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Research Article

# The Effect of Some Endophytic Bacteria on Seedling Growth and Physiological Properties of *Salvia officinalis* L.

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**Abstract:** In order to meet the demand for medicinal sage (*Salvia officinalis* L.), which is an important economic product, harvesting from nature has economic value. However, it may not always be of the desired standard and quality. Also, the harvesting from nature endangers their natural population causing their genetic base to decline. For this reason, it is important to produce it in an agrosystem and to increase yield in a sustainable way. In this study, the effects of eleven endophyte bacteria (EB) isolates applications on the development, morphology, and physicochemical properties of *Salvia officinalis* L. were investigated by climate chamber experiments. Peat+perlite+soil (1:1:2) mixture was used as the growing medium and EB was applied two times by soaking method. Effects of EB applications on shoot/root length, root/stem fresh and dry weight, Dualex values (Nitrogen balance index (NBI), flavonol, anthocyanin, and chlorophyll), leaf area, leaf temperature and color values (L*, a) *, b * C and Hue° were examined. All EB applications increased the plant height and leaf area. Also, the majority of EB isolates enhanced the root dry weight. The effect of EB applications on flavonol and chlorophyll content was not found statistically significant. However, there was a statistically significant increase in the nitrogen balance index (NBI). It was also observed that EB applications caused changes in plant color. According to the results obtained, it has been seen that it is possible to produce environmentally friendly and sustainable medicinal sage with appropriate plant-bacteria combinations.

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## 1. Introduction

Türkiye, which is the gene center of many plants cultivated in world agriculture, is home to a total of 92 *Salvia sp.* (Lamiaceae), 47 of which are endemic (Altındal ve Akgun, 2015). The essential oil and leaves of sage, are widely used in alternative or traditional medicine as an additive, herbal medicine, food, cosmetics, and perfumery (Elmas, 2021). In 2020, approximately 2 176 tons of sage, which is one of the important export products, was exported and 8 155 503 dollars were brought to the economy (Elmas, 2021). The trade of sage is carried out by cultivation in different provinces and collecting from nature. It is difficult to obtain products of desired standards and quality by collecting from nature. In addition, unconscious and over-collecting sage populations lead to unsustainable extinction, overexploitation, and the narrowing of their genetic base. For this reason, in order to ensure its sustainable use, it is necessary to expand the production of sage, which is in the standards and quantities required by the world markets. Studies have shown that yield and quality in sage production differ according to the climate and soil conditions of the region where it is grown, harvest periods, drying conditions, and agricultural practices (Elmas, 2021). Although collecting from nature is economical, it makes it difficult to obtain the product of the desired standard and quality. For this reason, in order to meet the demands of industrialists and consumers, sage production has been started in different provinces of our country in recent years in the quality required by the world markets. (Bayraktar et al., 2017).

The excessive and unconscious use of chemical fertilizers and pesticides negatively affects the environment and human health. It is important to avoid the use of chemical fertilizers and pesticides in the cultivation of medicinal and aromatic plants such as sage. (Egamberdieva and Teixeira da Silva, 2015). Nowadays, symbiotic or free-living microorganisms, which are one of the environmentally friendly sustainable agriculture approaches, have an important potential to increase the development of plants, their nutrient uptake, and their tolerance to biotic and abiotic stress conditions (Altunlu et al., 2019).

Beneficial bacteria to plants are named in general as "Plant growth promoting rhizobacteria/bacteria" (PGPR-PGPB). PGPBs live or colonize the plant as endophytes (inside the plant) and/or epiphytes (on the plant surface). Endophyte bacteria (EB) are defined as microorganisms that can spread throughout the plant without harming the plant and spend at least a part of their life in the internal tissues of the plant (Hallmann 1997; Hardoim et al., 2008; Hardoim, 2011). EB can affect the growth and development of plants directly and indirectly (Imriz et al., 2014; Ucar and Akkopru, 2022). EB contributes directly to the plants in ways such as fixing nitrogen, dissolving phosphorus, supporting the uptake of iron and other nutrients, produce phytohormones (Grobelak et al., 2015). They can make an indirect contribution by providing protection against harmful organisms by activating antibiosis, competition, hyperparasitism, and activated plant-induced resistance or tolerance (Glick, 2014). Endophytic bacteria have some advantages over epiphytic bacteria. Since they live in the internal tissues of plants, the metabolites they produce can be taken directly by plants (Romano et al., 2020; Akkopru et al., 2021). EB can reach and affect all tissues in the plant thanks to the transmission systems of plants (Hardoim et al., 2008; Hardoim, 2011; Mercado-Blanco and Lugtenberg 2014; Romano et al., 2020).

This study aimed to investigate the effects of endophytic bacteria on seedling growth and some physiological parameters of medicinal sage (*Salvia officinalis* L.), an important medicinal and aromatic plant with high market value.

## 2. Material and Methods

### 2.1. Plant material and running of the experiment

The seeds of the *S. officinalis* were obtained from Van Yüzüncü Yıl University Medicinal and Aromatic Plants Garden. The study was carried out in the Van YYU Faculty of Agriculture, Field Crops Department, Climate room, Physiology laboratory, and Plant Protection Department laboratory. The seeds were sown in a growth medium consisting of peat + perlite + soil (1:1:2) 500 cc pots. After sowing, the plants were grown in a dark/light photoperiod of 8/16 hours, in a controlled climate chamber at 25°C and 65% humidity. The experiment was carried out in seven replications according to the Randomized Complete Plots Experimental Design. The hoagland nutrient solution was applied to supply the nutritional needs of the seedlings after the cotyledon leaves emerged (Hoagland ve Arnond 1950).

### 2.2. Endophyte bacteria and their application

Endophyte bacteria isolates used in the study, which were previously isolated from plants belonging to the Poaceae family in the Van Lake basin, were obtained from the bacteriology laboratory stocks of Van Yüzüncü Yıl University, Faculty of Agriculture, Department of Plant Protection. The isolation and detection of some characteristics of the EBs were carried out within the scope of the project supported by the Scientific Research Projects Department of Van Yüzüncü Yıl University (Grant no: FBA-2020-8551) (Table 1). For this purpose, EB suspensions with a density of $10^8$ CFU/mL were prepared from 48-hour cultures grown in King B medium. The EB suspension was applied to 15 mL/plant by drenching method (Ucar and Akkopru, 2022).

Table 1. Data on the endophytic bacteria (EB) isolates used and some of their characteristics that may contribute to plant health and growth

| | EB Isolate Code | Gram React.* | IAA | ACCd | Nitrogen fix. | " P " solubilization (EI) | Sid. (EI) |
|---|---|---|---|---|---|---|---|
| 1 | G58S1 | - | 0.938 | + | - | 1.69 | 1.47 |
| 2 | G129K1-1 | - | 1.419 | - | + | 2.12 | 1.75 |
| 3 | G113Y1 | + | 0.024 | - | - | 1.60 | 1.51 |
| 4 | G43K2 | - | 0.430 | - | + | 1.89 | 1.28 |
| 5 | G106Y1 | + | 0.941 | - | - | 1.57 | - |
| 6 | G91S2 | + | 0.554 | - | + | 6.75 | 1.33 |
| 7 | G21Y1 | - | 1.472 | + | - | 1.12 | 2.04 |
| 8 | G100Y2 | - | 0.740 | + | - | 3.94 | 1.33 |
| 9 | G118K1T | - | 0.045 | - | - | 6.91 | 2.30 |
| 10 | G59S2 | - | 0.655 | - | + | 8.13 | 2.42 |
| 11 | G104Y1 | - | 1.108 | + | - | 2.61 | 1.89 |

*Gram React.: Gram reaction according to KOH test, IAA: indole acetic acid production, ACCd: 1-aminocyclopropane-1-carboxylate deaminaze Nitrogen fix.: Nitrogen fixation ability, P solubilization (EI): ability to dissolve phosphorus in vitro (enzyme index), Sid. (EI): Siderophore production ability (enzyme Index).

### 2.3. Determination of plant growth parameters

The study was terminated after 8 weeks. The plant was cut from the root collar and, the roots and shoots were washed separately. Roots were placed between blotting papers for the washing water to come out. Root and stem lengths were measured by the ruler as centimeters. Fresh and dry weights of root and stem were determined with the help of precision scales. The shoots and roots were then dried at 60°C for 72 hours and their dry weights were weighed. Leaf surface temperatures (Spectrum Technologies Inc.) were measured with the help of a portable infrared thermometer using the "Easy Leaf Area" program (Tuncturk et al., 2020). Nitrogen balance index (NBI), chlorophyll, flavonol, and anthocyanin content in leaves were measured in real-time and non-destructively on the leaf with Dualex scientific+ (FORCE-A, France). Color values were determined by (Minolta CR-400) brand colorimeter as L*, a*, b* C, and Hue° angle values. L* lightness ((L*=0 black and L*=100 white), a* red/green (+a* red, – a* green), b* yellow/blue (+b* yellow, -b* blue), Chroma is the brightness or opacity, Hue is the perceived color and the values that determine the name of the color (Anonim, 2022).

### 2.4. Analysis of Data

Statistical analyzes of the obtained data were made using the COSTAT (version 6.03) package program and multiple comparison tests were performed according to the Duncan test.

## 3. Results

The effects of EB isolates on morphological and physiological development and color parameters (L*, a*, b*, Chroma, and Hue) in medicinal sage were investigated. The obtained values are given in Tables 2, 3, and 4. The effects of endophyte bacteria were found to be statistically significant on all parameters except shoot dry weight. All EB treatments provided a significant increase in plant height compared to the control group. While the most successful application was obtained from the G43K2 isolate with 27.64 cm, it was observed that it was in the same statistical group with G129K1, G106Y1, and G59S2 isolates. In addition, it was determined that the difference between the applications was statistically significant at the level of 1% on the fresh weight of the shoots. Among the bacterial applications, the highest shoot fresh weight was obtained using the G104Y1 (5.11 g), while six bacterial isolates were found to be in the same statistic group. The lowest value was obtained with G58S1 isolate

(3.37 g). The control group, G129K1, G43K2, and G118K1T isolates are in the same statistical group. There was no statistically significant effect of experimental factors on shoot dry weight, which varies between 0.58-1.05 g observed (Table 2).

The effects of bacterial applications on means of the root length were statistically significant at the level of 5% level. While the highest plant root length was measured with G113Y1 (30.42 cm) application group, it was included in the same statistical group with seven different bacterial applications. The lowest root length was obtained from the G129K1 (22.14 cm) application, and also it is in the same statistical group as the control and other three isolates (Table 2).

The effect of EB on means of plant root fresh and dry weight was statistically significant at the 1% level. While the highest root fresh weight of 1.55 g was obtained from the application of G91S2 isolate, the lowest value was obtained with the use of G129K1 and G43K2 isolates with 0.76 g and 0.94 g, respectively, and they are in the same statistical group. While the lowest value in terms of root dry weight was determined from the control group (0.14 g), it was statistically found in the same group as G59S2, G129K1, G129K1, and G104Y1 isolates. The highest root dry weight was observed with 0.27 g from the G91S2 isolate (Table 2).

Table 2. Effects of EB treatments on the morphological parameters of *S. officinalis*

| Applications | Plant Shoot height (cm) | Shoot weight (g) | | Plant Root length (cm) | Root weight (g) | |
|---|---|---|---|---|---|---|
| | | Fresh | Dry | | Fresh | Dry |
| Control | 14.71$^{f}$ | 3.77$^{de}$ | 0.88 | 23.57$^{cd}$ | 0.99$^{cd}$ | 0.14$^{d}$ |
| G58S1 | 19.35$^{e}$ | 3.37$^{e}$ | 0.85 | 24.85$^{bcd}$ | 1.00$^{cd}$ | 0.20$^{bc}$ |
| G129K1 | 24.35$^{abc}$ | 4.01$^{cde}$ | 0.87 | 22.14$^{d}$ | 0.76$^{e}$ | 0.15$^{d}$ |
| G113Y1 | 22.61$^{cde}$ | 4.25$^{a-d}$ | 0.91 | 30.42$^{a}$ | 1.17$^{bc}$ | 0.22$^{b}$ |
| G43K2 | 27.64$^{a}$ | 4.07$^{b-e}$ | 0.92 | 24.42$^{abcd}$ | 0.94$^{de}$ | 0.20$^{bc}$ |
| G106Y1 | 25.57$^{abc}$ | 4.92$^{ab}$ | 0.98 | 28.14$^{abc}$ | 1.20$^{bc}$ | 0.22$^{b}$ |
| G91S2 | 19.64$^{e}$ | 4.83$^{abc}$ | 1.05 | 29.00$^{ab}$ | 1.55$^{a}$ | 0.27$^{a}$ |
| G21Y1 | 22.57$^{cde}$ | 4.77$^{abc}$ | 1.03 | 29.78$^{ab}$ | 1.00$^{cd}$ | 0.20$^{bc}$ |
| G100Y2 | 19.35$^{e}$ | 4.56$^{a-d}$ | 0.89 | 28.64$^{abc}$ | 1.26$^{b}$ | 0.19$^{bc}$ |
| G118K1T | 20.57$^{de}$ | 4.15$^{b-e}$ | 0.90 | 28.33$^{abc}$ | 1.16$^{bc}$ | 0.19$^{bc}$ |
| G59S2 | 26.42$^{ab}$ | 4.61$^{a-d}$ | 0.85 | 23.71$^{cd}$ | 1.00$^{cd}$ | 0.14$^{d}$ |
| G104Y1 | 23.68$^{bcd}$ | 5.11$^{a}$ | 1.05 | 28.02$^{abc}$ | 1.29$^{b}$ | 0.17$^{cd}$ |
| Average | 22.20 | 4.37 | 0.93 | 17.46 | 1.11 | 0.19 |
| F value | 7.909** | 2.794** | 1,001ns | 2.200* | 7.464** | 8.70** |
| LSD | 3.66 | 0.87 | 0.20 | 5.14 | 0.21 | 0.03 |
| CV (%) | 15.50 | 18.73 | 20.90 | 18.09 | 17.93 | 17.78 |

**P < 0.01 level; * P < 0.05 level significance: ns: no significance.

The effect of bacterial isolates on NBI content was significant at the 1% level. While the lowest NBI value was obtained from the control group as 26.32 dualex index, the highest value was determined as 45.98 dualex index with the G129K1 isolate. It was also determined that G129K1 was in the same statistical group with G100Y2 and G59S2 isolates (Table 3).

The effects of bacterial applications on chlorophyll and flavonol content were statistically insignificant (Table 3). In the study, chlorophyll content was determined between 21.32-25.44 dx and flavonol content between 0.54-0.71 dx. On the other hand, anthocyanin content was found to be significant at the 1% level. While the highest anthocyanin content was detected in the control group (0.074 dx), it was in the same group with five different bacterial isolates. The lowest anthocyanin content (0.050 dx) was determined from the G21Y1 application, but there was no statistically significant difference between them and the three bacterial isolates (G58S1, G113Y1, G100Y2).

The effect of the treatments on leaf area was significant at the 1% level. The highest leaf area with 14.79 cm2 was obtained from the G21Y1 treatment, while it was found to be in the same statistical group with seven different bacterial isolates. The lowest leaf area was determined as 8.82 $cm^2$ in the control group and the G118K1T (10.92 cm2) isolate (Table 3).

The effect of EB on plant temperature was found to be significant at the 1% level. The highest plant temperature was obtained as 27.31°C, 27.30°C, 27.27°C and 26.68°C from G129K1, control, G58S1, and G106Y1 treatments, which are in the same statistical group, respectively. The lowest plant temperature (24.25°C) was determined from G100Y2. However, it is in the same statistical group as G43K2 (25.58 °C), G113Y1 (25.45 °C), and G59S2 (25.42 °C) isolates (Table 3).

Table 3. Effect of endophytic bacterial applications on some biochemical parameters of *Salvia officinalis*

| Applications | NBI (Dualex index) | Chlorophyll (Dualex index) | Flavonol (Dualex index) | Anthocyanin (Dualex index) | Leaf area ($cm^2$) | Plant temperature (°C)§ |
|---|---|---|---|---|---|---|
| Control | 26.32f | 21.32 | 0.71 | 0.074a | 8.82d | 27.30a |
| G58S1 | 39.47bcd | 25.35 | 0.64 | 0.058cde | 12.17bc | 27.27a |
| G129K1 | 45.98a | 22.10 | 0.54 | 0.065a-d | 14.54ab | 27.31a |
| G113Y1 | 34.50e | 24.64 | 0.70 | 0.055de | 14.20ab | 25.45d |
| G43K2 | 35.94de | 22.91 | 0.65 | 0.061bcd | 12.86abc | 25.58d |
| G106Y1 | 41.15bc | 23.48 | 0.57 | 0.061bcd | 14.28ab | 26.68ab |
| G91S2 | 36.62de | 23.50 | 0.65 | 0.071ab | 13.73ab | 26.15bcd |
| G21Y1 | 41.09bc | 25.44 | 0.66 | 0.050e | 14.79a | 26.41bc |
| G100Y2 | 43.78ab | 24.11 | 0.57 | 0.057cde | 14.58ab | 24.25e |
| G118K1T | 37.29cde | 23.41 | 0.60 | 0.072a | 10.92cd | 25.66cd |
| G59S2 | 41.56abc | 23.50 | 0.59 | 0.067abc | 12.57abc | 25.42d |
| G104Y1 | 39.23cd | 24.65 | 0.63 | 0.065a-d | 12.34bc | 25.68bcd |
| Average | 38.58 | 23.70 | 0.63 | 0.063 | 12.98 | 26.10 |
| F value | 10.360** | 0.690ns | 1.384ns | 3.825** | 4.295** | 10.643** |
| LSD | 4.45 | 4.15 | 0.12 | 0.01 | 2.41 | 0.80 |
| CV (%) | 10.82 | 16.46 | 18.60 | 15.82 | 17.47 | 2.90 |

**P < 0.01 level; * P < 0.05 level significance: ns: no significance.

It was noted that the difference of color measurements between of EB tretment groups was statistically insignificant. The lowest and highest values were determined as L*; 41.94 - 47.49, a*; (-14.18)-(-12.03), b*; 18.85-24.63, Chroma; 22.50-28.59 and Hue; 121.06-122.97 (Table 4).

Table 4. Effect of EB applications on color values of *Salvia officinalis*

| Uygulamalar | L* | a* | b* | Chroma | Hue |
|---|---|---|---|---|---|
| Control | 41.94b§ | -12.03a | 18.85b | 22.50b | 122.97 |
| G58S1 | 44.14ab | -13.58ab | 21.68ab | 25.59ab | 122.37 |
| G129K1 | 44.00ab | -14.18b | 22.20ab | 26.35ab | 122.86 |
| G113Y1 | 46.08ab | -13.19ab | 21.17ab | 24.97ab | 122.28 |
| G43K2 | 45.98ab | -13.35ab | 21.68ab | 25.47ab | 121.91 |
| G106Y1 | 43.80ab | -13.56ab | 20.97ab | 24.98ab | 122.94 |
| G91S2 | 46.42a | -13.49ab | 21.66ab | 25.54ab | 122.42 |
| G21Y1 | 46.15a | -14.83b | 23.25a | 27.59a | 122.81 |
| G100Y2 | 46.33a | -14.38b | 23.05ab | 27.20a | 122.46 |
| G118K1T | 47.49a | -14.45b | 24.63a | 28.59a | 121.06 |
| G59S2 | 45.76ab | -14.82b | 23.47a | 27.77a | 122.42 |
| G104Y1 | 45.33ab | -13.43ab | 21.32ab | 25.22ab | 122.42 |
| Average | 45.29 | -13.77 | 21.99 | 25.98 | 122.40 |
| F value | 1.147ns | 1.963ns | 1.014ns | 1.134ns | 0.443ns |
| LSD | 4.16 | 1.66 | 4.30 | 4.49 | 2.31 |
| CV (%) | 5.45 | 7.17 | 11.60 | 10.25 | 1.12 |

**P < 0.01 level; * P < 0.05 level significance: ns: no significance.

## 4. Discussion

Secondary metabolites of medicinal plants such as *S. officinalis* are the main source of bioactive products and are considered as important pharmaceuticals. Medicinal sage is grown commercially in many countries around the world since it is difficult to obtain products of the desired standard and quality with pick up from nature (Phillipson, 2001; Bayraktar et al., 2017). In agricultural production, many chemical inputs are used in order to increase yield and protect plants from diseases and pests

(Egamberdieva and Teixeira da Silva, 2015). In our study, the effects of 11 EB isolates (Table 1) on some morphological, physiological, and color properties of medicinal sage were investigated.

It was determined that the obtained data were compatible with the relevant literature. Many studies have shown that growth-promoting bacteria (EB or PGPR) can increase plant growth. PGPRs can contribute to plant growth by increasing the availability of mineral phosphate and other nutrients (Hayat et al., 2010; Akkopru and Ozaktan, 2018), by biological fixation of nitrogen (Ardakani et al., 2010); by the production of IAA (Mishra et al., 2010; Etesami et al., 2015). In addition, they contribute to plant growth by reducing the amount of ethylene in the roots of plants under stress through the production of ACC deaminase (Glick, 2014). It is known that some of the EB isolates we used in our study have different properties such as nitrogen fixation, IAA, ACC deaminase, phosphate solubilization, and siderophore production (Table 1).

It was determined that each of the EB isolates had different; positive, negative, or neutral effects on the plants in terms of the investigated parameters. It was observed that all EB applications significantly increased plant height and leaf area (except G118K1T). Rahimi et al., (2013) reported that bacteria with nitrogen fixation ability increase the height of the basil plant (*Ocimum bacilicum* L.). Similarly, Anbi et al., (2020) reported that bacteria of different characteristics increase plant height and leaf area at different levels. It was shown that nitrogen and phosphorus have an important role in increasing the number and surface area of leaves by affecting the cell division process and chlorophyll production as a growth factor (Larimi et al., 2014). Zhang et al., (2019) revealed that there is a positive correlation between root and stem length, secondary root number, and leaf area index in plants inoculated with bacteria. It was indicated that PGPR (*Bacillus pumilus* TV-67C, *Bacillus subtilis* TV-13B, and *Bacillus megaterium* TV6D + *Brevibacillus choshiensis* TV-53D + *Pantoea agglomerans* RK-92+) application decreased damage caused by stress at the cabbage plant (Samancıoglu et al., 2016). Tuncturk et al., (2021), in their study on soybean, showed that rhizobacteria applications increased the leaf area index compared to the control. It was shown that the results of the researchers and our findings are similar.

Ghorbanpour and Hatami (2014) and Ghorbanpour et al., (2016) stated that the increase in shoot fresh and dry weights and root length in *S. officinalis* may be related to IAA production of the PGPR isolates used. In our study, isolates that contributed positively to root and stem development had the ability to produce high or low levels of IAA (Table 1). Also, these EB isolates are also successful in terms of “1-aminocyclopropane-1-carboxylate deaminase” (ACCd) production, nitrogen fixation, and siderophore production ability, and they can contribute in this way. Therefore, the effects of these characters should not be ignored tooThe results of the study conducted by Samani et al. (2019) using the plant *S. officinalis* support our findings.Anbi et al., (2020) determined that bacteria that can fix nitrogen and have the ability to dissolve phosphorus increased the chlorophyll content of *S. officinalis*. Researchers conducted studies on garlic (Esringu et al., 2016), broccoli (Yıldırım et al., 2011), cabbage (Turan et al., 2014), calendula (Selem et al., 2022), and broad beans (Cirka et al., 2022) revealed that different bacterial applications are effective on the increasing amount of chlorophyll in plants.

It is known that the temperature increases in plants due to stress. It was revealed that the temperature increases in the plants are in a negative correlation with the yield (Blum, 2009). In the study carried out, it was indicated that the temperature in the plants treated with bacteria was lower than the control group and that the bacterial applications had positive effects on plant growth in terms of temperature parameters.

All the EB applications increased the nitrogen balance index (NBI) in leaves significantly compared to the control group. G129K1-1 isolate, which produced the most successful application, appears to have nitrogen fixation ability (Table 2). It was determined that bacteria with nitrogen fixation ability increased the leaf nitrogen content of Salvia spp. more than other phosphorus-dissolving bacteria (Anbi et al., 2020). Researchers stated that stress reduces nitrogen use efficiency and NBI values (Shakiba et al., 2010). The nitrogen balance in the sorghum plant decreases with stress and causes early leaf maturation (Chen et al., 2015). Similarly, Oral et al., (2021), in their study on soybean, showed that plant growth-promoting bacteria increased the NBI value in the plant compared to the control group. According to the findings obtained from our experiments, the lowest NBI value was obtained from the control group as a dualex index of 26.32, and it was determined that it was compatible with the relevant literature.

Flavonols are low molecular weight polyphenolic compounds found in many plants. They have important roles in flowering in plants. During pollination, they produce red-blue or yellow pigments that serve to attract insect and bee populations (Birman, 2012). In this study, no significant difference was found between the plants in the control group and the plants treated with PGPR in terms of flavonol content. Anthocyanins are water-soluble compounds of various colors such as flavonols. They are found in the cavities within the epidermal and mesophyll tissue in the plant. It creates a secondary defense mechanism in plants by providing color change in various organs against abiotic factors such as drought stress (Aztekin and Kasım, 2016). Studies have shown that there is an increase in the amount of anthocyanins under stress (Hanson et al., 2011). In our study, however, the highest anthocyanin value was found in the control group with a dualex index of 0.074. The amounts of anthocyanins in bacterial treatment groups were found to contain lower than the control.

The values of the color characteristics, L* (brightness), a* (red-green), b* (yellowish), Chroma, and Hue (color tone), of sage treated by EB the difference between them, were not statistically significant. In the study performed on the basil plant, the color values were determined as L*(48.60), a*(-14.18), and b*(25.19) (Inan, 2010). In the study performed on the basil plant, the color values were determined as L*(48.60), a*(-14.18), and b*(25.19). The hue angle is defined as a color circle and it has been observed in the study that it corresponds to the range of 90 to 180. Positive a* values are obtained in red, while negative a* values are obtained in green. In our study, it was observed that a* values were negative and the highest values were obtained in bacterial applications. The positive b* value represents yellow, while a negative value represents blue. In the study, the b* value was positive and the lowest value was determined as 18.85 in the control group. The researchers found that the effects of compost and chicken manure application in lettuce and tomatoes, respectively on color values were found insignificant (Sonmez et al., 2017 and 2019). It has been reported that Chroma (C) value expresses vividness-opacity and Chroma values are high in vivid colors and low in dull colors (Ozbay, 2021). It has been reported that the Chroma value in the spinach plant, where different fertilizer applications are applied, varies between 21.48 and 24.86 (Demir and Sonmez, 2019). In the study carried out, the lowest Chroma value was found to be 22.50 in the control group, while the highest was determined as 28.59 (Table 4). It was concluded that the plants treated with bacteria were brighter.

## Conclusion

Due to the fact that high yield is the priority in agricultural production, producers tend to use excessive fertilizers and the majority of these fertilizers negatively affect the environment and human health. Alternative methods have been developed in terms of reducing fertilizer applications to a low level, and PGPRs, which positively affect plant growth, have come to the fore. It was concluded that the bacteria used in the study made positive contributions to plant growth parameters. The appropriate plant-bacteria combinations will enable the production of environmentally friendly and sustainable *S. officinalis*. The studies to be carried out with different bacterial strains and sage varieties can make significant contributions to our country's agriculture.

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