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# EFFECTS OF FOLIAR APPLICATION OF PLANT GROWTH PROMOTING BACTERIUM ON CHEMICAL CONTENTS, YIELD AND GROWTH OF TOMATO (*LYCOPERSICON ESCULENTUM* L.) AND CUCUMBER (*CUCUMIS SATIVUS* L.)

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**Abstract**

This study was conducted to determine the effects of spraying a growth promoting bacterium on chemical contents, yield and growth of tomato (*Lycopersicon esculentum* L.) and cucumber (*Cucumis sativus* L.) vegetable species. Different strains of bacteria, *Bacillus subtilis* BA-142, *Bacillus megaeorium- GC subgroup A.* MFD-2, *Acinetobacter baumannii* CD-1 and *Pantoea agglomerans* FF, were used. The effects of the bacterial treatments on the plant nutrient elements of tomato and cucumber fruit were determined. Its effects on average fruit weight, fruit number per plant, fruit weight per plant, plant length, fruit width and length, total soluble solid and dry matter in tomato and cucumber fruit were also determined. The effects of bacterial application on plant mineral contents were significant. Bacterial applications increased mineral contents of tomato and cucumber fruit as compared to control treatment. All bacterial applications particularly affected on improving the in N, P, Mg, Ca, Na, K, Cu, Mn, Fe and Zn contents of the fruit. Growth promoting effects of bacterial application on the plant growth parameters except TSS (Total soluble solid) were significant. The highest average fruit weight, fruit weight per plant and plant length were obtained from *Pantoea agglomerans* FF applications in tomato as comparing to that of the other applications. Fruit number per plant was high in *Acinetobacter baumannii* CD-1 application and fruit width, fruit length and dry matter were highest in *Bacillus megaterium-GC subgroup A.,* MFD-2 application than that of the other application in tomato. The highest fruit number per plant, fruit weight per plant, plant length, fruit width, fruit length and dry matter were obtained from *Pantoea agglomerans* FF applications in cucumber as compared to that of the other applications and the highest average fruit weight was found in *Bacillus megatorium-GC subgroup A.* MFD-2 application when compared to the other applications. The results of this study showed that *Pantoea agglomerans* FF, *Acinetobacter baumannii* CD-1 and *Bacillus megaterium-GC subgroup A.* MFD-2 have a great potential to increase the yield, growth and mineral contents of tomato and cucumber vegetable species.

## Introductıon

Vegetables presently grown in Turkey consist of annual crops including Solanaceous, Crucifers, Cucurbits, Bulb crops, Leguminous and other indigenous vegetable species. In terms of economic value, nutrition, consumer’s preference, general adaptability and extent of cultivation, the most commonly grown vegetable crops are tomato, watermelon, cucumber, pepper, eggplant, squash, cabbage, onion, snap bean, melon, salad vegetables etc. Microorganisms such as bacteria are important to promote the circulation of plant nutrients and reduce the need of chemical fertilizers as much as possible. Intensive farming practices require extensive use of chemical fertilizers, which are costly and create environmental problems, for warranting high yield and quality. Hence, there has recently been a resurgence of interest in environmentally friendly, sustainable and organic agricultural practices (Esitken *et al*., 2005, 2006). Because of the reason, use of bio-fertilizers containing beneficial microorganisms instead of inorganic chemicals is positively known to affect on plant growth in terms of supplying of plant nutrients and may help to sustain environmental health and soil productivity (O'Connell, 1992, Eşitken *et al*., 2006). A number of inoculated bacterial species mostly associated with the plant rhizosphere have been tested and found to be beneficial for plant growth, yield and crop quality so far. They have been called plant growth promoting rhizobacteria (PGPR)' including the strains in the genera *Acinetobacter Alcaligenes, Arthrobacter, Azospirillium, Azotobacter, Bacillus, Beijerinckia, Burkholderia, Enterobacter, Erwinia, Flavobacterium, Rhizobium* and *Serratia* (Rodriguez & Fraga, 1999; Sturz & Nowak, 2000; Sudhakar *et al*., 2000, Eşitken *et al*., 2006). These bacteria were previously reported as plant growth promoting bacteria and had potential bio-control agents against a wide range of bacterial and fungal pathogens causing economically important problems in agriculture (Cuppels *et al*., 1999; Kotan *et al*., 1999; Cakmakci *et al*., 2001; Esitken *et al*., 2002, 2003, 2006). They affect on fixation of Nitrojen (Reis *et al*., 1994; Dobereiner, 1997; Vance, 1997) and are one of the most plausible mechanisms of action affecting plant growth (Eşitken *et al*., 2006). The reason is that $N_2$-fixing bacteria may be important for plant nutrition by increasing N uptake by the plants and playing as significant role as plant growth PGPR in the biofertilization of the crops (Marschner, 1995, Aslantaş *et al*., 2007). Many researchers determined that PGPR can stimulate growth and increase yield in pepper and tomato (Sahin *et al*., 2000), in sugar beet (Elkoca *et al*., 2001, Çakmakçı *et al*., 2006), in spring barley (Salantur *et al*., 2005), in apricot (Eşitken *et al*., 2003, Altindağ *et al*., 2006), in raspberry (Orhan *et al*., 2006), in apple (Aslantaş *et al*., 2007) and in rocket (Dursun *et al*., 2008). However, not much is known about their promoting effects on yield, growth and nutrient contents of tomato and cucumber vegetable species.

The objective of this study was to determine the effects of inoculation bacteria (*Bacillus subtilis* BA-142, *Bacillus megaterium- GC subgroup A.* MFD-2, *Acinetobacter baumannii* CD-1 and *Pantoea agglomerans* FF) on chemical contents, yield and growth of tomato and cucumber vegetable species in unheated greenhouse conditions.

## Materials and Methods

**Bacterial strains, culture conditions, media and treatment:** Strains of bacteria, *Bacillus subtilis* BA-142, *Bacillus megaterium-GC subgroup A.* MFD-2, *Acinetobacter baumannii* CD-1 and *Pantoea agglomerans* FF, were obtained from Department of Plant Protection at Ataturk University. Bacteria were grown on Nutrient Agar (NA, including 3 g/l beef extract, 5 g/l peptone and 15 g/l agar) for routine use, and maintained in Nutrient Broth (NB, including 3 g/l beef extract and 5 g/l peptone) with 30% glycerol at -80°C for long-term storage. For this experiment, the bacterial strains were grown on nutrient agar. A single colony was transferred to 250 ml flasks containing NB, and grown aerobically in flasks on a rotating shaker (95 rpm) for 48 h at 27°C. The bacterial suspension was then diluted in sterile distilled water to a final concentration of $10^8$ CFU $ml^{-1}$, and the resulting suspensions were used to treat tomato and cucumber plants. The plant leaves were sprayed with bacterial suspension ($10^8$ CFU $ml^{-1}$) until getting wet at ten days interval for three times during seedling development.

**Unheated greenhouse experiment:** The experiment was carried out on tomato and cucumber vegetable species in the Department of Horticulture at Ataturk University under unheated greenhouse condition in Erzurum, Turkey, in 2007. It was made based on a completely randomized design with three replicates. Seedlings of the plants were planted on 18 May in 2007. The experiment ended on 15 October in the same year.

The effect of the bacterial treatments on the plant nutrient elements of fruit in tomato and cucumber was evaluated. Growth promoting effects of bacterial treatments were, also, evaluated on 10 plants by determining average fruit weight, fruit number per plant, fruit weight per plant, plant length, fruit width and length, total soluble solid and dry matter in tomato and cucumber fruit.

**Fruit analysis:** In order to determine the mineral contents of plant fruit, the samples were oven-dried at 68°C for 48 h and then ground. The micro-Kjeldahl procedure was applied for determination of N. $K^{+}$, $Ca^{2+}$, and $Mg^{+2}$ after wet digestion of dried and ground sub-samples in a $H_2SO_4$-Se-*salisilic* acid mixture. In the diluted digests, P was measured spectrophotometrically by the indophenol-blue method and after reaction with ascorbic acid. Potassium and $Ca^{+2}$ were determined by flame photometry, $Mg^{+2}$, Mn, Zn and Cu by atomic absorption spectrometry using the method of AOAC (1990).

**Data analysis:** All data were subjected to a one-way analysis of variance (ANOVA) and separated by Duncan's multiple range tests using SAS statistical software (Anon., 1982).

## Results and Discussion

**Fruit mineral contents:** The effects of bacterial application on mineral (N, P, Mg, Ca, Na, K, Cu, Mn, Fe and Zn) contents of tomato and cucumber fruit were significant at $p<0.05$, 0.01 and 0.001 (Tables 1 and 2). All bacterial applications particularly affected on increasing in N, P, Mg, Ca, Na, K, Cu, Mn, Fe and Zn contents of the fruit. The highest N, P and Mg contents were obtained from BA-142, FF, MFD-2 applications in both of tomato and cucumber. The highest Ca, Na, K and Cu contents were obtained from FF, MFD-2, BA-142 and CD-1 applications in the both species. Mn, Fe and Zn contents were highest in *FF and MFD-2* application in tomato and cucumber, MFD-2 application in tomato, and BA-142 application in cucumber, respectively. Bacterial strains effecting on Na and Cu in tomato and Ca, K Mn and Fe in cucumber were not significant (Tables 1 and 2). The higher mineral contents in the bacteria treated plant may have resulted from the producing plant hormones (Aslantaş *et al*., 2007) ability of these bacteria, as reported that many kinds of bacteria had given same results on different plant species in previous studies (Sahin *et al*., 2000; Cakmakci *et al*., 2001; Esitken *et al*., 2003 and 2006, Orhan *et al*., 2006, Dursun *et al*., 2008). Marschner (1995) and Aslantaş *et al*., (2007) stated that increasing mineral contents in plants results in greater uptake of nutrient elements from soil. This evidence confirms the data showing that the quantity of N, P, K, Zn, Fe, Mn, Na, Ca and Mg was significantly or relatively increased in the bacteria-treated plants, which may be explained by higher concentration of $N_2$ and P stimulated by bacterial application and resulted from the producing plant hormone.

**Table 1. Effects of bacterial applications on fruit chemical contents of tomato.**

| | N (%) | P (%) | Mg (ppm) | Ca (ppm) | Na (ppm) | K (ppm) | Cu (ppm) | Mn (ppm) | Fe (ppm) | Zn (ppm) |
|---|---|---|---|---|---|---|---|---|---|---|
| Control | 2,37 b** | 0,42 bc*** | 4351,33 bc* | 12470,00 a** | 1147,00 NS | 15456,00 b*** | 13,33 NS | 24,00 cd** | 92,67 b* | 35,00 NS |
| FF | 2,44 b | 0,65 a | 4717,33 ab | 13514,00 a | 1240,00 | 17741,00 a | 14,00 | 41,00 a | 94,33 b | 30,00 |
| MFD-2 | 2,56 b | 0,45 bc | 4880,00 a | 12934,00 a | 1550,00 | 15993,33 b | 16,00 | 34,00 ab | 108,00 a | 33,33 |
| CD-1 | 2,53 b | 0,49 b | 4229, 33 c | 12992,00 a | 1116,00 | 16531,00 b | 16,67 | 22,00 d | 105,00 a | 31,67 |
| BA-142 | 2,80 a | 0,41 c | 4595,33 abc | 10730,00 b | 1054,00 | 18412,67 a | 16,33 | 3,002 bc | 92,67 b | 21,67 |
| LSD (0.05) | 0,21 | 0,08 | 436,44 | 1198,45 | | 1100,41 | | 8,22 | 9,96 | |

*= Significantly important at p<0.05
**= Significantly important at p<0.01
***= Significantly important at p<0.001
NS= Not significant
LSD= Least significant difference

**Table 2. Effects of bacterial applications on fruit chemical contents of cucumber.**

| | N (%) | P (%) | Mg (ppm) | Ca (ppm) | Na (ppm) | K (ppm) | Cu (ppm) | Mn (ppm) | Fe (ppm) | Zn (ppm) |
|---|---|---|---|---|---|---|---|---|---|---|
| Control | 2,15 c** | 0,27 c*** | 4392,00 b* | 13186,67 NS | 724,33 b** | 15590,33 NS | 17,00 ab* | 29,00 NS | 97,67 NS | 43,33 ab* |
| FF | 2,34 bc | 0,40 a | 4310,67 b | 14413,33 | 777,33 b | 15724,67 | 21,00 a | 32,00 | 96,33 | 35,00 b |
| MFD-2 | 2,54 b | 0,34 b | 5124,00 a | 13800,00 | 795,00 b | 15254,33 | 22,00 a | 33,00 | 80,67 | 50,00 a |
| CD-1 | 2,27 bc | 0,30 bc | 4554,67 b | 13309,33 | 671,33 b | 15926,33 | 23,00 a | 31,00 | 90,00 | 43,33 ab |
| BA-142 | 2,87 a | 0,33 b | 4473,33 b | 13370,67 | 1148,33 a | 16934,33 | 13,00 b | 29,00 | 91,67 | 48,33 a |
| LSD (0.05) | 0,28 | 0,04 | 506,13 | | 189,60 | | 6,14 | | | 8,14 |

*= Significantly important at p<0.05
**= Significantly important at p<0.01
***= Significantly important at p<0.001
NS= Not significant
LSD= Least significant difference

**Plant growth:** Growth promoting effects of bacterial application on fruit weight, fruit number per plant, fruit weight per plant, plant length, fruit width and length and dry matter of tomato and cucumber fruit were significant and its effect on TSS (Total Soluble Solid) was not significant in both of the vegetable species (Tables 3 and 4). The highest average fruit weight (124,50g), fruit weight per plant (6120,57g) and plant length (368,33cm) were obtained from *Pantoea agglomerans* FF applications in tomato as compared to that of the other applications. Fruit number per plant (52,20) was high in *Acinetobacter baumannii* CD-1application and fruit width (70,50mm), fruit length (59,86mm) and dry matter (4,61%) were higher in *Bacillus megaterium- GC subgroup A.* MFD-2 application than that of the other application in tomato (Table 3). The highest fruit number per plant (68,07), fruit weight per plant (8857,47g), plant length (246,89cm), fruit width (37,76mm), fruit length (17,36cm) and dry matter (4,63%) were obtained from *Pantoea agglomerans* FF applications in cucumber as compared to that of the other applications and the highest average fruit weight (133,95g) was determined in *Bacillus megaterium-GC subgroup A.* MFD-2 application when compared to the other applications (Table 4). This is the first report on growth promoting effect of bacterial application on plant growth parameters of tomato and cucumber in our region. However, similar reports were obtained in different plant species. Researchers stated that bacterial applications including *Pseudomonas* and *Bacillus* strains can stimulate growth and increase yield in pepper and tomato (Kotan *et al*., 1999, Sahin *et al*., 2000), in sugar beet (Elkoca *et al*., 2001, Çakmakçı *et al*., 2006), in spring barley (Salantur *et al*., 2005), in apricot (Eşitken *et al*., 2003, Altindağ *et al*., 2006), in raspberry (Orhan *et al*., 2006) in apple (Aslantaş *et al*., 2007) and in rocket (Dursun *et al*., 2008). The reason of growth promoting effect of bacterial applications on plant growth is that they affect on fixation capacity of Nitrogen (Reis *et al*., 1994; Dobereiner, 1997; Vance, 1997) and are one of the most plausible mechanism of action affecting plant growth (Eşitken *et al*., 2006). The used bacterial strains showed the same results in our findings. Thus, our finding is in good agreement to previous studies mentioned above.

In conclusion, the effects of bacterial applications depend on the crop species. Bacterial application is safe, effective and easily adopted by farmers. The results of this study showed that *Pantoea agglomerans* FF, *Acinetobacter baumannii* CD-1, *Bacillus subtilis* BA-142 and MFD-2 have a great potential especially *Pantoea agglomerans* FF, *Acinetobacter baumannii* CD-1 and *Bacillus megaterium-GC subgroup A.* MFD-2 to increase the yield, growth and mineral contents of tomato and cucumber vegetable species. Such bacterial strains have the potential to benefit farmers in many ways and hence, its importance must be recognized by farmers as well as researchers. Therefore, they may be put to good use as biofertilizer for vegetable production in sustainable and especially ecological agricultural systems.

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**Table 3. Effects of spraying a growth promoting bacterium on yield, plant growth, total soluble solid and dry meter in tomato fruit.**

| Application | Average fruit weight (g) | Fruit number per plant | Fruit weight per plant (g) | Plant length (cm) | Fruit width (mm) | Fruit length (mm) | TSS (%) | Dry meter (%) |
|---|---|---|---|---|---|---|---|---|
| Control | 110,42 c** | 38,53 c*** | 4254,13 c*** | 338,34 c*** | 66,24 c* | 57,14 c** | 3,48 NS | 3,84 c*** |
| FF | 124,50 a | 49,17 b | 6120,57 a | 368,33 a | 67,14 bc | 57,36 bc | 3,63 | 4,06 b |
| MFD-2 | 117,65 b | 51,40 ab | 6046,73 a | 361,00 ab | 70,50 a | 59,86 a | 3,63 | 4,61 a |
| CD-1 | 117,22 b | 52,20 a | 6117,70 a | 364,17 ab | 69,39 ab | 58,28 bc | 3,63 | 4,13 b |
| BA-142 | 109,72 c | 50,50 ab | 5533,70 b | 354,00 b | 69,65 ab | 58,58 b | 3,47 | 4,06 bc |
| LSD (0.05) | 6,10 | 2,81 | 278,82 | 13,34 | 2,64 | 1,18 | | 0,17 |

*= Significantly important at $p<0.05$
**= Significantly important at $p<0.01$
***= Significantly important at $p<0.001$
NS= Not significant
LSD= Least significant difference
TSS= Total soluble solid

**Table 4. Effects of spraying a growth promoting bacterium on yield, plant growth, total soluble solid and dry meter in cucumber fruit.**

| Application | Average fruit weight (g) | Fruit number per plant | Fruit weight per plant (g) | Plant length (cm) | Fruit width (mm) | Fruit length (mm) | TSS (%) | Dry meter (%) |
|---|---|---|---|---|---|---|---|---|
| Control | 126,71 bc** | 56,70 c*** | 7182,60 d*** | 218,17 b** | 34,75 d*** | 15,84 b** | 2,96 NS | 2,56 c*** |
| FF | 130,14 b | 68,07 a | 8857,47 a | 246,89 a | 37,76 a | 17,36 a | 2,93 | 4,63 a |
| MFD-2 | 133,95 a | 62,10 b | 8314,70 b | 238,50 a | 35,73 c | 16,92 a | 3,08 | 4,16 a |
| CD-1 | 125,74 c | 62,10 b | 7803,07 c | 246,34 a | 36,99 b | 16,83 a | 3,03 | 3,49 b |
| BA-142 | 129,45 b | 55,00 c | 7121,67 d | 234,33 a | 36,52 b | 17,38 a | 2,94 | 3,20 b |
| LSD (0.05) | 3,43 | 4,28 | 442,90 | 13,98 | 0,66 | 0,67 | - | 0,52 |

*= Significantly important at $p<0.05$
**= Significantly important at $p<0.01$
***= Significantly important at $p<0.001$
NS= Not significant
LSD= Least significant difference
TSS= Total soluble solid

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