, and protein and carbohydrate are much higher in AVSW 1 inoculated plants compared to untreated control. GC-MS analysis revealed that *2-Pentanone,4-Hydroxy-4-methyl, Cyclopropane,1-(1-Methylethyl)-2-Nonyl-Glycine, N-Acetyl-N(Trifluoroacetyl), MethylEster2-Acetoxy Isobutyryl Chloride, propanoic Acid, 2-Oxo-, Methyl Ester Pentanoic Acid* 4-Oxo,5-Hydroxy pentane hydroxyl amine Ethanol,2-(Octyloxy), 2-Cyclopenten-1-One, 2-Hydroxy-3,4-Dimethyl and 2, 2- Di methyl tetrahydro pyran-4-ol are pivotal compounds of S. maltophilia AVSW1 responsible for fungal antibiosis and root colonization to promote growth in tomato seedlings.

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Fungal Antagonism; *Fusarium oxysporum*; GC-Ms; *Lycopersicon esculentum*; Plant Growth Promotion; *Stenotrophomonas Maltophilia.*

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Introduction

Tomato (*Lycopersicon esculentum* Mill) is one of the rich source of vitamins, minerals, organic acids, essential amino acids and dietary fibres. Abiotic and Biotic factors(microbes) decreasing the production of tomato crop. Many Microbial diseases like *Fusarium wilt,* Early blight, Late blight, damping off, Leaf spots, Bacterial fruit canker, Anthracnose, bacterial speck affecting Tomato crop. Previous studies reports that ,soil nitrogen deficiency is decreasing quality and production of tomato crop, which can be reclaimed by potential PGP Bacteria as an alternative and eco friendly alternative to chemical fertilizers.[1-5] Nowadays, bio inoculants have gained more global attention due to their eco-friendly, cost-effective and easily replicable nature and used in agricultural ecosystems because of the presence of plethora of secondary metabolites responsible for plant growth promotion ,stress tolerance and disease resistance.[6-11] Endophytic bacteria enhances plant growth by the production of IAA, Ammonia, Siderophore, secondary metabolites, solubilizing Phosphates, increasing pathogenicity and maintain the integrity of host cell wall.[1,2,12,13] Due to its close association with the host , they developed a unique mechanism of synthesizing secondary metabolites for amicable association with the host in a symbiotic manner. Such specific secondary metabolites and volatile compounds express defence against fungi and bacteria, induce systemic resistance, immunity and enhance plant growth under oxidative stress conditions.[14-19]

Siderophores produced by plant growth promoting bacteria are also involved in iron transportation and as biocontrol agents by scavenging the available iron present in the surroundings of the pathogen siderophore production.[20] However, the metabolic profiling and physiological attributes of metabolites from endophytic bacteria have not been much focussed. The present research aimed to understand the metabolic profiling of an endophytic bacteria, *Stenotrophomonas maltophilia* isolate, extracted from green fruits of chilli and its physiological attributes in the growth promotion of tomato.

Isolation of *Stenotrophomonas maltophila* AVSW 1

AVSW 1 an endophytic bacteria isolated from the Green fruit of the chilli plant, was collected from Guntur, Andhra Pradesh, India. It was collected from the corresponding author as a subject for the current research. AVSW 1 was purified by the streak plate method and grown in nutrient broth. After 48 hrs of incubation, the endophytic bacterial strain was stored.[21-23]

Materials and Methodology

Screening of fungal antagonism and PGP activities

The antifungal activity of AVSW 1 was assessed using the double culture method.[24] Bacterial isolates were streaked on Sabouraud dextrose sugar (SDA) medium at 3 cm from the pathogenic fungi inoculated at the centre. Antifungal activity was measured at room temperature after 4-7 days of incubation. Inhibition levels were measured as I= 1-(a/b) X as described earlier.[13-17,25,26]

Indole-3-Acetic Acid Production

One ml culture filtrate of 72 hrs old bacterial culture was supplemented with 1 µg. ml^{-1} L- tryptophan and 2 ml Salkowski reagent and incubated at 28 ± 20° C for 30 min. Appearance of Pink colour indicates the presence of IAA.[27]

Phosphate Solubilization

Solubilization of Tricalcium phosphate was detected in Pikovskaya's agar.[21] Bacterial isolate was streaked on the surface of the Pikovskaya agar medium, and activity was estimated after 1 to 5 days of incubation at room temperature. Development of the clear zone around the bacterial colony was a positive response for phosphate solubilization.

Ammonia Production

Fresh bacterial culture was inoculated into 10 ml of peptone water and incubated at 36 ± 2° C for 48-72 h. A change of colour from brown to yellow with the addition of 0.5 ml of Nessler's reagent was a positive reaction for ammonia production.[28]

Siderophore Production

The bacterial strain was grown on a succinate medium and incubated for 24-30 h with constant shaking at 120 rpm at 28º C. For every 20 minutes of interval, 5 ml broth was taken and centrifuged at 10,000 rpm for 10 min at 4º C. The cell-free supernatant was mixed with 0.5 ml CAS solution and colour was measured at 630 nm.0.5 ml un-inoculated succinate medium and 0.5 ml CAS solution was used as control.[28]

Biochemical Physiological and Molecular Identification

Biochemical tests such as The IMViC test, Indole test, MRVP, Citrate utilization, Hydrogen sulphide production, and sugar fermentation were done as per the standard protocols.[29]

Physiological Characteristics

Amylase,Cellulase, Catalase, Oxidase, Urease, Gelatin hydrolysis, Nitrate reduction, Haemolysis and Lecithinase were analysed as per the protocols.[29-32]

Molecular Identification

It was done according to Bergey's manual of determinative bacteriology[33] for tentative identification of Genus followed by 16S rRNA partial gene sequencing analysis using universal primer 1492R (5´-TACGGYTACCTTGTTACGACTT-3') and 27F (5' AGAGTTTGATCMTGGCTCAG-3'). Phylogenetic tree analysis was done by using Neighbour joining method with 1000 bootstrap replicates. Isolate was deposited in GenBank, NCBI.[34]

Optimization Studies

Physical parameters ,temperature, pH, incubation period, and chemical parameters NaCl, carbon, and nitrogen sources were studied in 10ml of broth inoculated with100 µl of bacterial culture and incubated for 48 h .O.D of the culture broth was read at 600 nm.[35] Growth optimization of endophytic bacteria was analyzed at different temperatures ranging from 20-45°C at an interval of 5°C, different pH value range [3-9], saline concentration range 1-6 %, 1% carbon sources (Glucose, Fructose, Sucrose, Lactose, Maltose, Glycerol, Mannitol, Starch, and Cellulose) , 0,5%nitrogen sources (KNO3, NaNO3, NH4SO2, Urea, Peptone, Beef extract, Yeast extract, Casein, and Malt extract) and incubation periods (24 h, 36 h, 48 h, 60 h and 72 h). Bacterial Growth Curve was also determined for 24 h bacterial culture at an interval of 4 h.

PGP Activity of Endophyte by Pot Assay Studies

Tomato seeds were treated with 48 h bacterial culture for 30 min and shade-dried for 1 h. Inoculated seeds were seeded into coco peat,and the pots were kept under greenhouse conditions.% of seed germination was evaluated at an interval of 4,6 and 8 weeks. Physiological parameters, root length, shoot height, biomass, no of leaves, no of lateral roots, and chemical constituents Protein content by Lowry's and carbohydrate content by DNS method were estimated.

Metabolite Profile Fingerprint of Endophytic Bacteria

Secondary metabolite fingerprint of endophyte was analysed in ethyl acetate extract using Fourier transforms infrared spectroscopy (FTIR) and GC-MS[36,37,39]

Scanning Electron Microscopy

Root colonization ability was studied using SEM. One cm of root pieces were fixed and then processed with the PATOTO method. The prepared samples were mounted on aluminium stubs with Scotch TM double-sided tape, coated with gold in a sputtering Hummer II (Technics, Springfield, VA) and examined in a Cambridge S360 Scanning Electron Microscope.

Results and Discussion

Fungal Antagonism and PGP Traits

Pure culture of AVSW 1 was screened for fungal antagonism against *Fusarium oxysporum* by dual culture method as shown in Plate -1. AVSW 1 showed significant inhibition of Fusarium oxysporum compared to control and positive to PGP traits such as IAA, Ammonia, Phosphate solubilisation and siderophore production in qualitative screening.

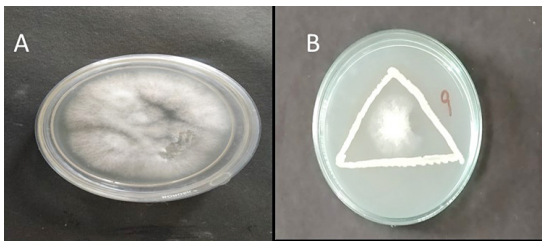


Plate 1: Screening of Fungal antagonism of AVSW 1 against *Fusarium oxysporum*
A. *F. oxysporum* on NAM as control and
B. Dual culture of *F. oxysporum* and AVSW 1

Effect of Physical and Chemical Parameters on Bacterial Growth

Physicochemical parameters were analyzed on a one-time factor (OFOT) basis to optimize the growth and maximise the production of PGP traits. Optimization studies revealed that AVSW 1 showed maximum growth at 37^0 C, 1% salinity and 60 h incubation period with minimal medium ameliorated with 1% Fructose and 1 %Peptone (Fig 1).

The growth curve of *S. maltophilia* AVSW 1 showed 4-8 h of Lag phase,12-44 h of Exponential phase, and 44-68 h of stationary phase. After 68 h, the decline phase was observed (Fig2).

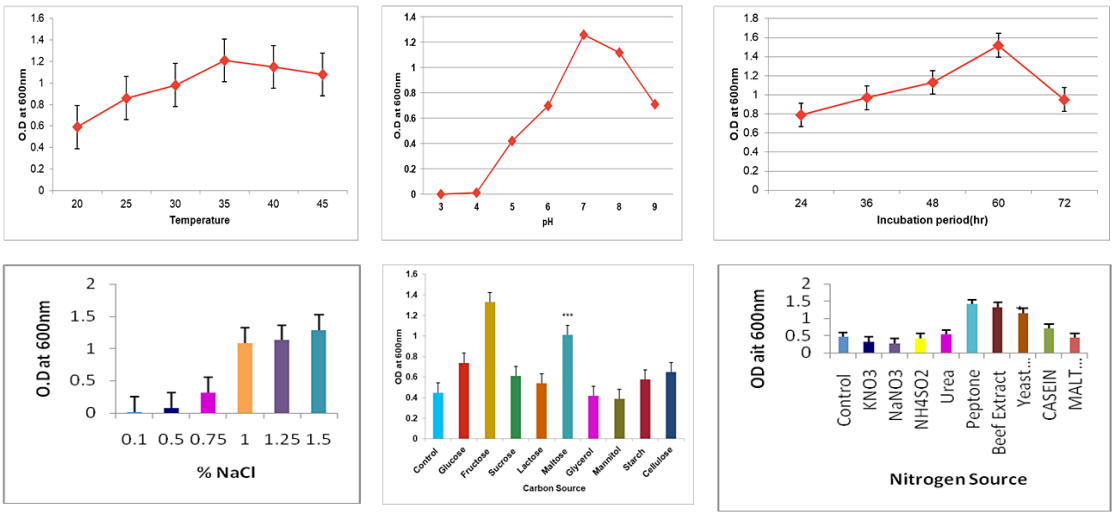


Fig. 1: Optimization of Physical and Chemical parameters on growth of AVSW 1

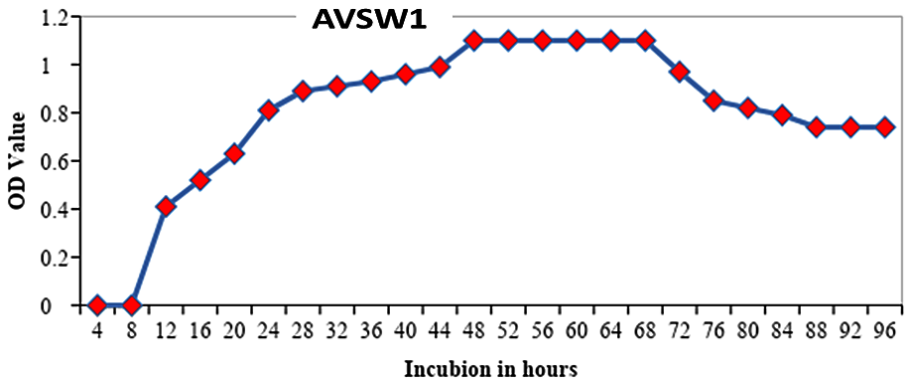


Fig 2: Growth analysis of AVSW 1 from 4 - 96 h of incubation in optimized medium

PGP traits of AVSW 1

Plant growth promotion (PGP) traits in *S. maltophilia* AVSW 1, such as IAA, Ammonia, PO4- solubilization and siderophore production, were evaluated after optimization.(Table1 and fig 2) In *S. maltophilia* AVSW 1, IAA production increased by 55.66 %, Ammonia production increased by 74.85 %, Phosphate ($po4^{-}$) solubilization increased by 69.42 % and Siderophore production increased by 49.62 % after Optimization. Whereas in earlier findings, *S. maltophilia JVB5*, root endophyte of sunflower showed optimization were IAA (30.0 µg/ml), Phosphate solubilisation (32.23 ppm) and Siderophore (79.90 psu) after optimization.[4] Previous findings reported that two *S.maltophila* IPR-Pv696 and 262XG2 root nodular endophyte strains of clover reported IAA (30.26 µg /ml and 31.15 µg/ml), Phosphate solubilization (100 and 75%) and phosphate liberated (278 mg/l and 208 mg/l).[11] 10.73 µg /ml phosphate solubilization, Indole acetic acid 3.16 µg/ml and gibberellic Acid 40 µg /ml were observed in *S. maltophilia*, SBP-9 rhizobacteria of Sorghum bicolour at pH 8, temperature 30º C, Nacl Concentration 6 % and broth incubated for 48 h.[6] 818 ppm Phosphate solubilisation, 1,62 IU /ml Acid phosphatase, 93µg/ml IAA production, Ammonia production (80 µg/ml) and ability to produce siderophore and HCN reported

earlier in *Stenotrophomonas maltophilia* AVP 27 rhizobacterium of chilli.[40] Similarly, present results also revealed that *S.maltophila* AVSW 1 is specific to the host in the expression of PGP traits and varies with endophytic and rhizospheric habitats. Further observed that the ability of phosphate solubilisation is less in endophytic strain compared to rhizosphere strain. It may be a marker characteristic feature to distinguish endophytic PGP strain from PGPR.

Table 1: Quantitative analysis of Plant growth promotion (PGP) traits in AVSW 1

Optimization	IAA (µg/ml)	Ammonia (µg/ml)	PO_4^- solubilization (ppm)	Siderophore (psu)
Before (control)	14.17	15.07	33.6	33.33
After	25.1	33.1	69.33	55.33

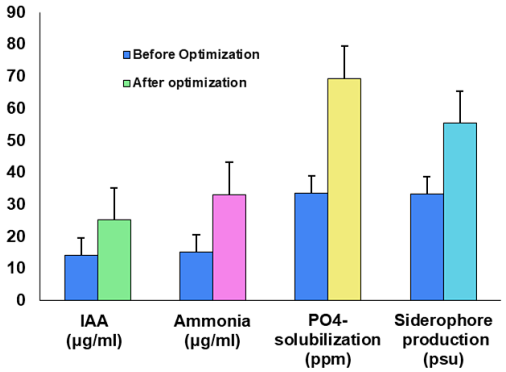


Fig 3: Impact of optimization on Plant growth promotion (PGP) traits of AVSW 1

Identification of AVSW 1

Based on colony morphology, morphological, and physiological characteristics (Table 2) followed by 16s rRNA partial gene sequencing analysis (Fig 4), AVSW 1 was identified as *Stenotrophomonas maltophilia* and deposited in NCBI GENBANK with the name *Stenotrophomonas maltophilia* AVSW 1 with the accession no OQ293900

Greenhouse Studies

The plant growth promotion potential of *Stenotrophomonas maltophilia* AVSW 1 was tested on tomato seedlings using the seed bacterization method followed by inoculation of culture at the collar region under greenhouse conditions. Root colonization efficacy of AVSW 1 inoculum was observed on 4, 6 and 8 week old seedlings treated with AVSW 1 under scanning electron microscope (Fig 5.1 & 5.2)

Table 2: Colony morphology and Morphological, Biochemical and physiological

Characteristics of AVSW 1	
Morphological Characteristics	
Colony Morphology	Small, circular and off white
Gram's reaction	Positive
Cell shape	Rod
Biochemical Characteristics	
Indole ,MR	Negative
VP/Citrate	positive
Sugar fermentations	
Glucose /Sucrose/Fructose /Maltose/Mannitol	A+/G-
Lactose / Arabinose / Inositol / Sorbitol/Dulcitol	A-/G-
Physiological chaacteristics	
Decarboxylation reactions	
Ornithine/ Arginine / Creatinine/ Lysine	Positive
Malonate	Negative
Enzymatic reactions	
Amylase/Urease /Super oxide dismutase/ Phenylalanine deaminase /Gelatin hydrolysis/ Oxidase	Negative
Cellulase peroxidases Poly phenyl oxidase Lecithinase Catalase Oxidase Nitrate reduction	positive
Haemolysis	Positive
Spore formation	Positive

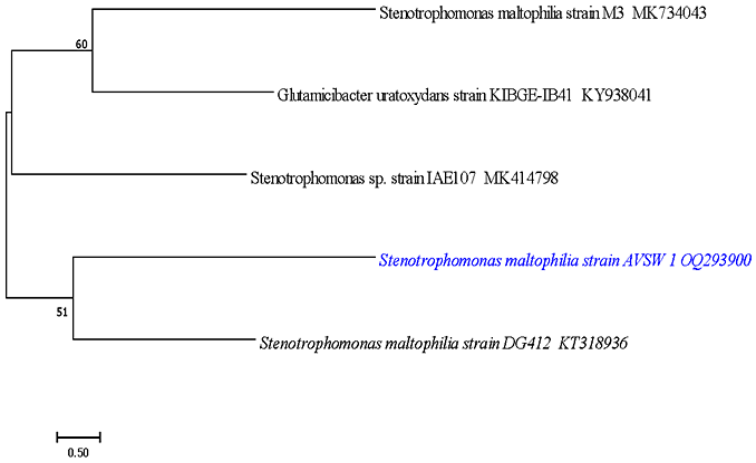


Fig: 4: Phylogenetic distance tree of Stenotrophomonas maltophilia AVSW 1 (OQ293900) constructed by the neighbour-joining method using Blast N of NCBI with 1000 bootstraps

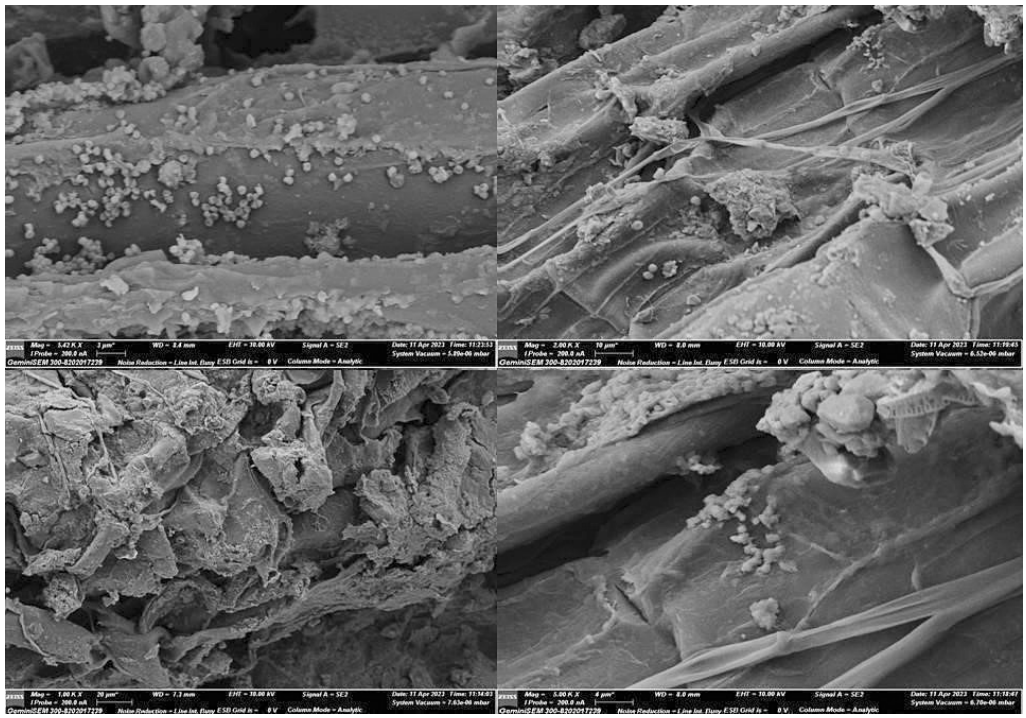


Fig 5.1: Scanning Electron Microgram of the root of tomato seedling treated with AVSW 1

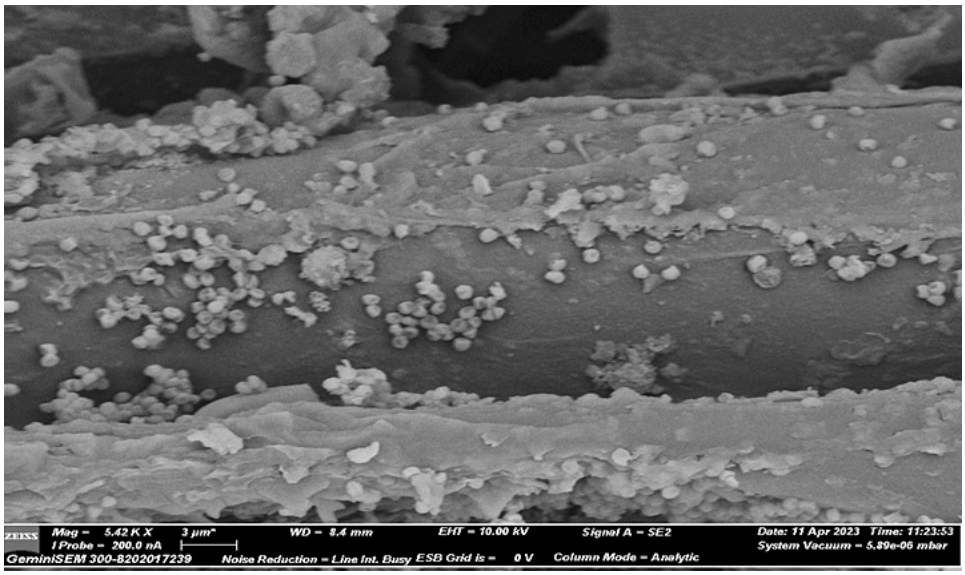


Fig 5.2: Scanning Electron Micrograph showing clear root Colonization in AVSW 1 treated tomato seedlings

Growth parameters such as root length, shoot height, root shoot ratio, No of leaves and fresh weight, and nutritional metabolites (carbohydrates and proteins) were studied at intervals of two weeks from 4th week onwards using un inoculated tomato seedlings as control.

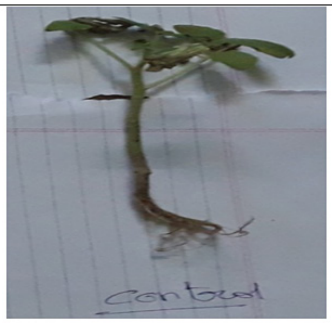
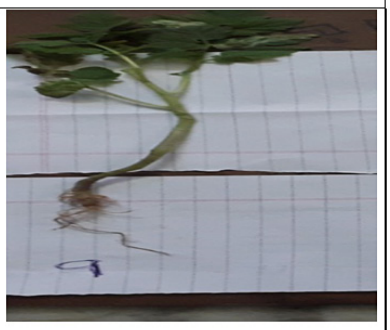
Week	Control Plant	AVSW 1 Treated plant
IV		
VIII		

Fig. 6: Greenhouse studies of tomato seedlings treated with AVSW 1

In previous findings, endophytic bacterial strains, namely CT12, CT 13 and CT 16 of *S. maltophilia* isolated from fruit, stem and leaf of tomatoes, respectively, showed nearly 35.9 % enhancement of shoot height and 19.6-28.3% root length of tomato seedlings and also observed enhancement of fresh weight of shoot and root by 39.5 -57 % and 38.2 -58.8 % respectively under greenhouse conditions.[41]

Similarly, *Stenotrophomonas maltophilia* BJ01 isolated from halo tolerant grass species showed a 17.68 % enhancement of shoot height and 57.14 % enhancement of fresh weight in peanut plants compared to control under high salt stress conditions (100 mM NaCl)[9] In previous findings, *Stenotrophomonas maltophilia* SBP-9 rhizospheric isolate of Sorghum bicolour showed significant enhancement of 20 % shoot height, 28.81% root length, 28 % fresh weight and 42 % dry weight of the Plant and 18.40 % shoot fresh weight. It also reported that chlorophyll content increased by 55% at 150 mM NaCl, 39 % at 200 mM NaCl and 25 % at 200 mM NaCl when compared with 0 % NaCl in wheat plants.[6]

S.maltophilia JVB5 isolated from sunflower root endosphere showed significant enhancement of root length (30.23 %), lateral roots (37.45 %) and fresh weight (31.65%). IPR-Pv696 and 262XG2, two *Bacillus toyonensis* strains of root nodules of clover, showed a significant increase in fresh weight to (40.53 fed^{-1} and 42.68 tons fed^{-1}), chlorophyll content by 4.51 % and carbohydrate content by 1.519 %, 20.18 % respectively.[4]

Similarly, in the present investigation, AVSW 1 treated tomato seedlings showed significant response in terms of the growth attributes and nutritive metabolites from the fourth week to the 8th (fig 5.1 to 5.3). A significant increase of Shoot length (53.47 % ,41.85 % and 40.55 %) root length(48.97 % , 48.76% and 52.97 %), number of leaves (92.29 %, 69.07 %, 84.33 %), lateral roots (52.81 %,43.77 %, 37.08 %), fresh weight of Plant (90.10 %, 99.60 %, 81.6 %) in 4th, 6th and 8th weeks respectively in AVSW 1 treated plants compared to control (Fig 6). AVSW 1 treated plants also showed significant enhancement of protein and carbohydrate (66.66 %, 43.83 %, 45.75 % & 33.33 %, 33.96 %, 29.50 %) from 4th week to 8th week. Our results emphasized that the isolate AVSW 1 can promote plant growth and nutritional health and its antifungal activity against *Fusarium oxysporum.*

Based on the study, there is a significant increase in root and shoot length, number of leaves and lateral roots when compared with control, protein and carbohydrate content and fresh weight is more in inoculated seeds and seeds treated with AVSW 1 inoculum showed better antifungal activity against *Fusarium oxysporum.*

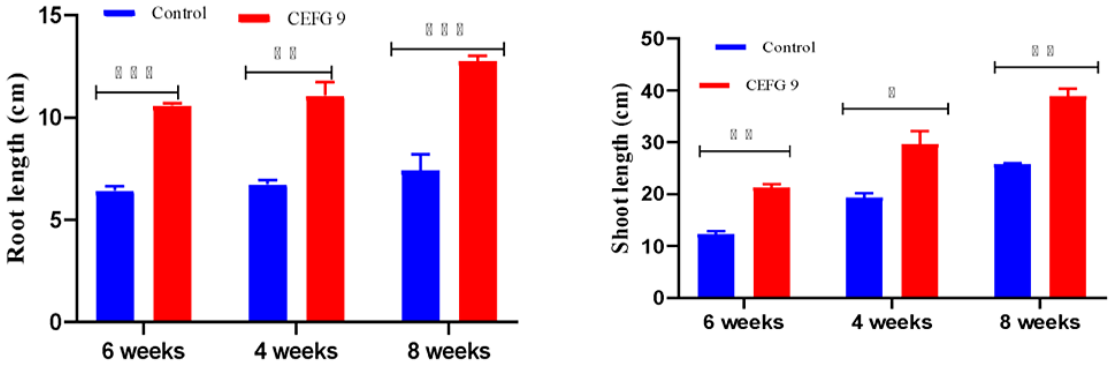


Fig. 7. 1: Root length and shoot height on 4th, 6th and 8th week in treated and control plant

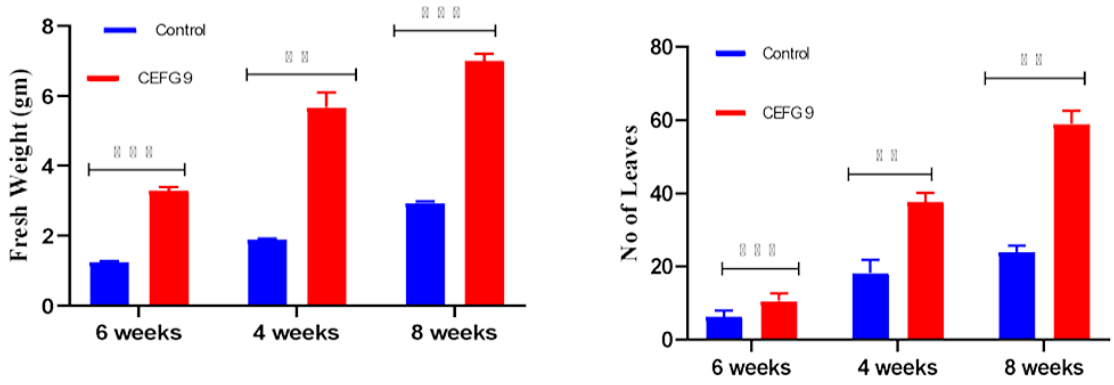


Fig. 7.2: Fresh weight and number of leaves on 4th, 6th and 8th week in treated and control plants

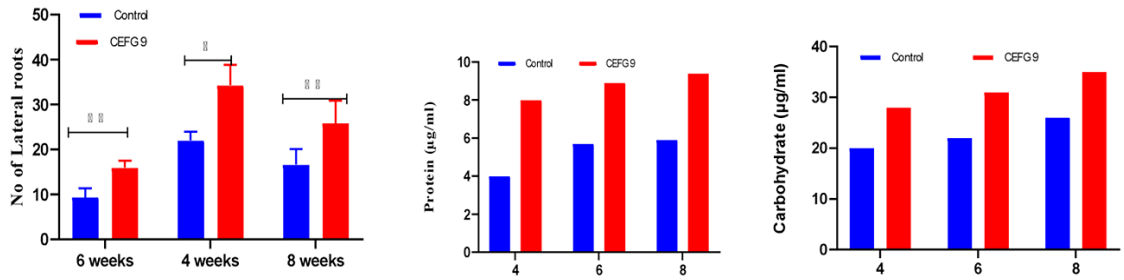


Fig. 7.3: Lateral roots, Protein and carbohydrate on 4th, 6th and 8th week in treated and control

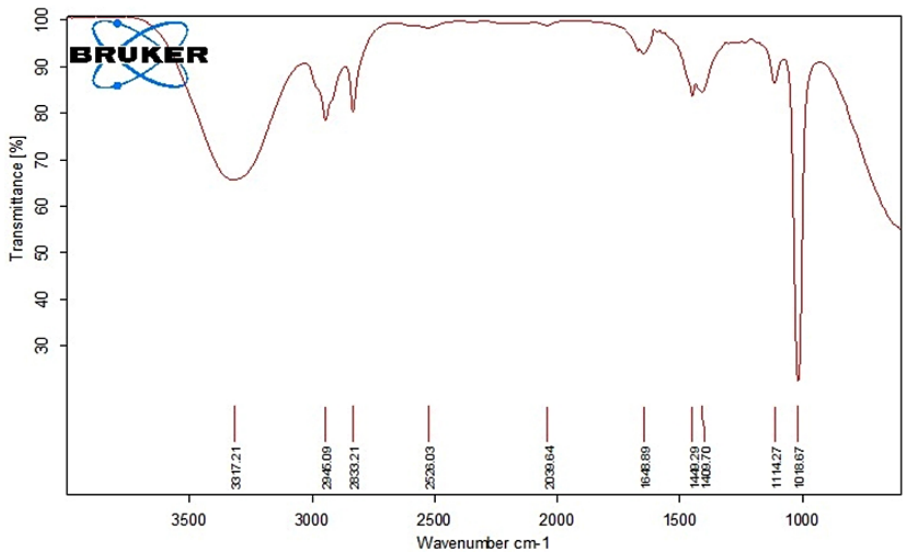
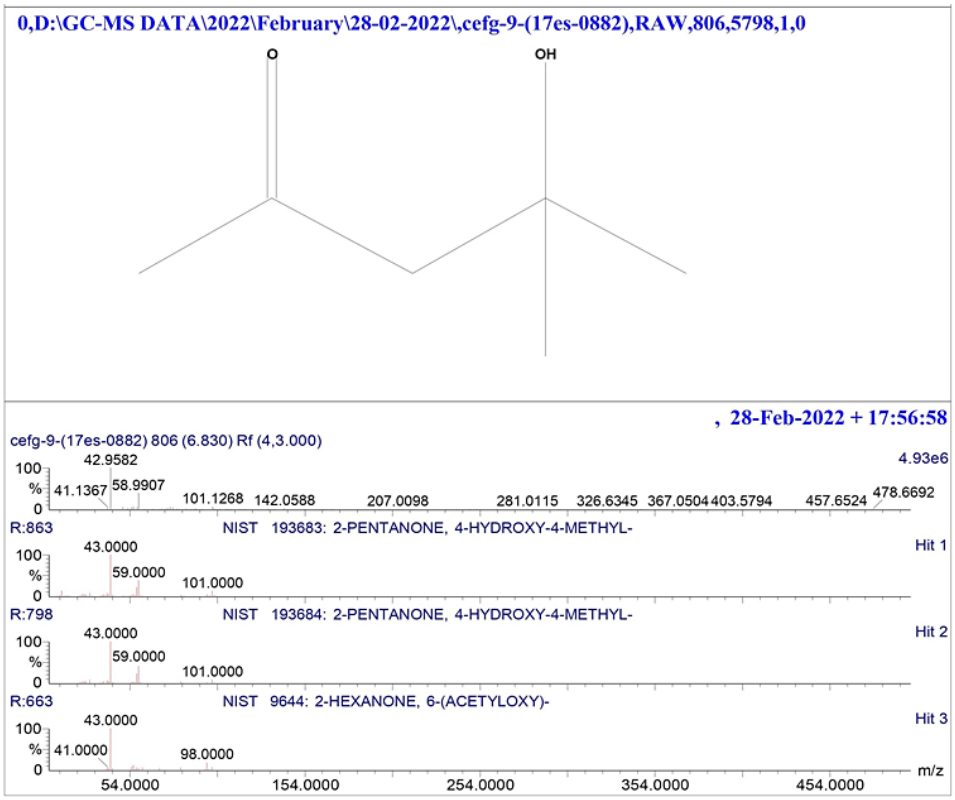


Fig. 8: FTIR spectral analysis and interpretation of bioactive metabolites of *Sphenotrophomonas maltophila* AVSW1

FTIR Spectral Data Analysis of Stenotrophomonas maltophila

Wavelength range:	Functional groups and their bonds
3314.25	C-H stretch, Alkyne (strong, sharp)
3303.73	OH stretch normal polymeric
2944.98	C-H stretch, Alkane (medium)
2832.35	N-H stretch, Alcohol (weak, broad)
2042.92	N=C=N stretch, Isothiocyanate (strong)
1659.67	C=C stretch, Alkene (medium)
1449.21	C-H bend, Alkane (medium)
1414.57	S=O stretch, Sulfate (strong)
1114.16	C-O stretch, secondary alcohol (strong)
1019.84	C-F stretch, Fluoro compound (strong)

In GC MS chromatogram major peak was observed at 6 min 8sec with the retention factor value of 43.00 with the molecular weight of 42.9582g/mol with the chemical formula C6H12O2. Compared to the NIST library, the primary compound was identified as 2-pentanone, 4-hydroxy, 4-methyl ester, which is confirmed by the FTIR results. Similarly, the compounds identified in the GC-MS analysis are confirmed to be the same as those in the FTIR analysis. As per the results of GC-MS, the profile of the AVSW 1 extract contains volatile organic compounds and secondary metabolites, which can efficiently control phytopathogens, increase plant growth, and induce systemic resistance.[37-39]



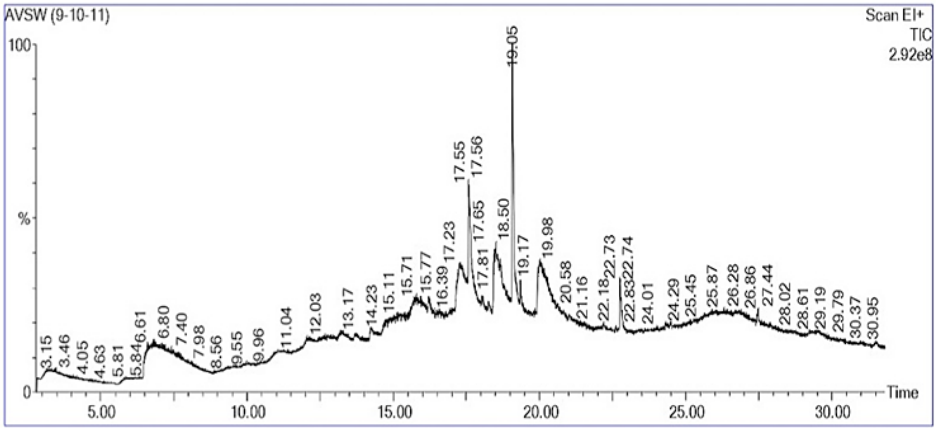


Fig: 9: GC-MS analysis of metabolites of Stenotrophomonas maltophila AVSW1 at RT 6.30
GC-MS Chromatogram of Stenotrophomonas maltophila AVSW 1

Table 3 : Metabolic Profile of Bacterial extract of AVSW 1 Stenotophomonas maltophilia

S. No	RT	Height	Name of the compound	Molecular Formula	Mol wt	Area % occupied	Norm %
1	6.830	30,461,028	2-Pentanone, 4-Hydroxy-4-Methyl-	C6H12O2	116	20.389	100
3	19.990	44,1313,68	Cyclopropane,1-(1-Methyethyl)-2-Nonyl-	C15H30	210	15.419	19.53
3	19.050	216,875,040	Glycine, N-Acetyl-N -(Trifluoroacetyl)-, Methyl Ester	C7H8F3NO4	227	13.917	18.88
4	17.559	106,691,120	2-Acetoxy Iso butyryl Chloride	C6H9O3Cl	164	13.507	6.54
5	17.274	40,798,592	Propanoic Acid, 2-Oxo-, Methyl Ester	C4H6O3	102	12.275	60.20
6	18.500	52,147,852	Pentanoic Acid, 4-Oxo-	C5H8O3	116	7.883	66.24
7	7.395	20,769,012	2-Pentanone, 4-Hydroxy-4-Methyl-	C6H12O2	116	3.983	38.66
8	7.660	15,587,187	2-Hexanone, 6-Acetyloxy)	C8H14O3	158	3.849	16.79
9	18.640	35,413,672	5-Hydroxy pentane hydroxyl amine, N,O, OTriacetyl-	C11H19O5n	245	3.423	68.26
10	22.727	42,736,244	Ethanol, 2-(Octyloxy)-	C10H22O2	174	2.664	6.66
11	19.330	24,171,646	2-yclopenten-1-One, 2-Hydroxy -3,4-Dimethyl-	C7H10O2	126	1.357	75.62
12	16.194	13,718,134	2-Pentanone, 4-Hydroxy -4-Methyl-	C6H12O2	116	1.333	13.06

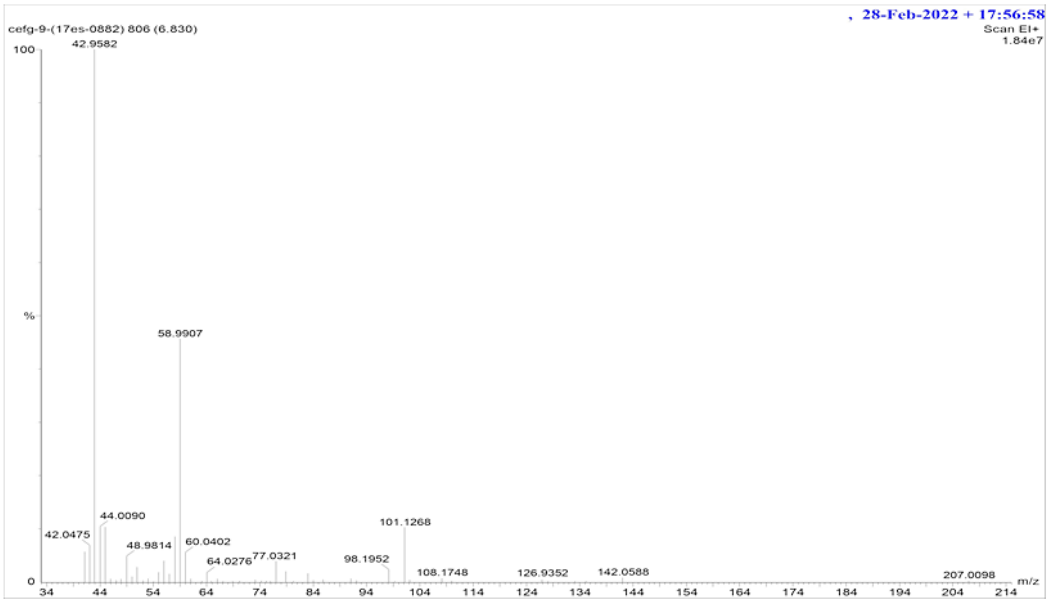


Fig: 10:GC-MS spectrum of Active metabolites of Sphenotrophomonas maltophila AVSW 1 at RT 6.830

Metabolic Profile of Bacterial Extract of AVSW 1 *Stenotrophomonas maltophilia* As per the NIST Database, bioactive metabolites such as 2-Pentanone, 4-Hydroxy-4-methyl, Cyclopropane, 1-(1-Methyl ethyl)-2-Nonyl-, Glycine, N-Acetyl-N-(Trifluoro acetyl)-, Methyl Ester, 2-Acetoxy Iso butyryl Chloride, Propanoic Acid, 2-Oxo-, Methyl Ester, Pentanoic Acid, 4-Oxo-,5-Hydroxypentanehydroxylamine, Ethanol, 2-(Octyloxy)-, 2-Cyclopenten-1-One, 2-Hydroxy-3,4-Dimethyl- , 2,2- Di methyl tetra hydro pyran-4-ol showed bioactivities like root colonization, antifungal, high salinity stress tolerance, production of secondary metabolites and agrochemicals, signalling of molecules, enhanced microbial tolerance, anti-oxidation, cryoprotection, immune response, regulating bacterial glycogen metabolism, gluconeogenesis, anti-inflammatory, plant defence mechanisms and promotes plant growth and metabolism.

In earlier studies, *Stenotrophomonas maltophilia* UN1512 of strawberries reported the presence of Benzothiazole, 1,3,5,7-Cyclooctatetraene-1-carboxaldehyde, Carbonic Acid, octadecyl phenylester, Benzaldehyde,2,5-bis [(trimethylsilyl) oxy]-, Estragole, and Benzaldehyde, and significantly inhibited the growth of fungal pathogens and promote growth of tomato plants *in vitro*.[43]

Volatile compounds like haloalkanetrichloromethane, 2,4-dimethyl heptane and 4-methyl octane, furans and dimethyl sulphide, α- and β-pinene, camphene, and Δ-3-carene were reported in *S.rhizophila* EP2.2 which are potential antifungal metabolites against *A.alternata* and *B.cinerea*[18 ,44-47,49,50]

Discussion

Currently, utilizing PGP bacteria in agriculture as Bio fertilizers and Bio inoculants is more winsome. Such bio inoculants protect plants from diseases and biotic and abiotic stress conditions by producing various regulatory chemicals, volatile organic compounds and secondary metabolites in the vicinity of the rhizosphere. Many Bacterial endophytes can colonize an ecological niche, making them potential biocontrol agents against diseases.

Phosphate solubilising endophytic bioinoculant is one of the alternative biotechnological solutions in sustainable agriculture to meet the phosphate demands of plants. IAA is one of the most crucial signal molecules in regulating plant growth. Salinity, pH and temperature are primordial abiotic factors which control plant growth, photosynthetic capacity, protein synthesis, energy, lipid metabolism, and total nitrogen content. PGP bacterial isolates which can have acclimatization to extreme high and low levels

of salinity, pH and temperature will be a potential bioresource for sustainable crop productivity. In present research the isolate, *Stenotrophomonas maltophilia* AVSW 1, showed multiple potential by having acclimatization to abiotic factors apart from direct (IAA, phosphate solubilisation, Ammonia) and indirect (HCN, volatile compounds, siderophore s) mechanisms. Secondary metabolites such as 2-Pentanone, 4-Hydroxy-4-methyl, Cyclopropane, 1-(1-Methyl ethyl)-2-Nonyl-, Glycine, N-Acetyl-N-(Trifluoro acetyl)-, Methyl Ester, 2-Acetoxy Iso butyryl Chloride, Propanoic Acid, 2-Oxo, Methyl Ester, Pentanoic Acid, 4-Oxo-, 5-Hydroxypentane Hydroxylamine, Ethanol, 2-(Octyloxy)-, 2-Cyclopenten-1-One, 2-Hydroxy-3,4-Dimethyl-, 2,2- Di methyl tetrahydro pyran-4-ol present in AVSW 1 are reported to be responsible for root colonization and suppression of soil borne fungal pathogens. Hence tomato plants inoculated with AVSW1 showed enhanced growth in greenhouse trials. Further advanced technological intervention can be useful to bring potential formulation with this isolate *Stenotrophomonas maltophila* AVSW1.

Conclusion

In conclusion, Tomato plants inoculated with AVSW 1,an endophytic plant growth promoting bacteria isolated from chill roots effectively suppressed fungal (*Fusarium oxysporum*) growth and exhibited significant growth-promoting traits and growth enhancement compared to untreated control. In this study, PGP isolate AVSW 1 showed maximum PGP traits in in-vitro conditions, perhaps can be used as a plant growth promoter in several vegetable or fruit crops. AVSW 1 improves plant growth directly through phosphate solubilization, production of secondary metabolites, volatile organic compounds, and growth hormones including nitrogen fixation. It reduces phytopathogens by sequestration of iron, secretion of siderophores, release of volatiles, etc. Further, the growth of plant pathogens may be directly inhibited by antibiosis. AVSW 1 may also induce systemic resistance in the host, where the plant defence mechanism gets activated against pathogen attack. The results indicate that Plant growth-promoting rhizobacteria AVSW 1 isolated from chilli roots can help the tomato plant withstand stress and support the Plant morphologically, physiologically, and biochemically. It can potentially promote tomato growth directly or indirectly, and further exploration of AVSW 1 as a bioinoculant can be tested at the field level to confirm its commercial significance. Genomic, proteomic and meta bolomics of holobiome (Plant and associated micro biome) and interdisciplinary research findings of AVSW 1 will be beneficial to understand its biocontrol potential, mode of action, regulatory mechanisms, and plant-microbe interaction in detail.

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Conflict of Interest

The authors declare that there is no conflict of interest

Data Availability Statement

The manuscript incorporates all datasets produced throughout the research study

Ethics Statement

Not applicable

Author's Contribution

Gadala Swapna carried out the laboratory work, conceptualization, wrote the manuscript text and methodology. Amrutha V. Audipudi was the supervisor of the research work

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