



Simultaneous growth promoting and managing bacterial wilt of tomato through *Bacillus amyloliquefaciens*

DHANANJAY KUMAR YADAV¹, DINESH SINGH² and NARENDRA KUMAR³

ICAR-Indian Agricultural Research Institute, New Delhi 110012, India

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ABSTRACT

Bacterial wilt disease, caused by *Ralstonia solanacearum* (Smith) Yabuuchi has become a severe problem in tomato crops and no satisfactory control methods are currently available to reduce damage caused by the disease. For biological control of the disease, plant growth promoting rhizobacteria (PGPR) isolates particularly from *Bacillus* spp. may be considered as a promising strategy for managing the disease. Thirty nine isolates of *Bacillus amyloliquefaciens*, isolated from rhizospheric soil of solanaceous crops were characterized antagonistic and plant growth promoting activities *in vitro* conditions. The isolate UKTBA-8 of *B. amyloliquefaciens* was found best among all other isolates, that inhibited maximum growth of *R. solanacearum* under *in vitro* conditions and formed inhibition zone of 3.96 cm² followed by HPTBA-1 (3.3 cm²) and MPBA-6 (2.56 cm²). UKTBA-8 solubilized the highest phosphorus (81.59 µg/ml) produced maximum siderophores to form orange yellow halo (1.96 cm diameter) and indole acetic acid ranging from 41.25–97.41 µg/ml under *in vitro* conditions. Biocontrol efficacy and plant growth ability of best six bacterial antagonists, viz. HPTBA-1, MPBA-6, JHBA-6, ORTBA-1, KCBA-4 and UKTBA-8 were evaluated to control bacterial wilt disease of tomato cv. Pusa Ruby at National Phytotron Facility, IARI, New Delhi. *B. amyloliquefaciens* UKTBA-8 treated plants showed least wilt incidence (12.23%) with maximum biocontrol efficacy (83.36%) after 30 days of inoculation.

Key words: *Bacillus amyloliquefaciens*, PGPR, 16S rRNA, *Ralstonia solanacearum*, Wilt

Tomato (*Solanum lycopersicum* L.) is a major vegetable crop and having excellent source of vitamin C, biotin, molybdenum and vitamin K. However, the productivity of the crop is low due to many biotic and abiotic stresses during cultivation. Among various biotic stresses, bacterial wilt incited by *Ralstonia solanacearum* (Smith) Yabuuchi is a serious disease in India particularly in coastal and hilly areas and North- eastern states and causes very heavy losses, ranging from 2–90% in different climatic conditions, seasons, cultivars and strains of pathogen (Singh *et al.* 2010, 2018). There are methods such cultural, chemical, resistant varieties and biological control applied to manage the wilt disease but all have a limited success. Furthermore, use of chemicals has its adverse effects on the environment and the non-target organisms. In biological control, use of bacterial antagonists has emerged as one of the important methods for the management of the disease (Singh *et al.* 2013, 2016). Potential bacterial antagonists, such as *Pseudomonas fluorescens*, *Bacillus* spp. and *Actinomyces* are used to

control wilt disease on tomato. Among them, the *Bacillus* species such as *Bacillus amyloliquefaciens*, *B. pumilus* and *B. subtilis* isolated from rhizosphere of tomato and other crops are effectively used for management of bacterial wilt disease in tomato (Nguyen and Ranamukhaarachchi 2010, Singh *et al.* 2016, Cao *et al.* 2018). *B. amyloliquefaciens* has advantages over other genera of bacterial antagonists by having resistant to desiccation, better survivability at higher temperature due to endospore forming nature and also plant growth promoting ability (Zhang *et al.* 2012). These antagonists exert a beneficial effect on the plant they colonize; on the other hand, they interact with the plant roots and with the other microbes present in the rhizosphere. The characterization of new bacterial antagonists is done by morphological, biochemical, physiological and advanced methods such as DNA based techniques particularly 16S rRNA sequence analysis to distinguish *Bacillus* species (Wattiau *et al.* 2001). The present study was undertaken to isolate *Bacillus* spp. from the rhizosphere of solanaceous crops growing in the bacterial wilt infested field, characterize them and find out potential biocontrol agents and plant growth promoting ability to control bacterial wilt disease of tomato.

MATERIALS AND METHODS

Collection of rhizospheric soil: Rhizospheric soil

¹Ph D student (dhananjaymkp@gmail.com), ²Principal Scientist (dinesh_iari@rediffmail.com), ³Assistant Professor (narendra.microbiology@rediffmail.com), Amity Institute of Biotechnology, Amity University, Manesar, Gurgaon, Haryana.

Table 1 List of *Bacillus amyloliquefaciens* strains isolated from rhizosphere of tomato, potato, brinjal and chilli samples collected from different agro-climatic regions of India.

Strains of <i>B. amyloliquefaciens</i>	Host	Place of collection	Agro-climatic regions	Year of collection	Source of isolation
MPBA-1, MPBA-2, MPBA-3, MPBA-4, MPBA-5, MPBA-6, MPBA-7, MPBA-8, MPBA-9	Potato	Shillong, Meghalaya	Temperate climate Temp. (3–23°C). Altitude- 1966 meters	2015	Rhizosphere
ORTBA-1, ORTBA-2, ORTBA-4 ORTBA-3, ORTBA-5, ORTBA-6	Tomato	Khurda, Odisha	Tropical savannah Temp. (15–40°C)	2015	Rhizosphere
KCBA-1, KCBA-2, KCBA-4	Chili	Bhubneshwar, Odisha, Dharwad, Karnataka	Tropical Wet and Dry climate (15–40°C).	2015	Rhizosphere
HPTBA-1, HPTBA-2	Tomato	Shimla, Himachal Pradesh	Subtropical. Temp. (1–28°C). Alt-2397 m.	2015	Rhizosphere
JHBA-1, JHBA-2, JHBA-3, JHBA-4	Brinjal	BAU, Ranchi, Jharkhand	Agro climatic Zone-IV (Central and North-Eastern Plateau Zone)	2015	Rhizosphere
UKTBA-1, UKTBA-2, UKTBA-3, UKTBA-4, UKTBA-5, UKTBA-6, UKTBA-7, UKTBA-8, UKTBA-9, UKTBA-10, UKTBA-11, UKTBA-12, UKTBA-13, UKTBA-14, UKTBA-15	Tomato	Mukteshwar, Nainital, Chorgaliya Haldwani, Chaffi, Nainital	Subtropical high land	2015	Rhizosphere
DSBA-11*, DSBA-12*	Tomato	IARI, New Delhi	Influenced humid subtropical and semi-arid	2011	Rhizosphere

*Reference strains of *Bacillus amyloliquefaciens*

samples were collected from tomato, potato, brinjal and chilli from different locations of India such as Chorgaliya and Chaffi (Uttarakhand), East Khasi hills (Meghalaya), Bhubneshwar (Odisha), CPRI, Shimla (Himachal Pradesh), Birsa Agriculture University (BAU) Ranchi and Dharwad (Karnataka) under temperate, subtropical, tropical savanna as well as tropical wet and dry agro-climatic regions during 2013–2015. The samples were brought to the laboratory for isolation and characterization of *Bacillus* spp.

Isolation of *Bacillus* spp.: Ten gram rhizospheric soil was taken from each sample and put it in a 250 ml of flask containing 100 ml of sterilized distilled water and mixed it thoroughly. The mixed samples, then heated for 15 min at 60°C. 100 µl of diluted aliquot of soil suspension was plated on to Luria Bertani (LB) agar medium and incubated at 28 ± 1°C for 48 h (Tan *et al.* 2013). After 24–48 hr of incubation the colonies were appeared in the petri plates. Colonies of bacteria like typical irregular, dry, white were picked up and purified on Luria agar medium to get pure colony of *Bacillus* spp. The purified colonies were preserved at 4°C.

Characterization of *Bacillus* spp.: Thirty nine isolates of *Bacillus* spp. was characterized by using standard procedure (Schaad *et al.* 2001). Prototype strains DSBA-11 and DSBA-12 used in taxonomic comparison were obtained from Division of Plant Pathology, IARI, New Delhi, India. CTAB method was used for extraction of bacterial DNA

(Murray and Thompson 1980). Molecular characterization was done based on 16S rRNA sequence analysis using a set of universal primers UNI_OL5 and outer reverse UNI_OR (Sauer *et al.* 2005). Sequencing of PCR product was done as outsource from by Scigenome (<http://www.scigenome.com>) and these nucleotide sequences were aligned with the partial sequences of 16S rRNA gene of other *Bacillus* spp. taken from NCBI Gen Bank database (Fig 1). Neighbor-joining method of MEGA 5.0 software was used to prepare phylogenetic tree (Tamura *et al.* 2011).

Antagonistic activity against *R. solanacearum*: Dual culture method was used to evaluate antagonistic ability of 39 isolates of *B. amyloliquefaciens* along with two reference strains i.e. DSBA-11 and DSBA-12 to inhibit growth of *R. solanacearum* in vitro (Singh *et al.* 2016). The area of inhibition zone formed by bioagent was calculated as:

$$\text{Area of inhibition (circle)} = \pi r^2.$$

Estimation of plant growth promoting attributes in vitro: The isolates *B. amyloliquefaciens* showing antagonistic ability was estimated their plant growth promoting attributes, viz. phosphate solubilization, and siderophores production and indole acetic acid (IAA) production *in vitro* conditions. Quantitative estimation of phosphate solubilization was measured as described by Mehta and Nautiyal (2001). Indole acetic acid was assayed colorimetric using ferric

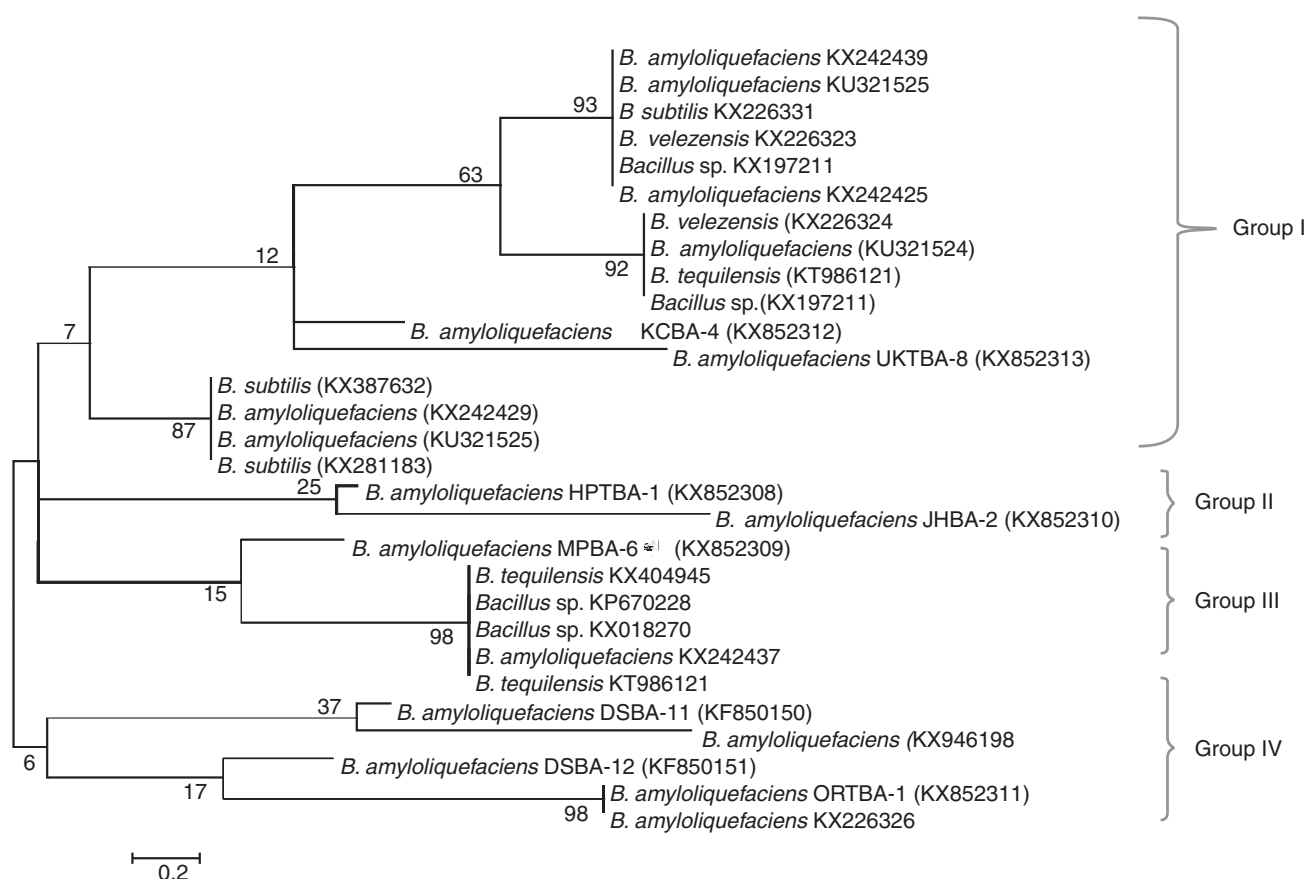


Fig 1 Phylogeny tree inferred from six isolates of *Bacillus amyloliquefaciens* based on 16S rRNA gene sequences from rhizosphere obtained from NCBI database.

chloride-perchloric acid reagent (Vikram *et al.* 2007) and total soluble protein estimation was done as described by Lowry *et al.* (1951). Production of siderophores was measured as described by Swanson *et al.* (2005).

Bio-efficacy and plant growth promoting ability of *B. amyloliquefaciens* under glasshouse conditions: Out of thirty nine isolates of *B. amyloliquefaciens*, six isolates, viz. HPTBA-1, MPBA-6, JHBA-6, ORTBA-1, KCBA-4 and UKTBA-8 were taken to evaluate their bio-efficiency against bacterial wilt disease of tomato under controlled conditions ($28 \pm 2^\circ\text{C}$) at National Phytotron Facility, ICAR- IARI, New Delhi. Seedlings of tomato cv. Pusa Ruby was grown and after 21 days they were transplanted into the pots (size 15 cm) containing autoclaved soil mixture of peat moss, vermiculite and sand in the ratio 2:1:1. The culture of *R. solanacearum* UTT-25 and bioagents harvested after 48 h growth from the Petri plates and mixed it in sterile distilled water to maintain 0.1 OD. About 5.0 ml culture of *R. solanacearum* was inoculated after 5 days of transplanting at root zone of the plants. Then 5.0 ml culture of *B. amyloliquefaciens* isolates was inoculated at root zone of the plants (Table 3). A set of plants treated only *R. solanacearum* and untreated plants were also maintained as positive and negative control. Wilt intensity was recorded at 6 days intervals up to 30 days of transplanting using 1–5 scale and biological control efficacy (BCE) of *B. amyloliquefaciens* isolates was estimated as

described Singh *et al.* (2016). The plant growth parameters in terms of plant height (cm), fresh and dry weight (g) of root and shoot were assayed. The comparative growth promotion efficacy (GPE) by *B. amyloliquefaciens* was calculated as described by Singh *et al.* (2016).

Data analysis: The analysis of variance for antagonistic ability was performed by using standard procedure of Gomez K A. (1984). Mean comparisons were conducted using a least significant difference (LSD) test ($P = 0.05$).

RESULTS AND DISCUSSION

Characterization of *Bacillus* spp.: Out of 123 isolates of *Bacillus* grown on Luria agar medium after 48 h, 39 isolates were characterized as *Bacillus* spp. (Table1) based on milky white, flat and dry colonies. Gram positive, rods shaped, cells often form chains and having peritrichous flagella. They were similar in morphological characters as reported earlier by Singh *et al.* (2016). This was further confirmed by biochemical test, specific for *Bacillus* species as per Bergey's Manual of Determinative Bacteriology. A representative of 39 isolates, six isolates HPTBA-1, MPBA-6, JHBA-2, ORTBA-1, KCBA-4 and UKTBA-8 of *B. amyloliquefaciens* were characterized at molecular level using universal primer of bacteria based on partial sequence analysis (=709 bp) of 16S rRNA and they showed pair-wise sequence similarity >99% with *B. amyloliquefaciens* and

Table 2 Evaluation of antagonistic and plant growth promoting attributes of *B. amyloliquefaciens* strains

<i>B. amyloliquefaciens</i> isolates	Area of inhibition zone against <i>R. solanacearum</i> (cm ²)	Phosphorus solubilization (µg/ml)	IAA production (µg ml)	Siderophores production [Diameter of the orange yellow halo produced (cm)]
MPBA-1	2.4 ^{hi}	51.61 ^k	71.42 ^{gh}	1.33 ^{cd}
MPBA-2	1.6 ^{no}	29.84 ^v	69.95 ^{gh}	1.01 ^{cd}
MPBA-3	1.1 ^p	27.51 ^x	59.26 ^{ij}	0.85 ^d
MPBA-4	0.86 ^p	23.84 ^z	41.25 ^l	0.52 ^d
MPBA-5	1.6 ^{no}	15.23 ^{zg}	72.41 ^{fg}	1.07 ^{cd}
MPBA-6	2.6 ^{efgh}	57.52 ⁱ	65.25 ^{hi}	1.54 ^{bcd}
MPBA-7	2.5 ^{gh}	49.45 ^l	62.2 ⁱ	1.12 ^{cd}
MPBA-8	1.81 ^{mno}	59.52 ^g	41.39 ^l	1.16 ^{cd}
MPBA-9	2.2 ^{ij}	17.58 ^{zf}	48.9 ^{jk}	1.14 ^{cd}
ORTBA-1	3.4 ^b	79.76 ^b	81.24 ^{cde}	1.87 ^{abc}
ORTBA-2	1.96 ^{klm}	21.56 ^{za}	61.2 ⁱ	1.33 ^{bcd}
ORTBA-3	2.53 ^{fgh}	27.54 ^x	93.26 ^{ab}	1.08 ^{cd}
ORTBA-4	2.76 ^{efg}	41.43 ^p	81.26 ^{cde}	1.51 ^{bcd}
ORTBA-5	1.66 ^{no}	19.56 ^{zb}	41.3 ^l	0.85 ^a
ORTBA-6	2.8 ^{ef}	31.25 ^u	53.55 ^{jk}	1.36 ^{cd}
KCBA-1	1.66 ^{no}	31.43 ^t	41.4 ^l	0.9 ^a
KCBA-2	1.83 ^{lmn}	19.25 ^{zc}	81.25 ^{cde}	0.74 ^a
KCBA-4	2.16 ^{ij}	29.59 ^w	71.40 ^{gh}	0.61 ^a
HPTBA-1	3.3 ^b	75.67 ^c	37.59 ^l	1.74 ^{bcd}
HPTBA-2	3.06 ^{cd}	71.4 ^e	95.4 ^a	1.62 ^{bc}
JHBA-1	2.13 ^j	59.59 ^g	75.9 ^{fg}	1.87 ^{bcd}
JHBA-2	2.83 ^e	45.38 ⁿ	83.27 ^{cd}	1.21 ^{bc}
JHBA-3	2.73 ^{efg}	17.6 ^{lzf}	61.3 ⁱ	1.45 ^{bcd}
JHBA-4	1.9 ^{klmn}	17.88 ^{ze}	41.4 ^l	0.6 ^a
UKTBA-1	3.2 ^{bc}	71.54 ^{de}	62.35 ⁱ	1.24 ^{abc}
UKTBA-2	2.6 ^{efgh}	61.39 ^f	53.31 ^{jk}	1.4 ^{bcd}
UKTBA-3	2.4 ^{hi}	58.33 ^h	83.89 ^{cd}	1.16 ^{abc}
UKTBA-4	1.6 ^{no}	43.58 ^o	71.35 ^{gh}	1.76 ^{abcd}
UKTBA-5	2.73 ^{efg}	55.85 ^j	29.56 ^m	0.93 ^a
UKTBA-6	1.6 ^{no}	19.41 ^{zbc}	62.56 ⁱ	1.02 ^{abcde}
UKTBA-7	2.1 ^{jk}	34.9 ^r	71.29 ^{gh}	1.53 ^{abc}
UKTBA-8	3.9 ^a	81.59 ^a	98.4 ^a	1.96 ^{abc}
UKTBA-9	1.8 ^{lmno}	17.62 ^{zf}	78.27 ^{def}	1.34 ^{bcd}
UKTBA-10	1.53 ^{no}	34.4 ^s	59.30 ^{ij}	1.54 ^{bcd}
UKTBA-11	2.73 ^{efg}	71.63 ^d	71.35 ^{gh}	1.63 ^{abcd}
UKTBA-12	3.3 ^{bc}	24.37 ^y	93.3 ^{ab}	1.85 ^{abcd}
UKTBA-13	2.03 ^{jkl}	18.36 ^{zd}	87.6 ^{bc}	0.94 ^a
UKTBA-14	1.7 ^{mno}	19.4 ^{zc}	96.29 ^a	0.85 ^a
UKTBA-15	1.63 ^{no}	37.56 ^q	85.28 ^{bc}	0.75 ^a
DSBA-11*	3.46 ^b	46.58 ^m	97.41 ^a	0.81 ^a
DSBA-12*	3.2 ^{bc}	41.44 ^p	95.20 ^a	0.89 ^a

* Reference strains of *Bacillus amyloliquefaciens*. Means ±SE (standard error) followed by the same letter do not differ significantly according to Duncan's multiple range test at P=0.05. Scheffe post hoc test. Means sharing different alphabetical (a, b, c, d, e) superscripts in a column significantly different (P <0.05).

other *Bacillus* spp. (Fig 1). Combination of phenotypic, physiological tests and 16S rRNA sequence analysis, these 39 isolates were belonging to *B. amyloliquefaciens*. The nucleotide sequences of six isolates *B. amyloliquefaciens* HPTBA-1, MPBA-6, JHBA-2, ORTBA-1, KCBA-4 and UKTBA-8 were submitted to Gene bank NCBI and obtained accession number as KX852308, KX852309, KX852310, KX852311, KX852312, KX852313) respectively.

In this study, out of 39 isolates of *B. amyloliquefaciens*, maximum 23 isolates were isolated from tomato, 9 from potato, 3 from chilli and 4 isolates from brinjal (Table 1). A plenty of antagonistic and plant growth promoting bacteria are found in the rhizosphere of solanaceous crops and majority of them belonging to *Bacillus* species and *Bacillus* derived genera especially from the rhizospheric soil of tomato plants (Tan *et al.* 2013, Singh *et al.* 2016).

Antagonistic ability of *B. amyloliquefaciens*: *In vitro* study, 39 isolates of *B. amyloliquefaciens* along with DSBA-11 and DSBA-12 used as positive control were tested for their antagonistic ability against *R. solanacearum* UTT-25 on CPG medium. Among them, the isolate UKTBA-8 of *B. amyloliquefaciens* isolated from wilted tomato rhizosphere was inhibited maximum growth of *R. solanacearum* (3.96 cm²) followed by isolate HPTBA-1 (3.3 cm²), MPBA-6 (2.56 cm²) and JHBA-2 (2.83 cm²) (Table 2). However, the other strains had a wide range of antagonistic activity to form inhibition zone ranging from 0.866–3.96 cm². The variation in formation of inhibition zone by *B. amyloliquefaciens* isolates against *R. solanacearum* is directly related to the type of secondary metabolites produced by the bacterial isolates particularly in mechanism of (Aliye *et al.* 2008).

Stein (2005) reported that *Bacillus* spp. has the potential to produce >24 structurally diverse antimicrobial compounds, which affects their inhibitory activity against various plant pathogens. The formation of inhibition zone is also affected by providing FeCl₃ into the KB medium to increase the antibacterial activity of *B. amyloliquefaciens* CM-2 and T-5 against *R. solanacearum* (Tan *et al.* 2013).

Plant growth promoting attributes: All the isolates showed positive in solubilization of phosphorus. The isolate UKTBA-8 solubilized the highest phosphorus (81.59 µg/ml) followed by ORTBA-1 (79.76 µg/ml), UKTBA-11 (71.63 µg/ml), and UKTBA-11 (71.54 µg/ml) (Table 2). The bacteria isolated from soil and endophytes have ability to solubilize phosphorus (Mehta and Nautiyal 2001) which are supported our findings. It was observed that all the isolates had ability to produce siderophores but variation ranging from 0.52–1.96 cm diameter. Maximum orange yellow halo zone (1.96 cm in diameter) on chromoazurool-S medium was formed by isolate UKTBA-8 followed by UKTBA-11 (1.85 cm) due to siderophores production, which is another plant growth promoting attributes. This was confirmatory to earlier report as rhizobacteria and endophytes (Beneduzi *et al.* 2008) produce siderophores, which is an iron chelating, compounds. These isolates have also ability to produce IAA with range of 41.25–97.41 µg/ml. However, isolate DSBA-11 produced the highest amount of IAA (97.41 µg/ml) followed by UKTBA-14 (96.29 µg/ml), and UKTBA-8 (93.4 µg/ml) (Table 2) and similar results has also been reported by Idris *et al.* (2007) in strain of *B. amyloliquefaciens* FZB42, produces IAA to promote the plant growth.

Table 3 Reduction of bacterial wilt disease intensity and enhancement of biomass of tomato plants treated with best six antagonistic *B. amyloliquefaciens* isolates

Treatments	Wilt disease incidence (%)	Biocontrol efficacy (%)	Length of tomato plant (cm)		Dry weight (g/ plant)		GPE (%) based on dry wt. of root and shoot
			Root	Shoot	Root	Shoot	
<i>B. amyloliquefaciens</i> HPTBA-1 + <i>R. solanacearum</i> UTT-25	31.53c	58.71	3.41e	44.33d	0.62b	1.19f	94.62
<i>B. amyloliquefaciens</i> MPBA-1 + <i>R. solanacearum</i> UTT-25	21.25e	71.1	3.12f	47.60b	0.54d	1.31c	98.92
<i>B. amyloliquefaciens</i> JHBA-2 + <i>R. solanacearum</i> UTT-25	27.32d	62.84	3.93d	45.33c	0.47f	1.22e	81.72
<i>B. amyloliquefaciens</i> ORTBA-1 + <i>R. solanacearum</i> UTT-25	20.87f	71.61	2.95g	29.39h	0.486f	1.12h	72.04
<i>B. amyloliquefaciens</i> KCBA-4 + <i>R. solanacearum</i> UTT-25	37.60b	48.86	3.94d	31.52g	0.71a	1.16g	101.07
<i>B. amyloliquefaciens</i> UKTBA-8 + <i>R. solanacearum</i> UTT-25	12.23h	83.36	7.15a	51.33a	0.7a	1.93a	182.79
<i>B. amyloliquefaciens</i> DSBA-11 + <i>R. solanacearum</i> UTT-25	17.95g	75.58	6.52b	39.52e	0.59 c	1.72b	148.38
<i>R. solanacearum</i> UTT-25 only	73.53a	-	2.60h	21.5i	0.39f	0.83h	31.18
Un-inoculated	0i	-	4.59c	35.6f	0.12e	0.81d	-

Means ±SE (standard error) followed by the same letter do not differ significantly according to Duncan's multiple range test at P = 0.05. Scheffe post hoc test. Means sharing different alphabetical (a, b, c, d, e) superscripts in a column significantly different (P < 0.05)

Biocontrol of bacterial wilt disease and plant growth attributes: Out of 39 isolates of *B. amyloliquefaciens*, six isolates HPTBA-1, MPBA-6, JHBA-2, ORTBA-1, KCBA-4 and UKTBA-8 with one positive strain DSBA-11 having better antagonistic and plant growth promoting ability were selected to test their efficacy to reduce bacterial wilt disease incidence and promote growth of tomato cv. Pusa Ruby under glass house conditions. The wilt disease was initiated after 5 days of inoculation of *R. solanacearum* (UTT-25), while *B. amyloliquefaciens* treated plant delayed appearance of wilt disease 8-10 days. Minimum disease incidence 12.23% was recorded in isolate UKTBA-8 treated plants followed by ORTBA-1 (20.87%) and MPBA-6 (21.25%) with maximum biocontrol efficacy of *B. amyloliquefaciens* UKTBA-8 (83.36%) followed by ORTBA-1 (71.76%) and MPBA-6 (71.1%) after 30 days (Table 3). The isolates UKTBA-8, ORTBA-1 and MPBA-6 of *B. amyloliquefaciens* showed better bio-efficacy to reduce wilt incidence in tomato under glasshouse conditions in accordance with the earlier reports, in tomato (Singh *et al.* 2016, Cao *et al.* 2018) and potato (Aliye *et al.* 2008). In plant growth promoting ability of these isolates, maximum shoot length of tomato plant (51.33 cm) was recorded in UKTBA-8 followed by MPBA-6 (47.60 cm) and JHBA-6 (45.33 cm), whereas, maximum root length was recorded in UKTBA-8 (7.15 cm) followed by KCBA-4 (3.94 cm) after 30 days of inoculation (Table 3). Maximum root dry weight (0.71 g) and shoot weight (1.93 g) was recorded in isolate UKTBA-8 treated plants with higher growth promoting efficacy 182.78% in UKTBA-8 treated plants followed by DSBA-11 (148.4%), and KCBA-4 (101.07%). Accordingly to our findings, the *B. amyloliquefaciens* isolates were positive in phosphorus solubilization, IAA and siderophores production. It indicates that *B. amyloliquefaciens* isolated from rhizosphere of tomato showed both antagonistic as well as plant growth promoting ability *in vitro* and under glass house conditions. Similarly, Yanti *et al.* (2018) reported that plant growth promoting rhizobacteria (PGPR) like *Bacillus* species act as biocontrol control agent for the suppression of bacterial wilt disease of plants and also promote the plant growth.

It was noticed during study that variation in antagonistic ability and in plant growth promoting attributes was found among the isolates of *B. amyloliquefaciens* isolated from different agro-climatic areas. Among them, isolate UKTBA-8 was found most effective in both controls of bacterial wilt disease as well as promoting plant growth of tomato.

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