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Short communication

Alteration of tomato fruit quality by root inoculation with plant growth-promoting rhizobacteria (PGPR): *Bacillus subtilis* BEB-13bs

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Abstract

In this study the effect of inoculation of tomato (*Lycopersicon esculentum* Mill.) roots with plant growth-promoting rhizobacteria (PGPR) on yield and fruit quality was evaluated. The control treatment was non-inoculated (CTL) and the PGPR treatment was inoculated with *Bacillus subtilis BEB-lSbs* (BS13). Yield per plant and marketable yield, as well as fruit weight and length were increased by the BS13 treatment when compared to the CTL treatment. Texture of red fruits was also enhanced by the BS13 treatment compared to that in the CTL treatment. These results demonstrated that PGPR have positive effects on tomato fruit quality attributes, particularly on size and texture.

Keywords: Plant growth-promoting rhizobacteria; *Bacillus*; Quality; Texture; Tomato

1. Introduction

The challenge of producing fresh fruits and vegetables is increasing both yield and quality to satisfy consumers avoiding deleterious effects on the environment (Mader et al., 2002). Therefore, the utilization of biofertilizers has become a feasible production practice. Many marketable biofertilizers are mainly based on plant growth-promoting rhizobacteria (PGPR) that exert beneficial effects on plant development often related to the increment of nutrient availability to host plant (Vessey, 2003). However, not all the PGPR strains exert their positive effect on plant growth via increasing nutrient status of host plants. PGPR seem to promote growth through suppression of plant disease (Zehnder et al., 2001), or through production of phytohormones and peptides acting as biostimulants (Glick et al., 1998; Jimenez-Delgadillo, 2004).

On the other hand, agricultural product quality is affected by both pre and postharvest factors. Preharvest factors influencing the quality of the harvested product include biological factors (pathological, entomological and animal); physiological factors (physiological disorders, nutritional imbalances and maturity) and cultural factors (fertilization and growth regulators) among others (Mattheis and Fellman, 1999; Kays, 1999; Sams, 1999). Thus, PGPR could be considered as preharvest biotic factors affecting crop yield and quality. It is known that sensory attributes of fruit quality appearance (shape, size and color) and texture are influenced by ripening (Hobson and Grierson, 1993). The objective of this work was to determine the effect of tomato root inoculation with PGPR on yield and tomato fruit quality.

2. Materials and methods

2.1. Greenhouse experiments

Two greenhouse experiments were set as follows: tomato (*Lycopersicon esculentum* Mill, cv Rio Fuego) was grown from seeds in a mixture of peat and perlite (1:1, v/v), and seedlings were transplanted after 21 days separately into 3500 cm^3 pots containing 5 kg of a mixture methyl bromide sterilized coarse sand and sandy loam soil (1:1, v/v). PGPR were pre-selected mainly for their plant growth stimulation in *Arabidopsis thaliana* under greenhouse conditions (Jimenez-Delgadillo, 2004). *Bacillus subtilis* BEB-13bs strain was grown for 12 h at 28 °C in modified potato-dextrose-agar medium (MPDA) in which agar was not added (Johnson and Curl, 1972). Bacterial suspension was adjusted to yield 1×10^7 CFU ml^{-1} using MPDA. For treatments, either 3 ml of MPDA medium (CTL treatment) or

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3 ml of bacteria suspension (BS13 treatment) were applied into 8 cm deep holes made in pots for transplanting. Plants were irrigated as needed and fertilized weekly with 300 ml per pot of Long Ashton Solution (Hewitt, 1966).

Plants height, shoot (leaves + stem) dry weight, roots length, root dry weight, yields and fruit quality evaluation were determined after 50 days of transplant.

Shoot (leaves + stem) and root dry weights of four plants were determined after drying at 80 °C for 48 h to constant weight. Each plant was a single replication and there were four replications per treatment ($n = 4$).

2.2. Yield and quality evaluation

All red fruits were harvested from four plants per treatment and the total yield per plant was obtained. Fruits were graded into three categories according to length: economical grade (<4 cm in length), marketable grade (4–7 cm in length) and selected grade (>7 cm in length). The yield of each category was calculated as the percentage of yield per plant. Each plant was a single replication and there were four replications per treatment ($n = 4$).

Size was determined by measuring fruit fresh weight, length and diameter of all red fruits from four plants per treatment. Average weight, length and diameter per fruit were calculated. Each plant was a single replication and there were four replications per treatment ($n = 4$). For texture determinations fruits were harvested at three different ripening stages: breaker (B), light red (LR), and red (R) (USDA, 1991). Fruits were evaluated by a puncture test using a Texture Analyzer TA-XT2 (Stable Micro Systems), loading at 1 mm s^{-1} to a specified distance of 15 mm on two opposite points along the equatorial plane and the average maximum force per fruit (F_{max}) was recorded. A total of 10 fruits from each of the treatments and ripening stages were measured. Each fruit was a single replication and there were 10 replications per treatment ($n = 10$).

2.3. Experimental design and statistical analysis

Experiments consisted in eight plants per treatment randomly arranged. Four plants in each experiment were used to get biometric parameters and yields while four plants were used to evaluate fruit quality. Data were analyzed under a completely randomized design with the replications number indicated above. Statistical significance of the data was determined using analysis of variance (ANOVA). Mean separation was tested by minimum significant difference (MSD) at $P < 0.05$ (FAUANL, 1994).

Table 1
Effect of the growth promoting rhizobacteria *Bacillus subtilis BEB-13bs* on root growth in tomato (*Lycopersicon esculentum* L. cv Río Fuego)

	Treatments	Root Length (cm)	Root dry weight (g)
Experiment 1	CTL	46 b	6.8 b
	BS13	53 a	8.6 a
Experiment 2	CTL	43 b	5.9 b
	BS13	51 a	7.8 a

CTL: non-inoculated control plants. BS13: *B. subtilis BEB-13bs*-inoculated plants. In each experiment means followed by different letter within each column are significantly different based on DMS test ($P < 0.05$, $n = 4$).

3. Results and discussion

Although the impact of root inoculation with beneficial rhizosphere microorganisms on some quality parameters is being explored (Charron et al., 2001; Kaya et al., 2003; Mena-Violante et al., 2006), this is the first report of physical changes in fruit due to PGPR root inoculation. The results have demonstrated that the PGPR strain *B. subtilis* BEB13-bs has capacity to modify some tomato fruit quality aspects. Both experiments showed that PGPR inoculation improved the radical system of tomato plants (Table 1). Root dry weight and root length increased significantly in plants of the BS13 treatment (18–26% and 13–15%, respectively) compared to those in the control treatment. No marked differences were detected in biometric parameters of aerial system (data not shown). Enhanced yield has been reported before for other crops (Baset-Mia et al., 2005), in a similar way, inoculation of tomato with PGPR led also to increasing yield per plant and marketable grade yield in the BS13 treatment which were higher than those found in the CTL treatment in the first experiment (21% and 6%, respectively) (Table 2). In the second experiment, yield per plant was 25% higher in the BS13 treatment compared to that found in the CTL treatment. Moreover, we found a power effect on marketable grade yield this time. The number of marketable fruits was significantly larger (20%) in the BS13 treatment than in the control treatment. These findings were in concordance with the improved size observed in fruits of the BS13 treatment (Table 2). Fruits in the BS13 treatment were significantly

Table 2
Effect of the growth promoting rhizobacteria *B. subtilis BEB-13bs* on yield and fruit size of tomato (*L. esculentum* Mill. Río Fuego)

	Treatments	Yield/plant (g)	Marketable grade yield %	Weight/fruit (g)	Length (cm)	Diameter (cm)
Experiment 1	CTL	648 b	66.0 a	45.1 b	5.3 b	4.1 a
	BS13	787 a	72.0 a	58.2 a	5.8 a	4.3 a
Experiment 2	CTL	675 b	44.5 b	62.7 b	4.9 b	4.6 a
	BS13	844 a	64.8 a	74.3 a	5.8 a	4.8 a

CTL: non-inoculated control plants. BS13: *B. subtilis BEB-13bs* -inoculated plants. Marketable grade: fruits 4–7 cm in length. In each experiment means followed by different letter within each column are significantly different based on DMS test ($P < 0.05$, $n = 4$).