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INDUCTION OF SYSTEMIC RESISTANCE IN TOMATO BY ENDOPHYTIC MICROBIAL CONSORTIUM AGAINST BACTERIAL WILT PATHOGEN *RALSTONIA SOLANACEARUM*

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ABSTRACT
Induction of systemic resistance by endophytic microbial consortium was studied in both tomato root and stem, against challenge inoculation of *Ralstonia solanacearum* by assay of defense related compounds such as phenols, oxidative enzymes and PR proteins. Higher accumulation of phenols was noticed in stem samples and the activity of oxidative enzymes and PR proteins was higher in roots. The plants treated with microbial consortium showed higher activity of the defense related compounds with the maximum in plants inoculated with both consortium and pathogen. These results suggest that induction of defense enzymes involved in phenyl propanoid pathway and accumulation of phenols and PR proteins might have contributed to restriction of invasion of *R. solanacearum* in tomato plants.

KEY WORDS: Endophytes, microbial consortium, phenols, oxidative enzymes, PR proteins.

INTRODUCTION

Among the various diseases affecting tomato, bacterial wilt caused by *Ralstonia solanacearum* (Smith) Yabuuchi *et al.* is the major production limiting factor and it causes extensive losses in Asia and South Pacific regions. The warm humid tropical climate and soil conditions prevailing in Kerala are conducive for the occurrence of bacterial wilt and a yield loss up to cent percent has been reported in susceptible varieties. Bacterial wilt is caused by a genetically diverse soil borne pathogen with wide host range and is difficult to manage once it established in the field. Indiscriminate use of chemicals results in environmental pollution, development of resistant strains and detrimental effects on non target organisms including human beings. Among the biological management strategies, utilization of antagonistic endophytes is considered as one of the novel approaches for efficient disease management due to their intimate systemic association with the plants. Endophytes are microorganisms that inhabit inside plant tissues without causing any apparent harm to the hosts (Petrini, 1991) and they benefit the host by promoting plant growth and prevent pathogenic organisms from colonization. This novel method of biological control has entered the arena of disease management with attempts to make the plant, defend itself from the pathogens by induced systemic resistance. Recently, a greater thrust is being given for the development of microbial consortium, since it consists of microbes with different biochemical and physiological capabilities, which permit interactions among themselves and provide better management of diseases by way of synergistic effect and multiple mode of action. Recognizing the potentiality of endophytes and the consortial effect of the microorganisms, the present study was undertaken with the objective of enhancing defense mechanism by endophytic microbial consortium against bacterial wilt pathogen in tomato as a result of induced systemic resistance.

MATERIALS & METHODS

A pot culture experiment was conducted to study the changes in the defense mechanism in the root and stem of tomato plants against *R. solanacearum* due to application of endophytic microbial consortium against *R. solanacearum*. Healthy tomato plants were collected from 16 different locations representing north, central and south Kerala and endophytic microorganisms were isolated from both root and stem of tomato. After *in vitro* and *in planta* evaluation, these endophytes were tested for mutual compatibility and an endophytic microbial consortium consisted of five different microorganisms including *Trichoderma viride-1*, *T. viride-2*, *T. harzianum-1*, *Bacillus subtilis*, and *Streptomyces thermo diastaticus* was developed. Consortial inoculum was prepared by inoculating PDB with 48 h old bacterial culture (@ 1 loopful/ 100 ml) and five and seven day old fungal and actinomycete culture (@ 1 cm disc/100 ml) separately. The inoculation dates of different endophytes were adjusted accordingly to complete the incubation period of all endophytes on the same day. The cultures of the specific consortium were mixed and diluted with sterile water to prepare 30 % consortium suspension.

The treatments details are given below:
T_1: Microbial consortium alone
T_2: Microbial consortium +pathogen
T_3: Control (pathogen alone)
T_4: Medium alone
T_5: Absolute control
Consortial suspension of 30 % was applied to the soil at the time of planting and challenge inoculation of the pathogen was done 30 days after consortial treatment with

fresh ooze of the bacterial pathogen. Plants in each treatment were carefully uprooted at 0, 5, and 10 days after inoculation of the pathogen, and the roots were washed in running tap water. These samples were stored at -80^{0}C for biochemical analysis.

Extraction and estimation of phenols in both root and stem of tomato

Phenols were extracted and analysed as described by Mahadevan and Sridhar (1982). Root and stem tissues of one gram each, were cut into small pieces and immersed in boiling absolute alcohol@10 ml alcohol/ 1 g of tissue and kept in boiling water bath for 5-10 min. After cooling, the tissues were crushed with sterilized mortar and pestle using 80 % alcohol and were filtered. Residues were again extracted with 80% alcohol, boiling for 3 min and filtered. Both filtrates were mixed and final volume was made up to 4 ml with 80 % alcohol. Extracts were stored at 4^{0}C in screw capped vials covered with black paper. The whole extraction was done in dark to prevent light induced degradation of phenol.

Estimation of total phenols

Total phenol was estimated by Folin-Ciocalteau's method as described by Mahadevan and Ulaganathan (1991). To 1 ml of phenolic extract, 1 ml of Folin-Ciocalteau's reagent and 2 ml of 20 % Na_2CO_3 were added, shaken properly and heated on a boiling water bath for 1 min. Finally, the volume was adjusted to 25 ml with double distilled water. Absorbance was measured using spectrophotometer at 650 nm. Quantity of total phenol was estimated using catechol as standard.

Estimation of ortho-dihydroxy phenols (OD phenols)

OD phenol was estimated as described by Mahadevan and Ulaganathan (1991). To 1 ml of phenolic extract, 2 ml of 0.5 N HCl, 1 ml Arnow's reagent ($NaNO_3$-10g, Na_2MoO_4-10g, distilled water 100ml) and 2 ml of 1N NaOH were added. This was diluted to 10 ml with double distilled water. The tubes were shaken well and absorbance was recorded by spectrophotometer at 515 nm. Quantity of OD phenol was estimated using catechol as standard.

Assay of defense related oxidative enzymes

The important oxidative enzymes involved in the defense mechanism include phenylalanine ammonia-lyase (PAL), peroxidase (PO) and polyphenol oxidase (PPO). These enzymes were assayed at three intervals *viz.* 0, 5 and 10 days after inoculation of the pathogen.

Assay of peroxidase (PO)

One gram root and stem samples were homogenized separately in 2 ml 0.1 M phosphate buffer, pH 7.0, at 4^{0}C. The homogenate was centrifuged at 16000 g at 4^{0}C for 15 min and the supernatant was used as the enzyme source. The reaction mixture consisted of 1.5 ml of 0.05 M pyrogallol, 0.5 ml of enzyme extract and 0.5 ml of 1 % H_2O_2. The changes in OD were recorded at 30 sec intervals for 3 min at 420 nm. The enzyme activity was expressed as changes in the OD min^{-1} g^{-1} protein (Hammerschmidt *et al*., 1982).

Assay of polyphenol oxidase (PPO)

Polyphenol oxidase activity was determined as described by Mayer *et al.* (1965). Freeze dried root and stem samples of one gram each were homogenized in 2 ml 0.1 M sodium phosphate buffer (pH 6.5) and centrifuged at 16000 g for 15 min at 4^{0}C. The supernatant served as the enzyme source. The assay mixture comprised 0.2 ml of enzyme extract, 1.5 ml of 0.1 M sodium phosphate buffer (pH 6.5) and 0.2 ml of 0.01 M catechol. The rate of increase in absorbance was recorded in 30 sec interval up to 3 min at 420 nm. The enzyme activity was expressed as changes in absorbance min^{-1} g^{-1} fresh weight of tissue.

Assay of phenylalanine ammonia-lyase (PAL)

One gram of root and stem samples were homogenized separately in 3 ml of ice-cold 0.1 M sodium borate buffer, pH 7.0, containing 1.4 mM 2-mercapto ethanol and 0.1 g insoluble polyvinyl pyrrolidone. The extract was filtered through cheese cloth and the filtrate was centrifuged at 16000 g at 4^{0}C for 15 min. The supernatant was used as the enzyme source. Sample containing 0.4 ml of enzyme extract was incubated with 0.5 ml of 0.1 M borate buffer, pH 8.8, and 0.5 ml of 12 mM L-phenylalanine in the same buffer for 30 min at 30^{0}C. OD value was recorded at 290 nm. Activity of phenylalanine ammonia-lyase was determined as trans-cinnamic acid as described by Dickerson *et al.* (1984). Enzyme activity was expressed as µmol trans-cinnamic acid min^{-1} g^{-1} protein.

Assay of pathogenesis related (PR) proteins

The major PR proteins involved in the defense mechanism are β-1,3-glucanase and chitinase. The changes in the activity of these enzymes were assayed in different intervals after the inoculation of the pathogen.

Assay of β-1, 3-glucanase

B-1, 3-glucanase activity was assayed by the laminarin dinitrosalicylic acid method (Pan *et al.*, 1991). One gram of tomato root and stem tissue were extracted separately in 5 ml of 0.05 M sodium acetate buffer (pH 5.0). The homogenates were centrifuged at 16000 g for 10 min at 4^{0}C and the supernatant was used as enzyme source. The enzyme extract (62.5 µl) was mixed with equal volume of 4 % laminarin solution and incubated at 40^{0}C for 10 min. 375 µl of dinitrosalicylic acid was added to stop the reaction and heated for 5 min in boiling water bath. This coloured solution was diluted with 4.5 ml of distilled water, vortexed and its absorbance at 500 nm was determined. The enzyme activity was expressed as mg glucose released min^{-1} g^{-1} of sample.

Assay of chitinase activity

Chitinase enzyme activity was estimated according to the protocol described by Jeuniaux (1966) with slight modification.

Root and stem samples of one gram each were homogenized separately in 2 ml of 0.1 M sodium citrate buffer of pH 5.0. The homogenates were centrifuged at 16000 g for 15 min at 4^{0}C and the supernatant was used for the enzyme assay.

Preparation of colloidal chitin

Two gram of pure chitin was dissolved in 64 ml of prechilled conc. H_2SO_4 with constant stirring and the temperature maintained at 4^{0}C. It was allowed to stand at 4^{0}C for 1 h with occasional stirring. This viscous solution was transferred to 90 ml prechilled distilled water with continuous stirring and kept overnight at 4^{0}C. The precipitate formed was centrifuged and resuspended in distilled water several times, to remove excess acid and then dialysed against tap water until pH reaches 5. Chitin content of the suspension was determined by drying the sample *in vacuo* and adjusted with distilled water to a final

concentration of 10 mg ml^{-1} and stored at 4^0C for further use.

Preparation of P-dimethyl aminobenzaldehyde reagent (DAMB)

DMAB reagent was prepared by the procedure described by Reissig *et al.* (1955). Stock solution of DMAB was prepared by mixing 8 g of DMAB in 70 ml of glacial acetic acid along with 10 ml of concentrated HCl. One volume of stock solution was mixed with 9 ml of glacial acetic acid immediately before use and the reagent was prepared fresh.

Colorimetric method of chitinase activity

The reaction mixture consisted of 10 µl of 0.1 M sodium acetate buffer (pH 4.0), 0.4 ml enzyme solution and 0.1 ml colloidal chitin. After incubation for 2 h at 37^0C, the reaction was stopped by centrifugation at 8000 g for 3 min. An aliquot of 0.3 ml supernatant was added into a glass reagent tube containing 30 µl of 1 M potassium phosphate buffer of pH 7.1. After the addition of 2 ml of DMAB, the mixture was incubated for 20 min at 37^0C. Immediately thereafter, the absorbance was measured at 585 nm. N-acetyl glucosamine (G1cNAc) was used as a standard and the enzyme activity was expressed as moles G1cNAc equivalents $min^{-1}g^{-1}$ fresh weight.

RESULTS & DISCUSSION

It is observed that, the total and OD phenols were more in stem than the root, whereas, the activities of oxidative enzymes and PR proteins were higher in root as compared to stem. The lower level of phenolics in the roots might be due to the increased rate of oxidation of phenolics to more toxic compounds like quinones by the oxidative enzymes like polyphenol oxidase and peroxidase (Mahadevan, 1970). It is also noted that, in all cases, the plants treated with microbial consortium showed higher activity of the defense related compounds before the inoculation of the pathogen. Among the different treatments, plants treated with microbial consortium (T_1 and T_2) showed higher levels of phenols, oxidative enzymes and PR proteins especially in T_2, where plants treated with consortium and pathogen. In all treatments, an initial increasing trend was observed and later decreasing/increasing pattern noticed depending upon different treatments.

Estimation of phenols

Total phenol content

Total phenol content was more in stem as compared to root and varied from 2.69 to 3.21 and 2.86 to 3.64 mg g^{-1} in root and stem respectively before inoculation of the pathogen. After challenge inoculation, all treatments showed an increasing trend upto 5 DAI in both root and stem samples with maximum in T_2 (5.13 mg g^{-1}) followed by T_1 (3.35 mg g^{-1}). At 10 DAI, an increasing trend was noticed in consortium treated plants (T_1) whereas decreasing trend was observed in plants treated with consortium and pathogen (T_2) and pathogen alone (T_3). However, consortium treated plants (T_1 and T_2) showed high phenol contents compared to other treatments in both root and stem during all the intervals of observations (Table 1). Higher accumulation of phenolics by prior application of *P. fluorescens* challenged with the pathogen has been reported in various crops (Meena *et al.*, 2000, Vivekananthan *et al.*, 2004). Malinowski *et al.* (1998) reported 20 % more total phenolic concentration in endophyte infected plants than control plants. Rajendran *et al.* (2006) also reported enhanced mechanical strength of host cell walls and inhibition of invading *Xanthomanas axonopodis* pv. *malvacearum* in cotton by endophyte treatment.

TABLE 1. Effect of different treatments on total phenol content in tomato

Treatments	* Total phenol content (mg g^{-1} sample)					
	Root			Stem		
	0 DAI	5 DAI	10 DAI	0 DAI	5 DAI	10 DAI
T_1	3.1	3.35	3.42	3.47	3.61	3.95
T_2	3.21	5.13	4.02	3.64	5.81	5.37
T_3	2.74	3.27	2.58	2.95	3.84	2.85
T_4	2.71	2.78	2.88	2.90	3.02	3.64
T_5	2.69	2.72	2.78	2.86	3.13	3.23

* Mean of three replications
DAI – Days after inoculation
T_1 – Endophytic consortium alone
T_4 –Medium alone
T_2 - Endophytic consortium+pathogen
T_5 – Absolute control
T_3 – Pathogen alone

Estimation of ortho dihydroxy (OD) phenols

It is noticed that, OD phenol contents before inoculation varied from 0.29 to 0.46 and 0.37 to 0.55 mg g^{-1} in root and stem respectively. At 5 DAI, all treatments showed increasing trend with highest accumulation in consortium and pathogen treated one (T_2) followed by consortium alone (T_1) in both root and stem samples, whereas treatments T_1, T_2 and T_3 showed decreasing trend at 10 DAI. Plants treated with pathogen alone (T_3) showed less content as compared to control treatments (T_4 and T_5) (Table 2). This is in accordance with Tomiyama (1963) who reported that, mono and dihydric phenols increased in host tissues invaded by parasites as a part of resistance mechanism. The high toxicity of OD phenols and its role in resistance was also observed by Mahadevan (1966). He reported that, orthodihydroxy phenolic compounds such as caffeic acid, and chlorogenic acid, and orthoquinones and tannins were shown to strongly inhibit the activities of extracellular enzymes produced by microorganisms.

TABLE 2. Effect of different treatments on OD phenol content in tomato

Treatments	* OD phenol content (mg g^{-1} sample)					
	Root			Stem		
	0 DAI	5 DAI	10 DAI	0 DAI	5 DAI	10 DAI
T_1	0.46	0.49	0.48	0.52	0.50	0.57
T_2	0.45	0.56	0.46	0.55	0.71	0.61
T_3	0.32	0.38	0.28	0.37	0.42	0.28
T_4	0.29	0.32	0.34	0.39	0.38	0.36
T_5	0.31	0.34	0.36	0.43	0.35	0.36

* Mean of three replications
DAI – Days after inoculation
T_1 – Endophytic consortium alone
T_4 –Medium alone
T_2 - Endophytic consortium+pathogen
T_5 – Absolute control
T_3 – Pathogen alone

Assay of defence related enzymes

Peroxidase (PO) activity

The activity of PO as expressed by change in absorbance ranged from 1.06 to 1.42 $min^{-1}g^{-1}$ and 1.03 to 1.29 $min^{-1}g^{-1}$ in root and stem respectively before inoculation. The enzyme activity increased in all the treatments upto five days after inoculation with the maximum in consortium and pathogen treated root and stem (T_2) recording 2.41 and 2.23 $min^{-1}g^{-1}$ respectively followed by T_1 (consortium alone). However, at 10 DAI, a decreasing trend was observed in pathogen inoculated treatments, T_2 (consortium + pathogen) and T_3 (pathogen alone) in both root and stem samples which ranged from 1.22 to 1.96 and 1.18 to 2.02 $min^{-1}g^{-1}$ respectively with higher activity in consortium treated plants (T_1 and T_2), compared to other treatments. Any noticeable difference was observed in treatments with medium alone (T_4) and untreated control (T_5) (Table 3). Kurian (2011) reported increased PO activity in cocoa upto five days after inoculation and later decreasing trend in all treatments with endophytes. Uppala (2007) also noticed enhanced activity of peroxidase in amaranth by endophyte application. Various studies reported that, PGPR induced peroxidase in response to pathogen attack (Vijayaraghavan, 2007; Anita and Samiyappan, 2012).

TABLE 3. Effect of different treatments on peroxidase activity in tomato

Treatments	*Δ_{436} ($min^{-1}g^{-1}$ fresh tissue)					
	Root			Stem		
	0 DAI	5 DAI	10 DAI	0 DAI	5 DAI	10 DAI
T_1	1.42	1.78	1.62	1.29	1.98	1.76
T_2	1.38	2.41	1.96	1.26	2.23	2.02
T_3	1.18	1.38	1.22	1.12	1.44	1.34
T_4	1.06	1.21	1.34	1.03	1.12	1.18
T_5	1.10	1.32	1.41	1.09	1.24	1.45

Polyphenol oxidase (PPO)

Before inoculation of the pathogen, PPO activity in root and stem ranged from 0.37 to 0.5 and 0.24 to 0.32 $min^{-1}g^{-1}$ respectively. On inoculation of the pathogen, enhanced activity of PPO was observed in plants treated with consortium (T_2) at 5 DAI, with maximum of 0.77 and 0.49 $min^{-1}g^{-1}$ in root and stem respectively. Though the enzyme activity was found to be declined at 10 DAI in T_1, T_2 and T_3 treatments, the consortium treated plants (T_1 and T_2) showed better activity as compared to others with maximum of 0.69 $min^{-1}g^{-1}$ (root) and 0.45 $min^{-1}g^{-1}$ (stem) in T_2 (Table 4). There are earlier reports of enhanced levels of PPO by endophyte treatment in cotton against *X. malvacearum* (Rajendran *et al.*, 2006); in black pepper against *P. capsici* (Barka *et al.*, 2002) with reduction in disease. Enhanced activity of PO and PPO were observed in tomato roots treated with *P. fluorescens* and *Fol* (Ramamoorthy *et al.*, 2001). Chen *et al.* (2000) also reported similar enhanced activity of PPO in cucumber roots treated with various rhizobacteria and the pathogen, *P. aphanidermatum*.

TABLE 4. Effect of different treatments on polyphenol oxidase activity in tomato

Treatments	*Δ_{420} ($min^{-1}g^{-1}$ fresh tissue)					
	Root			Stem		
	0 DAI	5 DAI	10 DAI	0 DAI	5 DAI	10 DAI
T_1	0.47	0.65	0.63	0.29	0.38	0.36
T_2	0.50	0.77	0.69	0.32	0.49	0.45
T_3	0.45	0.47	0.36	0.27	0.32	0.30
T_4	0.42	0.39	0.41	0.24	0.22	0.26
T_5	0.37	0.40	0.42	0.24	0.27	0.29

Phenyl alanine ammonialyase (PAL)

Initially, the activity of PAL varied from 2.01 to 2.94 and 1.02 to 1.98 n mol transcinnamic acid in root and stem respectively. Same trend as in PO and PPO was observed at 5 DAI with maximum in T_2 showing 4.29 and 3.32 nmol trans cinnammic acid in root and stem respectively. Though the activity was found to be reduced at 10 DAI in all treatments except control (T_5), a drastic decline was noticed in T_2 from 4.29 to 3.06 in root and 3.32 to 2.09 n mol trans cinnammic acid in stem. In this case also, higher activity was observed in T_1 and T_2 of consortium treatments (Table 5). Chen *et al.* (2000) noticed high levels of PAL in cucumber roots treated with *P. corrugata* and later decreased after inoculation of *P. aphanidermatum*. Uppala (2007) observed increased PAL activity in amaranth with treatments of endophytes. Induction of phenols is also linked with induced PAL activity, which catalyses the first step in synthesis of phenols. Increased PAL activity and phenol accumulation in the present study may, thus be correlated with enhanced defense response by the microbial consortium.

TABLE 5. Effect of different treatments on phenylalanine ammonia lyase activity in tomato

Treatments	*Nmol trans cinnammic acid g^{-1} fresh tissue					
	Root			Stem		
	0 DAI	5 DAI	10 DAI	0 DAI	5 DAI	10 DAI
T_1	2.75	2.84	2.83	1.78	1.97	1.86
T_2	2.94	4.29	3.06	1.98	3.32	2.09
T_3	2.21	2.42	2.31	1.27	1.45	1.34
T_4	2.09	2.18	2.13	1.12	1.21	1.18
T_5	2.01	2.04	2.08	1.02	1.01	1.09

* Mean of three replications
DAI – Days after inoculation
T_1 – Endophytic consortium alone
T_4 –Medium alone
T_2 - Endophytic consortium+pathogen
T_5 – Absolute control
T_3 – Pathogen alone
DAI – Days after inoculation

TABLE 6. Effect of different treatments on glucanase activity of tomato

Treatments	*Glucose released (mg $min^{-1}g^{-1}$ fresh tissue)					
	Root			Stem		
	0 DAI	5 DAI	10 DAI	0 DAI	5 DAI	10 DAI
T_1	0.62	0.66	0.69	0.20	0.33	0.41
T_2	0.60	0.83	0.71	0.22	0.64	0.47
T_3	0.51	0.7	0.55	0.17	0.22	0.28
T_4	0.50	0.54	0.51	0.19	0.21	0.22
T_5	0.48	0.51	0.53	0.16	0.18	0.21

* Mean of three replications
DAI – Days after inoculation
T_1 – Endophytic consortium alone
T_4 –Medium alone
T_2 - Endophytic consortium+pathogen
T_5 – Absolute control
T_3 –Pathogen alone

Assay of pathogenesis related proteins (PR proteins)

B-1, 3-glucanase activity

The activity of β-1,3-glucanase was found higher in root samples as compared to stem. Before the challenge inoculation, the activity ranged from 0.48 to 0.62 and 0.16 to 0.22 glucose units released in root and stem respectively. The treatments with consortium and pathogen showed maximum activity at 5 DAI followed by T_3. The activity was found increased at 10 DAI in consortium alone treated plants (T_1) in both root and stem whereas the activity was found to be reduced in plants inoculated with pathogen (T_2 and T_3). However, the activity was higher in consortium treatments (T_1 and T_2) in both root and stem at 10 DAI (Table 6). Kurian (2011) also observed enhanced activity of β-1,3-glucanase induced by endophytes in cocoa on challenge inoculation with *P. palmivora*. Increased β-1,3-glucanase activity on application of endophyte, *B. subtilis* has also been confirmed by Wilhelm *et al.* (1998) in chestnut against chestnut blight and Jayaraj *et al*. (2004) in rice against sheath blight.

Chitinase activity

The root samples showed higher chitinase activity as compared to stem. On fifth day of inoculation, enhanced activity was observed in all treatments in root varied from 1.3 to 2.2 n mol G_1cNAc $min^{-1}g^{-1}$ with maximum in consortium + pathogen treatment (T_2) followed by consortium alone (T_1). Among the stem samples also, treatment T_2 showed highest activity and the activity reduced in all treatments at 10 DAI, however higher activity was noticed in consortium treatments (T_1 and T_2) (Table 7). Similar results of increase in PR proteins after application with biocontrol agents have been reported by several workers in different crops (Kuc, 1995; De Meyer *et al.*, 1998, Meena *et al.*, 2000).

Summing up the above results, it is clearly evident that, consortium treated plants either before or after inoculation exhibited higher phenol contents, activity of oxidative enzymes and PR proteins, which might have collectively contributed to induced systemic resistance in tomato plants which is more pronounced after infection. In conclusion, the present study reveals the potential of endophytic microbial consortium in enhancing the plant's own defense mechanism against the bacterial wilt pathogen.

TABLE 7. Effect of different treatments on chitinase activity of tomato

Treatments	*Nmol G1cNAc ($min^{-1}g^{-1}$ fresh tissue)					
	Root			Stem		
	0 DAI	5 DAI	10 DAI	0 DAI	5 DAI	10 DAI
T_1	1.7	1.8	1.6	0.82	0.79	0.83
T_2	1.9	2.2	1.8	0.84	1.21	0.92
T_3	1.4	1.5	1.3	0.62	0.87	0.58
T_4	1.2	1.5	1.4	0.60	0.57	0.56
T_5	1.2	1.3	1.2	0.61	0.55	0.51

* Mean of three replications
DAI – Days after inoculation
T_1 – Endophytic consortium alone
T_4 –Medium alone
T_2 - Endophytic consortium+pathogen
T_5 – Absolute control
T_3 –Pathogen alone

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