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# A Study of Different Bacterial Formulations in Increasing the Nutrient Content of Bulb and Leaf of Tulips and Grown Soil Samples

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**Abstract:** This study was carried out in an open field under black plastic mulch conditions in order to determine the nutritional element contents of the bulb, leaf and cultivation medium of tulips by cultivating tulip bulbs being accepted as the reserve of nutrients without using any chemical or organic material and injecting different PGPR formulations into the root area in 2013. Three different tulip varieties belonging to *Tulipagesneriana* L. widely used in parks and gardens in our country were used as plant material in the study. The study was comprised of 5 applications, Formulation A (*Pantoeaagglomerans* RK-79 + *Pantoeaagglomerans* RK-92), formulation B (*Pantoeaagglomerans* RK-79 + *Pantoeaagglomerans* RK-92 + *Bacillus megaterium* TV-91C + *Bacillus subtilis* TV-17C), formulation C (*PantoeaAgglomerans* RK-79 + *Pantoeaagglomerans* RK-92 + *Bacillus megaterium* TV-3D + *Paenibacilluspolymyxa* TV-12E), formulation D (*Pantoeaagglomerans* RK-79 + *Pantoeaagglomerans* RK-92 + *Bacillus megaterium* TV-6D + *Pseudomonas putida* TV-42A) and the control (without fertilizer / bacteria application). On the basis of the examined properties, it has been determined that 'variety x applications' has a general interaction. The highest total nitrogen (2.53%) and P (0.34%) amounts were determined in the bulbs of the Pink Impression variety in Formation C compared to the control application. The highest total nitrogen, Ca, S, Mg and Fe amounts in leaves was determined in the Formulation C application. The highest available nitrogen obtained from soil samples (31.22 mg $kg^{-1}$), calcium (15.27 mg $kg^{-1}$), magnesium (2.94 mg $kg^{-1}$) and manganese (3.78 mg $kg^{-1}$) were determined in the Formulation C application while the highest available phosphorus (18.80 mg $kg^{-1}$), Fe (1.90 mg $kg^{-1}$), Zn (1.64 mg $kg^{-1}$) and Pb (0.13 mg $kg^{-1}$) were obtained in the Formulation D application. It has been determined that Formulation C and Formulation D applications in particular have important effects on bulb, leaf and soil nutrient element content. It has been concluded that this information can be used to develop more efficient and environmentally friendly fertilizer management plans for commercial bulb production and landscape use.

**Key words:** PGPR, *Tulipagesneriana* L. · Macro-micro element · Bacteria formulation

## INTRODUCTION

The tulip (*Tulipagesneriana*L.), is a perennial bulbous plant with flashy flowers in the genus *Tulipa* belonging to the family *Liliaceae*. The tulip has become one of the most important ornamental plants in the world [1]. One of the methods used by enterprises dealing in the bulbous plant trade is to produce with bulblets to replicate the same main plant and generate plenty of production. In the production of tulips for outdoor ornamental plants, cut flowers and potted plants, as production materials, bulbs are used once and they are renewed every year [2, 3]. Therefore, the cost of bulbs is the most important input of tulip production.

The size limits and usage of bulbs vary according to the type of culture. As bulbs grow, they have the strength to deliver better quality flowers. This increases in the commercial value of this type of bulb and ensures more bulblet production characteristics [4]. It is believed that the bulb can supply all nutrients that the plant needs, which has not been shown yet. However, a great heterogeneity in the yield and quality of bulbs is observed in tulip cultivation [5]. One of the solutions to this heterogeneity in bulb yield and quality is the addition of nutrients to the cultivation medium in tulip cultivation. Furthermore, since the development of tulip plants is as short as a few months, fertilization or plant feeding in the first period (green bud period) is especially important for

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the best completion of this process and for the bulb growth expected from the plant itself [6, 7]. While doing so, maintaining the production area is an important consideration. This is necessary for the continuation of production and the development of the market. PGPR application is assigned to increase in the plant growth and yield as well as develop soil quality [8, 9].

Asymbiotic nitrogen fixation, increasing in the solubility of inorganic phosphorus and mineralization of organic phosphorus compounds, the production of iron and organic acids through the production of siderophore and increasing in the uptake of some other trace elements and beneficial bacteria promote growth by improving the mineral nutrition of plants [10, 11]. The bacteria, appropriately called rhizobacteria, of the habitat of which is located in a zone surrounding the roots of the plants or rhizosphere are known as plant growth promoting rhizobacteria (PGPR) [10]. Scientific studies involve interdisciplinary approaches to understand the adaptation of PGPR to the rhizosphere, effects on plant physiology and growth, mechanisms of root colonization [12], biofertilization[13], induced systemic resistance, biocontrol of plant pathogens [14-16], production of determinants, etc. regarding plant growth.

There are several reports showing that PGPR have promoted the growth and reproductive parameters of plants ranging from vegetable crops [17-18], fruits [19] and field crops [20-22]. There are a few studies using PGPR as a plant growth promoting agent in the cultivation of ornamental plants around the world [23-25]. PGPR have gained a worldwide importance and acceptance for agricultural benefits. Plant growth-promoting bacterial effects may vary depending on the bacterial species and number, plant-bacteria combination, plant genotype, development period, harvest date, plant parameters, soil type, organic matter amount and environmental conditions [26-27].

A study has been carried out in order to determine the nutritional element contents of the bulb, leaf and cultivation medium of tulips by cultivating tulip bulbs being accepted as the reserve of nutrients without using any chemical or organic material and injecting different PGPR formulations into the root area. In many previous studies [23-25], only the effects of single bacterial strains have been studied whereas in our study the impact of different bacteria formulations established with bacteria strains have been examined. The objective is to benefit from the most appropriate bacterial formulation in the cultivation of tulip bulbs based on the obtained results.

## MATERIALS AND METHODS

The experiment was conducted in black inorganic mulch in field conditions at the Department of Horticulture of Agriculture Faculty, Atatürk University, Erzurum, Turkey in 2013. The soil texture was sandy-loamy and the general properties of the field soil are given in Table 1. A total of 360 bulbs of Pink Impression, Blue Aimable and Golden Parade cultivars belonging to *Tulipagesneriana* L. species were used in the experiments. The selected bulbs were free of cuts and rot and as homogeneous as possible in size (10 to 12 cm perimeter).

The region altitude is 1853 m and the climate is cold. According to the climatic values measured at the 12$^{th}$ Regional Directorate of Meteorology (Erzurum) between the months of January-August in 2013, the mean temperature is 6.88 °C. Annual rainfall is 28.24 kg/m$^2$ and average relative humidity is 66.20% (Table 2).

Research was established in a completely randomized design with 3 replications and there were 8 plants in each replication. All of the bacterial strains (*Pantoeaagglomerans* RK-79, *Pantoeaagglomerans* RK-92, *Bacillus megaterium* TV-91C, *Bacillus subtilis* TV-17C, *Bacillus megaterium* TV-3D, *Paenibacilluspolymyxa* TV-12E, *Bacillus megaterium* TV-6D, *Pseudomonas putida* TV-42A) were obtained from the culture collection unit in the Department of Plant Protection, Faculty of Agriculture at Atatürk University (Table 3).

There were 5 applications in the study: (1) Formulation A (RK-79 +RK-92), (2) Formulation B *(*RK-79 + RK-92 + TV-91C +TV- 17C),(3) Formulation C (RK-79 + RK-92 + TV-3D + TV-12E)*,* (4) Formulation D (RK-79 + RK-92 + TV-6D + TV-42A)and(5) Control (untreated bacteria) (Table 4).

The bulbs were planted in black inorganic mulch conditions in a field; subsequently 5 ml of prepared bacterial formulation was injected into the planting zone into each of the bulbson April 17 in 2013 and the bulbs were harvested on July 05 in 2013. The growing process

Table 1: The general properties of the field soil used in the experiment

| Soil properties | Values |
|---|---|
| pH (1:2,5 water) | 6.90 |
| Organic matter (%) | 2.48 |
| $CaCO_3$ (%) | 1.04 |
| Texture | Sandy -Loamy |
| N (%) | 0.002 |
| P (ppm) | 23.62 |
| K (ppm) | 996.45 |
| Ca (ppm) | 2794.00 |
| Mg (ppm) | 518.30 |

Table 2: The climatic values measured between the months of January-August in 2013 of Erzurum province (12th Regional Directorate of Meteorology (Erzurum-Turkey))

| | Months | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Meteorological Elements | I | II | III | IV | V | VI | VII | VIII | IX | X | XI | XII | Annual average |
| Mean temperature (°C) | -9.5 | -7.4 | -0.8 | 7.2 | 11.6 | 15.0 | 19.4 | 19.5 | | | | | 6.88 |
| Mean relative humidity (%) | 83.0 | 89.5 | 75.9 | 64.4 | 63.5 | 57.2 | 50.4 | 45.7 | | | | | 66.20 |
| Total rainfall (kg/m$^2$) | 28.7 | 28.5 | 30.9 | 36.3 | 36.3 | 32.3 | 25.1 | 7.8 | | | | | 28.24 |

Table 3: Bacterial strains, their host, nitrogen fixation (N) and phosphate-solubilising activity (P) properties [28-29]

| İsolate No | Bacterial strains (Diagnosed MIS results) | SIM | Isolated from | N | P | Siderophore |
|---|---|---|---|---|---|---|
| RK-79 | *Pantoeaagglomerans* | 0.762 | Rosaceae sp. (Malus L.) | + | + | - |
| RK-92 | *Pantoeaagglomerans* | 0.889 | Rosaceae sp. (*Pyrus*L.) | + | S+ | - |
| TV-17C | *Bacillus subtilis* | 0.677 | Rosaceae *sp. (Rubus L.)* | S+ | W+ | - |
| TV-12E | *Paenibacilluspolymyxa* | 0.551 | Poaceae sp. (*Triticum*L.) | S+ | + | - |
| TV-42A | *Pseudomonas putida* | 0.113 | Poaceae sp. (*Triticum*L.) | W+ | W+ | + |
| TV-91C | *Bacillus megaterium* | 0.474 | Poaceae sp. (*Triticum*L.) | + | W+ | - |
| TV-3D | *Bacillus megaterium* | 0.563 | Poaceae sp. (Secale L.) | S+ | + | - |
| TV-6D | *Bacillus megaterium* | 0.750 | Poaceae sp. (*Triticum*L.) | + | + | - |

(SIM: Similarity index; +: Positive; S+: Strongpositive; W+: Weak positive; -: Negative)

Table 4: Applications created in the studyand their codes

| Code of Application | Applications |
|---|---|
| Formulation A | RK-79 +RK-92 |
| Formulation B | RK-79 + RK-92 + TV-91C +TV- 17C |
| Formulation C | RK-79 + RK-92 + TV-3D + TV-12E |
| Formulation D | RK-79 + RK-92 + TV-6D + TV-42A |
| Control | Control (Uninoculated) |

of all bacterial isolates was as defined by Gunes *et al.* [30]. There was no nutrition application during the experiments. In addition, the flower buds formed in all applications were plucked before blooming [2] during the study.

Leaf samples taken from plants and bulb samples were put in paper bags and dried for 48 hours in a drying oven set at 65°C. The weight change method was applied on samples that did not dry out during this time and when the samples were fully dry, sufficient dry weights to be used for plant analyzes were measured with 0.001 gram sensitive digital scales and made ready for plant analysis. At the end of the experiment, a total of 45 samples were taken from the rhizosphere area, comprised of 3 samples from each application, to represent each application from the cultivation medium after the bulbs were harvested. Heavy metal and macro and micro nutrient amounts in the cultivation media samples were determined.

**Plant Analysis:** Nitrogen content of plant leaf and bulb samples were determined with the micro-kjheldahl method after wet decomposition with salicylic-sulfuric acid mixture [31]. The P, K, Ca, Mg, Fe, Mn, Zn, Cu, Pb, B and Cd contents of plant leaf and bulb samples were determined in 3 different steps with nitric acid-hydrogen peroxide (2: 3) acid (1st step; 75% at 145°C microwave strength for 5 minutes, 2nd step, 90% microwave strength at 180°C for 10 minutes and 3rd step at 100°C at 40% microwave strength for 10 minutes) at 40 bar pressure resistant microwave wet decomposition unit (speedwave MWS-2 Berghofproductts + (Harresstr.1. 72800 Enien Germany) after decomposition [32]. P, K, Ca, Mg, Fe, Mn, Zn, Cu B, Cd and Pb were determined from the readings in the ICP OES spectrophotometer (Perkin) Elmer, Optima 2100 DV, ICP / OES, Shelton, CT 06484-4794, USA) [32].

**Soil Analyses:** The soil reaction (pH) was measured potentiometrically with a glass electrode pH meter in a 1: 2.5 soil-water suspension [33]. Lime (% $CaCO_3$) was determined as volumetric with Scheiblercalcimeter[34]. The amount of organic matter (%) was determined with the Smith-Weldon method [35]. The determination of exchangeable cations was determined by rinsing and extracting with ammonium acetate (1 N, pH = 7.0) followed by reading with Na, K, Ca and Mg ICP-OES [36]. Phosphorus was determined by reading the light absorption of the blue colored solution formed according to the molybdophosphoric blue color method by reading in the 660 nm wavelength spectrophotometer [37]. Total nitrogen was determined by microkjheldahl method after wet decomposition with salicylic + sulfuric acid + salt

mixture [31]. Fe, Mn, Zn and Cu quantities absorbable by the plant were determined by reading ICP-OES in the percolators extracted according to DTPA method [38]. Total Pb, Cd and B heavy metals were determined according to AOAC [39].

All data have been treated by analysis of variance, which was performed using the SPSS version 17.0 statistical software package (SPSS Inc., Chicago, IL, USA). The means were separated by Duncan's multiple range tests. The maximum acceptable limit was set at 5% to be considered a significant result.

## RESULTS

### Plant Analysis

**Macro-Micro Nutrient Analysis of Tulip Bulbs:** The effects of different bacterial formulations on the amount of macro-micronutrients in bulb samples taken from different tulip varieties are given in Table 5. It was determined that the 'application' factor was significant (at $p<0.01$) while the 'cultivar' factor was insignificant in terms of effect (at $p>0.05$) on the total N (Table 5). According to the control application, the highest total amount of % N (2.53%) and % P (0.34%) was obtained from formulation C application with the Pink Impression cultivar.

When compared to the control application, the highest amount of K (%) was determined in formulation D application in the Golden Parade cultivar while, in terms of bacteria formulations, the average maximum K (%) was observed in formulation C and formulation D applications (Table 5).

It was determined that bacteria formulation applications had a statistically significant (at $p< 0.001$) effect on Ca (%) in the all cultivars in the study. According to the control, the highest amount of Ca (%) was obtained from formulation C application in the Golden Parade cultivar. The average maximum S (%) was observed in formulation A application. There were statistically insignificant ($p>0.05$) effects on Mg (%) in the bacteria formulation applications in all cultivars (Table 5).

The average maximum Na (mg $kg^{-1}$) was observed in formulation C application. The 'cultivar' factor had significant effects (at $p< 0.01$) on Na (mg $kg^{-1}$). The bacteria formulation applications had significant effects on Fe, Mn, Zn, Cu, Cd and B (mg $kg^{-1}$) nutrient elements at $p<0.001$ in the Pink Impression cultivar. There were no significant ($p>0.05$) differences in terms of bacterial application effects on Pb (mg $kg^{-1}$) in the Blue Aimable and Pink Impression when compared to the control application (Table 6).

Table 5: Macronutrient concentrations of tulip (*Tulipagesneriana* L.) bulbs (%)

| | N (%) | | | | P (%) | | | |
|---|---|---|---|---|---|---|---|---|
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 2.13 c$^{**}$ | 2.30 a$^{*}$ | 2.50 a$^{**}$ | 2.31 BC$^{***}$ | 0.33 ab $^{***}$ | 0.32 a$^{***}$ | 0.31 b$^{***}$ | 0.32 B$^{***}$ |
| Formulation B | 2.20 bc | 2.30 a | 2.30 a | 2.27 C | 0.31 bc | 0.34 a | 0.30 b | 0.32 B |
| Formulation C | 2.60 a | 2.40 a | 2.60 a | 2.53 A | 0.35 a | 0.33 a | 0.34 a | 0.34 A |
| Formulation D | 2.50 ab | 2.50 a | 2.50 a | 2.50 AB | 0.30 c | 0.34 a | 0.35 a | 0.33 AB |
| Control | 1.90 c | 1.80 b | 1.80 b | 1.83 D | 0.23 d | 0.22 b | 0.23 c | 0.23 C |
| Mean | 2.27 $^{ns}$ | 2.26 | 2.34 | 2.29 | 0.30 ns | 0.31 | 0.31 | 0.31 |
| | K (%) | Ca (%) | | | | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 1.86 a$^{**}$ | 1.86 $^{ns}$ | 1.90 c$^{***}$ | 1.87 B$^{***}$ | 0.88 a$^{***}$ | 0.86 a$^{***}$ | 0.82 c$^{***}$ | 0.85 A$^{***}$ |
| Formulation B | 1.90 a | 1.92 | 2.00 bc | 1.94 AB | 0.82 a | 0.90 a | 0.79 c | 0.84 A |
| Formulation C | 1.98 a | 1.96 | 2.10 ab | 2.01 A | 0.82 a | 0.88 a | 0.92 a | 0.87 A |
| Formulation D | 1.92 a | 1.89 | 2.20 a | 2.00 A | 0.78 a | 0.92 a | 0.88 b | 0.86 A |
| Control | 1.67 b | 1.66 | 1.60 d | 1.64 C | 0.42 b | 0.51b | 0.52 d | 0.48 B |
| Mean | 1.87 B$^{*}$ | 1.86 B | 1.96 A | 1.89 | 0.74 B$^{*}$ | 0.81 A | 0.79 AB | 0.78 |
| | S (%) | | | | Mg (%) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 0.59 ab$^{**}$ | 0.55 ab$^{***}$ | 0.62 a$^{*}$ | 0.59 A$^{***}$ | 0.45 a$^{*}$ | 0.52 $^{ns}$ | 0.44 a$^{***}$ | 0.47 $^{NS}$ |
| Formulation B | 0.55 ab | 0.59 a | 0.58 ab | 0.57 AB | 0.47 a | 0.55 | 0.47 a | 0.50 |
| Formulation C | 0.62 a | 0.58 ab | 0.55 bc | 0.58 AB | 0.47 a | 0.50 | 0.49 a | 0.49 |
| Formulation D | 0.52 b | 0.54 b | 0.58 ab | 0.55 B | 0.44 a | 1.98 | 0.47 a | 0.96 |
| Control | 0.42 c | 0.47 c | 0.50 c | 0.46 C | 0.30 b | 0.35 | 0.35 b | 0.33 |
| Mean | 0.54 $^{ns}$ | 0.55 | 0.57 | 0.55 | 0.43 $^{ns}$ | 0.78 | 0.44 | 0.55 |

ns: non-significant at $p>0.05$, * Significant at $P<0.05$, ** Significant at $p< 0.01$, *** Significant at $p< 0.001$; difference between the means shown with the same letter in a column is not significant

Table 6: Micronutrient and heavy metal concentrations of tulip (*Tulipagesneriana* L.) bulbs (mg kg$^{-1}$)

| | Na (mg kg$^{-1}$) | | | | Fe (mg kg$^{-1}$) | | | |
|---|---|---|---|---|---|---|---|---|
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 265.00 $^{ns}$ | 255.00 bc$^{***}$ | 258.00 b$^{***}$ | 259.33 AB $^{*}$ | 182.00 a$^{***}$ | 184.00 a$^{***}$ | 185.00 ab$^{***}$ | 183.67 A$^{***}$ |
| Formulation B | 258.00 | 245.00 d | 265.00 a | 256.00 B | 176.00 ab | 185.00 a | 179.00 bc | 180.00 B |
| Formulation C | 262.00 | 275.00 a | 247.00 c | 261.33 A | 178.00 ab | 180.00 ab | 176.00 c | 178.00 B |
| Formulation D | 264.00 | 254.00 c | 258.00 b | 258.67 AB | 174.00 b | 174.00 b | 187.00 a | 178.33 B |
| Control | 263.00 | 262.00 b | 249.00 c | 258.00 AB | 115.00 c | 119.00 c | 122.00 d | 118.67 C |
| Mean | 262.40 A$^{***}$ | 258.20 B | 255.40 C | 258.67 | 165.00 B$^{**}$ | 168.40 A | 169.80 A | 167.73 |
| | Mn(mg kg$^{-1}$) | | | | Zn (mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 38.00 a$^{***}$ | 39.00 ab$^{***}$ | 38.67 b$^{***}$ | 38.56 B$^{***}$ | 55.00 b$^{***}$ | 66.00 a$^{***}$ | 59.00 b$^{***}$ | 60.00B$^{***}$ |
| Formulation B | 37.33 a | 34.00 c | 39.00 b | 36.78 BC | 66.00 a | 58.67 b | 66.00 a | 63.56 A |
| Formulation C | 28.00 b | 38.00 b | 42.00 ab | 36.00 C | 47.00 b | 62.00 ab | 62.00 ab | 57.00 B |
| Formulation D | 42.00 a | 42.00 a | 44.00 a | 42.67 A | 59.67 b | 65.00 a | 64.00 a | 50.00 B |
| Control | 24.00 b | 23.00 d | 20.00 c | 22.33 D | 35.00 c | 38.00 c | 39.00 c | 37.33 C |
| Mean | 33.87 B$^{**}$ | 35.20 AB | 36.73 A | 35.27 | 50.60 B$^{***}$ | 57.73 A | 58.20 A | 55.51 |
| | Cu (mg kg$^{-1}$) | | | | Pb(mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 11.00 a$^{***}$ | 15.00 ab$^{***}$ | 15.33 a$^{*}$ | 13.78 A$^{***}$ | 1.70 $^{ns}$ | 0.50 $^{ns}$ | 0.20 b$^{***}$ | 4.13 $^{NS}$ |
| Formulation B | 11.00 a | 14.00 ab | 14.00 a | 13.00 A | 1.90 | 0.40 | 0.60 a | 0.97 |
| Formulation C | 9.00 a | 13.00 b | 16.00 a | 12.67 A | 0.30 | 0.60 | 0.50 a | 0.47 |
| Formulation D | 9.00 a | 16.00 a | 14.00 a | 13.00 A | 0.50 | 0.50 | 0.50 a | 0.50 |
| Control | 4.00 b | 8.00 c | 10.00 b | 7.33 B | 0.20 | 0.40 | 0.10 b | 0.23 |
| Mean | 8.80 B$^{***}$ | 13.20 A | 13.87 A | 11.96 | 2.92 $^{ns}$ | 0.48 | 0.38 | 1.26 |
| | B (mg kg$^{-1}$) | | | | Cd (mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 19.33c$^{***}$ | 20.67 a$^{***}$ | 26.00 $^{ns}$ | 22.00 B$^{***}$ | 2.00 c$^{***}$ | 2.00 b$^{***}$ | 2.00 b$^{*}$ | 2.00 B$^{***}$ |
| Formulation B | 22.00 bc | 26.00 a | 24.00 | 24.00 AB | 2.00 b | 3.00 a | 4.00 a | 3.00 A |
| Formulation C | 20.00 a | 24.00 a | 26.00 | 23.33 AB | 1.00 bc | 2.00 b | 3.00 ab | 2.00 B |
| Formulation D | 25.00 ab | 25.00 a | 25.00 | 25.00 A | 3.00 a | 3.00 a | 2.00 b | 2.67 A |
| Control | 14.00 c | 10.00 b | 19.00 | 14.33 C | 1.00 d | 1.00 c | 4.00 a | 2.00 B |
| Mean | 20.07 B$^{***}$ | 21.13 B | 24.00 A | 21.73 | 1.80 B$^{***}$ | 2.20 B | 3.00 A | 2.33 |

ns: non-significant at p>0.05, * Significant at P<0.05, ** Significant at p< 0.01, *** Significant at p< 0.001; difference between the means shown with the same letter in a column is not significant

**Macro-Micro Nutrient Analysis of Tulip Leaves:** Table 7 presents the results of the differences of the averages of two different tulip varieties showing the effect of the applications on the macro-micronutrient element amounts of leaf samples.

The effects of different bacterial formulation applications on the nutrient element content of leaf samples used in the experiment were found statistically significant in all nutrient elements except the Pb element. The highest average amounts of total nitrogen, Ca, S, Mg and Fe in the leaves were obtained from Formulation C application.

In comparison with the control application, the highest average maximum K (%) amount was determined in Formulation C and D applications (Table 7). The highest amounts of Na, Mn, Zn and Cd (mg kg$^{-1}$) (p <0.001) were determined in the Formulation D application, while the highest amounts of Cu and B (mg kg$^{-1}$) were determined in the control application (Table 8).

It was discovered that bacteria formulation applications had a statistically significant effect on all evaluated nutrients in the leaf samples of all cultivars except for N and Pb. According to the control application, the highest total amount of N (3.25 %), K (2.48 %), Ca (0.99%), S (0.72%), Na (327.50 mg kg$^{-1}$) and Fe (221.61 mg kg$^{-1}$) were obtained fromformulation C application in the Pink Impression cultivar. When compared to the control application, the highest amount of N, Ca, Mg (%) and Cu (mg kg$^{-1}$) was determined in the formulation C application in the Golden Parade cultivar. Furthermore, the highest K (%), Na and Mn(mg kg$^{-1}$) were obtained from the formulation D application in the Golden Parade cultivar. The highest N, P (%), Cu, Mn and Cd (mg kg$^{-1}$) amounts

Table 7: Macronutrient concentrations of tulip (*Tulipagesneriana* L.) leaves (%)

| | N (%) | | | | P (%) | | | |
|---|---|---|---|---|---|---|---|---|
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 2.13 b*** | 2.79 ab*** | 2.6 bc** | 2.51 B*** | 0.33 ns | 0.38 b*** | 0.39 ns | 0.37 A** |
| Formulation B | 2.20 b | 2.59 b | 2.62 bc | 2.45 B | 0.31 | 0.40 ab | 0.38 | 0.36 A |
| Formulation C | 3.25 a | 2.79 ab | 3.21 a | 3.08 A | 0.41 | 0.41 a | 0.42 | 0.41 A |
| Formulation D | 3.01 a | 2.96 a | 3.06 ab | 3.01 B | 0.25 | 0.42 a | 0.43 | 0.37 A |
| Control | 2.37 b | 1.98 c | 2.25 c | 2.20 C | 0.25 | 0.28 c | 0.26 | 0.26 B |
| Mean | 2.59 ns | 2.62 | 2.76 | 2.65 | 0.31 B* | 0.38 A | 0.38A | 0.35 |
| | K (%) | | | | Ca (%) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 1.86 c*** | 2.26 a** | 2.23 b*** | 2.12 B*** | 0.88 ab*** | 0.97 a*** | 0.95 a** | 0.93 B*** |
| Formulation B | 1.90 c | 2.19 a | 2.34 b | 2.12 B | 0.82 b | 1.05 a | 0.93 a | 0.93 B |
| Formulation C | 2.48 a | 2.31 a | 2.59 a | 2.46 A | 0.99 a | 1.04 a | 1.15 a | 1.06 A |
| Formulation D | 2.31 b | 2.23 a | 2.70 a | 2.41 A | 0.97 a | 1.01 a | 1.06 a | 1.01 AB |
| Control | 2.08 c | 1.83 b | 2.00 c | 1.97 C | 0.51 c | 0.60 b | 0.65 b | 0.59 C |
| Mean | 2.13 B*** | 2.16 A | 2.37 A | 2.22 | 0.83 B** | 0.93 A | 0.95 A | 0.90 |
| | S (%) | | | | Mg (%) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 0.59 ab * | 0.64 ns | 0.72 a*** | 0.65 A*** | 0.45 ns | 0.63 a*** | 0.52 c*** | 0.53 A*** |
| Formulation B | 0.55 b | 0.69 | 0.70 ab | 0.64 A | 0.47 | 0.62 a | 0.56 b | 0.55 A |
| Formulation C | 0.72 a | 0.72 | 0.61c | 0.68 A | 0.59 | 0.58 b | 0.61a | 0.59 A |
| Formulation D | 0.61 ab | 0.66 | 0.68 b | 0.65 A | 0.53 | 0.57 b | 0.58 b | 0.56 A |
| Control | 0.46 b | 0.59 | 0.58 c | 0.54 B | 0.37 | 0.39 c | 0.44 d | 0.40 B |
| Mean | 0.59 B* | 0.66 A | 0.66 A | 0.63 | 0.48 B* | 0.56 A | 0.54 A | 0.53 |

Table 8: Micronutrient and heavy metal concentrations of tulip (*Tulipagesneriana* L.) leaves (mg/kg)

| | Na (mg kg$^{-1}$) | | | | Fe (mg kg$^{-1}$) | | | |
|---|---|---|---|---|---|---|---|---|
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 265.00 c*** | 309.83 b*** | 302.51 c*** | 292.45 C*** | 182.00 c*** | 213.9 b*** | 228.48 a*** | 208.13B*** |
| Formulation B | 258.00 d | 275.63 e | 309.06 a | 277.38 D | 176.00 d | 218.76 a | 219.78 b | 202.98 C |
| Formulation C | 327.50 a | 319.69 a | 305.05 b | 317.41 A | 221.61 a | 198.45 d | 220.00 b | 213.35 A |
| Formulation D | 318.12 b | 300.36 c | 310.89 a | 309.79 B | 211.41 b | 204.02 c | 223.00 b | 212.81 A |
| Control | 327.44 a | 288.86 d | 310.01 a | 308.77 B | 129.38 e | 138.34 e | 141.83 c | 136.52 D |
| Mean | 299.21 B*** | 298.87 B | 307.39 A | 301.70 | 184.08C*** | 194.69 B | 205.68 A | 194.57 |
| | Mn(mg kg$^{-1}$) | | | | Zn (mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 38.00 b*** | 47.39 a*** | 44.56 b*** | 43.32 B*** | 55.00 b*** | 80.19 a*** | 68.59 b*** | 67.93AB*** |
| Formulation B | 37.33 b | 38.25 c | 46.34 b | 39.93 C | 66.00 a | 62.25 c | 78.55 a | 68.86AB |
| Formulation C | 35.00 bc | 44.18 b | 51.87 a | 43.68 B | 58.75 ab | 72.08 b | 68.36 b | 66.40 B |
| Formulation D | 50.61a | 49.67 a | 53.90 a | 51.39 A | 60.25 ab | 75.68 ab | 76.21 a | 70.71 A |
| Control | 29.88 c | 25.36 d | 25.00 c | 26.75 D | 42.58 c | 42.75 d | 45.34 c | 43.89 C |
| Mean | 38.16 C*** | 40.97 B | 44.19 A | 41.04 | 56.72 B*** | 67.19 A | 66.61 A | 63.50 |
| | Cu (mg kg$^{-1}$) | | | | Pb(mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 11.00 a** | 17.59 ab*** | 16.54 a** | 15.04 A*** | 0.48 ns | 0.61 b*** | 0.23 c** | 0.44 NS |
| Formulation B | 11.00 a | 16.28 b | 15.92 a | 14.21 B | 1.90 | 0.45 c | 0.73a | 1.06 |
| Formulation C | 10.46 a | 16.06 b | 18.60 a | 15.04 AB | 0.38 | 0.70 a | 0.55 b | 0.54 |
| Formulation D | 10.64 a | 19.60 a | 17.29 a | 15.84 A | 0.24 | 0.59 b | 0.59 b | 0.47 |
| Control | 4.41 b | 9.46 c | 12.25 b | 8.71 C | 0.25 | 0.45 c | 0.12 d | 0.27 |
| Mean | 9.5 0B*** | 15.80 A | 16.13 A | 13.76 | 0.65 ns | 0.56 | 0.42 | 0.55 |
| | B (mg kg$^{-1}$) | | | | Cd (mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 19.33 cd*** | 27.00 a*** | 32.50 a** | 26.28 B*** | 2.00 b*** | 2.35 c*** | 2.33 c*** | 2.23 B*** |
| Formulation B | 22.00 bc | 30.20 a | 28.50 a | 26.70 B | 2.00 b | 3.49 b | 4.74 a | 3.24 A |
| Formulation C | 24.10 b | 28.38 a | 31.33 a | 27.94 B | 1.16 bc | 2.47 c | 3.31b | 2.31 B |
| Formulation D | 31.13 a | 27.56 a | 31.13 a | 29.94 A | 3.55 a | 3.68 a | 2.35 c | 3.19 A |
| Control | 17.01d | 12.25 b | 23.09 b | 17.45 C | 1.10 c | 1.13 d | 4.65 a | 2.29 B |
| Mean | 22.71 C*** | 25.08 B | 29.37 A | 25.64 | 1.96 C*** | 2.62 B | 3.39 A | 2.64 |

Table 9: pH, $CaCO_3$, organicmatterand macronutrient concentrations of the growth soil

| | pH | | | | CaCO3 (%) | | | |
|---|---|---|---|---|---|---|---|---|
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 7.65 $^{ns}$ | 7.62 a $^{**}$ | 7.64 a$^{***}$ | 7.64 B$^{***}$ | 2.20 b$^{***}$ | 2.00 b$^{***}$ | 2.10 b$^{***}$ | 2.10B$^{***}$ |
| Formulation B | 7.64 | 7.65 a | 7.66 a | 7.65 AB | 1.80 c | 1.60 c | 1.50 c | 1.63 C |
| Formulation C | 7.50 | 7.30 b | 7.20 b | 7.33 C | 1.50d | 1.50 c | 1.60 c | 1.53 D |
| Formulation D | 7.67 | 7.65 a | 7.60 a | 7.64 B | 1.70 c | 1.60 c | 1.60 c | 1.63 C |
| Control | 7.75 | 7.75 a | 7.75 a | 7.75 A | 2.40 a | 2.40 a | 2.40 a | 2.40 A |
| Mean | 7.64 $^{ns}$ | 7.59 | 7.57 | 7.60 | 1.92 A$^{*}$ | 1.82 B | 1.84 B | 1.86 |
| | Organic matter (%) | | | | Available N (mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 1.66 $^{ns}$ | 1.62 $^{ns}$ | 1.65 $^{ns}$ | 1.64 $^{NS}$ | 14.00 c$^{***}$ | 17.00 $^{ns}$ | 19.00 b$^{***}$ | 16.67C$^{***}$ |
| Formulation B | 1.69 | 1.72 | 1.68 | 1.7 | 35.00 ab | 23.00 | 33.00 a | 30.33 A |
| Formulation C | 1.74 | 1.78 | 1.76 | 1.76 | 37.00 a | 21.67 | 35.00 a | 31.22 A |
| Formulation D | 1.77 | 1.74 | 1.68 | 1.73 | 32.00 b | 21.00 | 21.00 b | 24.67 B |
| Control | 1.14 | 1.58 | 1.58 | 1.43 | 11.00 c | 11.00 | 11.00 c | 11.00 D |
| Mean | 1.6 $^{ns}$ | 1.69 | 1.67 | 1.65 | 25.80 A$^{**}$ | 18.73B | 23.80 A | 22.78 |
| | Available P (ppm) | | | | Available K (cmol kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 12.50 c$^{***}$ | 13.70 b$^{***}$ | 12.40 c$^{***}$ | 12.87 C$^{***}$ | 2.77 $^{ns}$ | 2.74 $^{ns}$ | 2.75 $^{ns}$ | 2.75 $^{NS}$ |
| Formulation B | 16.40 b | 17.20 a | 16.80 b | 16.80 B | 2.66 | 2.75 | 2.68 | 2.70 |
| Formulation C | 17.80 b | 19.40 a | 18.70 a | 18.63 A | 2.74 | 2.75 | 2.74 | 2.74 |
| Formulation D | 20.20 a | 17.60 a | 18.60 a | 18.80 A | 2.76 | 2.74 | 2.75 | 2.75 |
| Control | 5.66 d | 5.66 c | 5.66 d | 5.66 D | 2.71 | 2.71 | 2.71 | 2.71 |
| Mean | 14.51 $^{ns}$ | 14.71 | 14.43 | 14.55 | 2.73 $^{ns}$ | 2.74 | 2.73 | 2.73 |
| | Available Ca (cmol kg$^{-1}$) | | | | SAvailable Mg (cmol kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 14.50 $^{ns}$ | 14.70 ab$^{*}$ | 15.00 a$^{*}$ | 14.73 AB$^{***}$ | 2.90 a$^{*}$ | 2.88 ab$^{***}$ | 2.91 b$^{***}$ | 2.90AB$^{***}$ |
| Formulation B | 14.20 | 14.50 ab | 14.60 a | 14.43 AB | 2.88 a | 2.89 a | 2.85 c | 2.87 B |
| Formulation C | 15.30 | 15.70 a | 14.80 a | 15.27 A | 2.94 a | 2.92 a | 2.95 a | 2.94 A |
| Formulation D | 13.50 | 13.70 bc | 13.50 ab | 13.57 B | 2.75 b | 2.84 b | 2.67 d | 2.75 C |
| Control | 12.42 | 12.42 c | 12.42 b | 12.42 C | 2.59 b | 2.59 c | 2.59 e | 2.59 D |
| Mean | 13.98 $^{ns}$ | 14.20 | 14.06 | 14.08 | 2.81 $^{ns}$ | 2.82 | 2.79 | 2.81 |

were determined in the formulation D application in the Blue Aimable cultivar in comparison to the control treatment. There were no significant ($p>0.05$) differences in terms of bacterial application effects on P (%) in the Golden Parade and Pink Impression when compared to the control application (Table 7; Table 8).

**Soil Analyses:** The effect on the macro-micro nutrient elements of the soil samples collected from the planting areas of tulip varieties is shown in Table 9. At the end of the study, different nutrient formulation applications showed statistically significant effects on all nutrient elements, pH and $CaCO_3$parameters except for organic material ratio, available K, Na, Cu and Cd elements compared to the control application.

It was determined that the 'cultivar' factor was significant (at $p<0.05$) in $CaCO_3$, available N and B. According to the averages of the general applications obtained from soil samples taken from different bacterial formulation applications, the highest available nitrogen (31.22 mg kg$^{-1}$), calcium (15.27 mg kg$^{-1}$), magnesium (2.94 mg kg$^{-1}$) and manganese (3.78 mg kg$^{-1}$) were found in the Formulation C application. Furthermore, the highest available phosphorus (18.80 mg kg-1), Fe (1.90 mg kg$^{-1}$), Zn (1.64 mg kg$^{-1}$) and Pb (0.13 mg kg$^{-1}$) were obtained from the Formulation D application (Table 9; Table 10).

The effect of bacterial formulation on pH, organic matter amount, K, Na, Ca, Cu, Mn and Cd for the Pink Impression variety was not statistically significant ($p>0.05$). The highest $CaCO_3$ in all cultivars used in the experiment was determined in the control application. The amount of available nitrogen in soil growing the Pink Impression variety was 70.27% higher while the available Mg amount increased in 11.90% with Formulation C application. An increase in 71.98% in available phosphorus content was achieved with the Formulation D application in comparison to the control group. An increase in 23.75% in the amount of determined

Table 10: Micronutrient and heavy metal concentrations of the growth soil (mg kg$^{-1}$)

| | Na(cmol kg$^{-1}$) | | | | Fe (mg kg$^{-1}$) | | | |
|---|---|---|---|---|---|---|---|---|
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 0.86 $^{ns}$ | 0.80 $^{ns}$ | 0.80 $^{ns}$ | 0.82 $^{NS}$ | 1.29 b$^{**}$ | 0.91 b$^{*}$ | 1.33 c$^{***}$ | 1.18BC$^{***}$ |
| Formulation B | 0.76 | 0.74 | 0.80 | 0.77 | 1.30 b | 1.46 ab | 1.45 b | 1.40 B |
| Formulation C | 0.74 | 0.72 | 0.75 | 0.74 | 1.88 a | 1.80 a | 1.92 a | 1.87 A |
| Formulation D | 0.72 | 0.72 | 0.70 | 0.71 | 1.88 a | 1.92 a | 1.90 a | 1.90 A |
| Control | 0.85 | 0.85 | 0.85 | 0.85 | 0.84 b | 1.20 b | 1.20 d | 1.08 C |
| Mean | 0.79 $^{ns}$ | 0.77 | 0.78 | 0.78 | 1.44 $^{ns}$ | 1.46 | 1.56 | 1.49 |
| | Cu (mg kg$^{-1}$) | | | | Mn (mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 1.33 $^{ns}$ | 1.88 c$^{***}$ | 1.88 a$^{***}$ | 1.70 $^{NS}$ | 3.67 $^{ns}$ | 1.88 c$^{***}$ | 3.65 c$^{***}$ | 3.07 B$^{*}$ |
| Formulation B | 1.90 | 1.90 b | 1.88 a | 1.89 | 3.44 | 3.69 a | 3.75 b | 2.88 AB |
| Formulation C | 1.90 | 1.93 a | 1.92 a | 1.92 | 3.79 | 3.78 a | 3.76 a | 3.78 A |
| Formulation D | 1.90 | 1.92a | 1.94 a | 1.92 | 3.77 | 3.78 a | 3.75 a | 3.77 A |
| Control | 1.75 | 1.75 d | 1.75 b | 1.75 | 3.66 | 3.66 b | 3.66 c | 3.66 A |
| Mean | 1.76 $^{ns}$ | 1.87 | 1.87 | 1.83 | 3.55 $^{ns}$ | 3.37 | 3.7 | 3.54 |
| | Zn(mg kg$^{-1}$) | | | | B (mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 1.36 bc$^{**}$ | 1.38 d$^{***}$ | 1.42 c$^{***}$ | 1.39 D$^{***}$ | 0.35 c$^{***}$ | 0.38 a$^{***}$ | 0.35 b$^{**}$ | 0.36 B$^{***}$ |
| Formulation B | 1.44 ab | 1.45 c | 1.44 c | 1.44 C | 0.39 a | 0.39 a | 0.35 b | 0.38 A |
| Formulation C | 1.56 a | 1.55 b | 1.56 b | 1.56 B | 0.37 b | 0.39 a | 0.38 a | 0.38 A |
| Formulation D | 1.60 a | 1.65 a | 1.68 a | 1.64 A | 0.36bc | 0.38 a | 0.38 a | 0.37 A |
| Control | 1.22 c | 1.22 e | 1.22 d | 1.22 E | 0.33 d | 0.33 b | 0.33 b | 0.33 C |
| Mean | 1.44 $^{ns}$ | 1.45 | 1.46 | 1.45 | 0.36 B$^{***}$ | 0.37 A | 0.36 B | 0.36 |
| | Pb(mg kg$^{-1}$) | | | | Cd(mg kg$^{-1}$) | | | |
| Applications | Pink Impression | Blue Aimable | Golden Parade | Mean | Pink Impression | Blue Aimable | Golden Parade | Mean |
| Formulation A | 0.10 b$^{*}$ | 0.10 $^{ns}$ | 0.10 c$^{***}$ | 0.10 D$^{***}$ | 0.14 $^{ns}$ | 0.14 $^{ns}$ | 0.12 $^{ns}$ | 0.13 $^{NS}$ |
| Formulation B | 0.10 b | 0.11 | 0.11 b | 0.11 C | 0.47 | 0.14 | 0.12 | 0.24 |
| Formulation C | 0.12 a | 0.12 | 0.10 c | 0.11 B | 0.13 | 0.14 | 0.12 | 0.13 |
| Formulation D | 0.12 a | 0.12 | 0.14 a | 0.13 A | 0.13 | 0.13 | 0.13 | 0.13 |
| Control | 0.09 b | 0.90 | 0.90 d | 0.90 E | 0.14 | 0.14 | 0.14 | 0.14 |
| Mean | 0.11 $^{ns}$ | 0.11 | 0.11 | 0.11 | 0.20 $^{ns}$ | 0.14 | 0.13 | 0.16 |

Zn was obtained with formulation D, while an increase of 21.79% was noted with the formulation C application. Formulation C and D applications were in the same statistical group in terms of available Zn amount (Table 9; Table 10).

The amount of pH in the soil samples of Blue Aimable tulip variety ($p < 0.01$) and the pH, $CaCO_3$, available N, P, Mg, Fe, Cu, Mn, Zn and Pb amounts in the soil samples of the Golden Parade tulip variety ($p < 0.001$) were statistically significant compared to the control application (Table 9; Table 10). Formulation C application generated an increase in 70.82% in the amount of available phosphorus determined in the soil of cultivated Blue Aimable variety compared to the control application. Formulation B and D applications were in the same statistical group in terms of available phosphorus amount. An increase in 20.89% in the amount of determined available Ca was generated with the application of Formulation C. The highest amounts of magnesium were determined in Formulation B and C applications compared to the control application (Table 9). The highest Cu and Zn levels determined in the soil of the related varieties were found in Formulation D application (Table 10).

An increase has been achieved in the available nitrogen amounts with Formulation C (68.57%) and Formulation B applications (66.67%) in the soil samples of the Golden Parade tulip cultivar compared to the control application. Formulation C (69.73 %) and Formulation D (69.57 %) applications enabled an increase in available phosphorus compared to the contol application. While the available Ca was 12.42 mg kg$^{-1}$ in the control application, the amounts determined in Formulation A, B and C were 15 mg kg$^{-1}$, 14.60 mg kg$^{-1}$ and 14.80 mg kg$^{-1}$ respectively and all three applications

were in the same statistical group (Table 9). While an increase in 12.20% was determined in the available Mg amount compared to the control application (Table 9), an increase of 27.38% was determined in the available Zn amount with the Formulation D application. The highest Fe, Mn and B amounts were obtained from Formulation C and D applications (Table 10).

## DISCUSSION AND CONCLUSION

It is well known that tulip plants are grown very rapidly in spring using nutrients (e.g. carbohydrates) preserved in the main plant's bulb scales [40, 41]. Tulip plants store a large amount of carbohydrates in bulb scales to support root and shoot growth and absorb nutrients in sub-soil life [42, 43]. Nutrient content is influenced by a large number of variables such as genetic structure of plant material, physical and chemical properties of the growing media, light and fertilization and irrigation programs [44]. The current study results showed that nutrient quantities differed according to tulip varieties and applications and were in parallel with the findings of Mickelbart [44].

The amount of optimum nitrogen for normal plant growth ranges from 2% to 5% of the dry weight of the plant [45]. Nitrogen is an element of basic cellular components such as amino acids, proteins and nucleic acids. It also intensifies the green leaf color. At the same time is controls P, K and other nutrients and increases in the efficiency of many products[46]. It promotes photosynthesis as it increases in the amount of chlorophyll [46-48]. The role of N as an osmotic agent that provides water retention in a vacuum is considered important for nutritional function [49]. As for all plants in general, nitrogen is a very important nutrient in tulip cultivation [50]. Nitrogen content is also important for the planting period of tulip bulbs. In addition, Baba *et al*. [51] reported that the N concentration in the bulb should be higher than 1.2% for the application of forcing in tulip cultivation. At the end of the study, the least total nitrogen analyzed in tulip bulbs, leaves and soil was found with the control application as 1.83%, 2.20% and 11.00 mg $kg^{-1}$, respectively and the highest amount of nitrogen was found with Formulation C as 2.60%, 3.08%, 31.22 mg $kg^{-1}$, respectively.Castaño *et al*., [45] stated that keeping the control application below the limits of the amount of nitrogen indicated that the addition of nutrients in tulip cultivation is important. Ohyama *et al*. [42] reported that although N was stored in the scales of bulbs to be used in tulip planting, this content was insufficient to counter the needs of the bulblets during peak growth period to ensure the highest yield. Thus, it has been concluded that it would be beneficial to inject PGPRs especially formulated with *Pantoea agglomerans*-RK-79+*Pantoea agglomerans*-RK-92+*Bacillus megaterium*-TV-3D +*Paenibacilluspolymyxa*- TV- 12Estrains to the growing medium in the cultivation of high quality bulbs for planting and also to promote blooming of cut flowers [51].

The amount of P obtained from tulip leaves was less than that obtained from bulbs. The highest amount of P according to the average of general applications was obtained with formulation C and the amounts of P from bulbs and leaves ranged between 0.23% and 0.41%. The highest amount of P determined in soil was 18.80 mg $kg^{-1}$ with Formulation D application. A low weight in main bulbs of tulips has been reported as an indication of deficiency in P [52, 53]. The amount of P in bacterial formulation was increased in C and D applications compared to the control application. These applications can be recommended to obtain larger bulbs. Increasing in the macro and micro element content of the bulbs, leaves and soil with the bacterial formulations used and manifesting a higher effect with some formulation C and D applications compared to the samples of the control application fed only with the nutrients present in the soil and bulb reserves without supplementary nutrients during the cultivation of these formulations (including *Pantoea agglomerans*-RK-79*,* *Pantoea agglomerans*-RK-92,*Paenibacilluspolymyxa*- TV- 12E,*Bacillus megaterium*-TV-3D, *Bacillus megaterium*-TV-6D and *Pseudomonas putida*-TV-42A) has been attributed to the use of more carbon sources, high N fixation and P-dissolving properties [54, 55].

van der Boon [56] reported that the tulip bulb had little or no response to K fertilizers. The amount of K obtained in bulbs in this study ranged from 1.97% to 2.46%. The amounts obtained from leaf samples ranged between 2.71% to 2.75%. When the differences between the K amounts analyzed from leaves and soil are compared, the K differences determined in bulbs were more pronounced, albeit slightly according to the applications. In addition, it was found that the application of potassium-dissolving microorganisms [57] provided an increase in the K content of the growth media which is in parallel with the results of this study.

Mengel and Kirkby [58] and Pérez-Pérez *et al*. [59] reported that, in addition to activating various enzyme systems, Ca is a macronutrient with important biochemical functions that promote many metabolic processes and therefore contribute to the proper development of plants. Among all the organs, leaves contain the highest

concentration [60]. The amounts of Ca determined separately in bulbs and leaves had changed according to the organs and the highest amounts were obtained with the Formation C application in our study. The Ca amounts in bulb samples had changed between 0.48% and 0.87% while the amount of Ca in leaf samples had changed between 0.59% and 1.06%. The amount of Ca increased in bacterial formulation C administration compared to the control application. Large amounts of Ca accumulate in plant cell walls and membranes [50]. This can account for the longer durability of bulbs obtained in the Formulation C application.

In fact, the indirect effects of PGPR bacterial strains on properties such as induced systemic resistance, biocontrol of plant pathogens [14-16] can be explained with this result. Furthermore, the collapse of flower stems can caused by a Ca deficiency in tulip bulbs [61] is also expected to be reduced with the use of this bacterial formulation.

While the difference in the amount of Mg determined in bulb samples in this study was not deemed significant, the Mg amount of leaf samples changed between 0.40% and 0.59%. The highest Mg content of leaf and soil samples was determined in Formulation C application. Previous studies have reported increased in tulip yield when fertilized with Mg salts [62].

As a result of the study, the increase in the amount of Mg in the leaf and soil samples is also considered to have a positive effect on tulip yield. Orhan *et al*. [63] also reported that bacterial applications significantly effect on available Mg in soil. In this study, the amount of available Mg has been increased in the application of bacterial formulation and it can be argued that this increase is due to the involvement of PGPRs such as growth hormone auxin having a role in the synthesis as well as other mechanism properties. The following literature on this finding by Pal *et al*. [64]; Sahin *et al*. [26]; Altın and Tayyar [65] and Çakmakçı *et al*. [27] can be presented as an example.

Research studies have manifested that elements such as sodium (Na), cobalt (Co) and silicon (Si) which are not necessary for the growth and development of a plant are commonly found in certain ratios in the structures of most plants [66, 67]. The analysis of soil samples of this study yielded available sodium however, no toxic indications were observed.

Zn in soils is easily soluble due to its easy adsorption by mineral and organic materials compared to other metals. Zinc functions as a structural and catalytic component of proteins, enzymes and co-factors for the normal development of pigment biosynthesis in the increase in chlorophyll content [68]. Zinc plays an important role in phosphorus and calcium intake and also in the availability of nitrogen [69]. As a result of this study, the highest zinc content was determined in leaf and soil samples with the Formulation D application. Considering the findings of Balashouri [68] and the current study suggests that there is an association between achieving the highest P content with Formulation C and D applications and determining the highest Zn in the same applications.

The amount of Cd determined in soil samples of bacterial formulation applications, the amount of B and Pb determined in bulb samples and the amount of Pb in leaf samples were statistically insignificant when compared to the control application.

In conclusion, the application of bacterial formulation in tulip varieties has increased in the total amount of N, P, K, Ca, S, Mg, Fe, Mn, Zn and Cu macro and micro elements in bulb and leaf samples compared to the control application and this increase is considered statistically significant. It has been manifested that the N, P, K, Ca, S, Mg, Fe, Mn, Zn and Cu content of tulip bulbs and leaves can be enhanced, especially using the formulation comprised of *Pantoea agglomerans*-RK-79+*Pantoea agglomerans*-RK-92+ *Bacillus megaterium*-TV-3D+ *Paenibacilluspolymyxa*- TV- 12E(Formulation C) and *Pantoea agglomerans*-RK-79+*Pantoea agglomerans*-RK-92+ *Bacillus megaterium*-TV-6D + *Pseudomonas putida*-TV-42A strains (Formulation D).Thus, these results can be benefited from both obtaining high quality and durable bulbs with the necessary N content required for planting as well as promote blooming in cut flowers in the cultivation of cut flowers. Furthermore, it has been concluded that this information can be used to develop more efficient and environmentally friendly fertilizer management plans for commercial bulb production and landscape use.

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