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## ARAŞTIRMA MAKALESİ RESEARCH ARTICLE

# Enhancing sugar beet yield and soil microbial activity under water-limited conditions through mycorrhizal and bacterial inoculations

Mikoriza ve bakteri uygulamalarının su stres koşulları altında şekerpancarı verimi ve toprak mikrobiyal aktivitesine etkisi

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## **ABSTRACT**

Aim of this study is to investigate the effects of mycorrhizal and bacterial inoculations on sugar beet yield and soil microbial activity under varying water stress conditions. The field experiment was conducted over two successive years with three different irrigation levels: 33% (I1), 66% (I2), and 100% (I3). Mycorrhiza (Mikostar BTH-100) containing Glomus intraradices, Glomus mosseae, Glomus fasciculatum, and Glomus etunicatum, along with Bradyrhizobium japonicum bacteria, were applied to seeds during planting. Results showed that both applications significantly improved sugar beet yield and soil microbial activity compared to the control treatment. Mycorrhiza was particularly effective under full irrigation (I3), while bacterial inoculations showed stronger effects under moderate and low irrigation levels (I2 and I1, respectively). The highest yield (10130 kg/da) was observed under full irrigation with mycorrhiza treatment, while the lowest yield (3917.33 kg/da) was recorded in the control group under low irrigation. Soil microbial analyses revealed significant enhancements in CO₂ respiration, dehydrogenase activity (DHA), and microbial biomass carbon (MBC) in treated soils. These findings highlight the potential of mycorrhiza and bacteria to enhance plant performance and soil health under water-limited conditions, contributing to sustainable agricultural practices.

## ÖZET

Bu çalışmanın amacı, farklı su stres koşulları altında mikoriza ve bakteri uygulamalarının şekerpancarı verimi ve toprak mikrobiyal aktivitesi üzerindeki etkilerini araştırmaktadır. Deneme, iki yıl boyunca arazi koşullarında ve üç farklı sulama düzeyinde (I₁: %33, I₂: %66, l<sub>3</sub>: %100) yürütülmüştür. Bradyrhizobium japonicum bakterileri ile birlikte Glomus intraradices, Glomus mosseae, Glomus fasciculatum ve Glomus etunicatum içeren Mycorrhiza (Mikostar BTH-100) ekim sırasında tohumlara uygulandı. Elde edilen sonuçlar, her iki uygulamanın da kontrole kıyasla şeker pancarı verimini ve toprak mikrobiyal aktivitesini önemli ölçüde geliştirdiğini gösterdi. Mikoriza özellikle tam sulama (13) altında etkili iken, bakteriyel aşılamalar orta ve düşük sulama seviyeleri altında daha güçlü etkiler gösterdi (sırasıyla I2 ve I1). En yüksek verim (10130 kg/da) mikoriza tedavisi ile tam sulama altında gözlenirken, en düşük verim (3917.33 kg/da) düşük sulama altında kontrol grubuna kaydedildi. Toprak mikrobiyal analizleri, işlenmiş topraklarda CO₂ solunum, dehidrojenaz aktivitesi (DHA) ve mikrobiyal biyokütle karbonunda (MBC) önemli gelişmeler ortaya koymuştur. Bu bulgular mikoriza ve bakterilerin su sınırlı koşullar altında bitki performansını ve toprak sağlığını artırma potansiyelini vurgulamakta ve sürdürülebilir tarım uygulamalarına katkıda bulunmaktadır.

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#### **INTRODUCTION**

The rapid growth of the global population and the impacts of climate change necessitate sustainable agricultural practices, particularly in water-scarce regions. The Southeastern Anatolia region contributes a relatively small proportion to Turkey's total sugar beet production. In 2022, approximately 2.95 million decares of land in Turkey were dedicated to sugar beet cultivation, resulting in a total production of 19.25 million tons. While Konya province alone accounted for 35.45% of this production, the Southeastern Anatolia region's share remained limited due to its semi-arid climate and restricted irrigation opportunities (TEPGE, 2023).

With the advancements brought by the Southeastern Anatolia Project (GAP), sugar beet cultivation has been increasing, especially in Şanlıurfa, Muş, and Malatya provinces. Although the region does not yet match the productivity levels of the Central Anatolia region, which benefits from more favorable climatic conditions, the potential for growth is significant with enhanced irrigation practices (Hektaş, 2023).

In the Southeastern Anatolia region of Turkey, where water stress is a significant challenge, sugar beet cultivation faces productivity losses. This study was conducted to address these challenges by evaluating the role of mycorrhiza and bacteria applications in improving water use efficiency, yield, and soil microbial health. Currently, traditional agricultural practices in the region often have problems in optimizing water usage and maintaining soil efficiency in which this study aims to improve. Water stress significantly impacts plant growth, yield, and physiological functions (Farooq et al., 2019).

Sugar beet (*Beta vulgaris* L.) is a critical crop not only for sugar production but also for contributing to global and national food security. As a staple crop, sugar beet plays a vital role in the agricultural economy, providing raw materials for the food and biofuel industries (Bojović et al., 2024). However, maintaining sugar beet yields amid increasing water scarcity remains a significant agricultural challenge, necessitating innovative approaches to enhance crop resilience under drought conditions.

Water-limited conditions negatively affect not only crop yields but also soil health and nutrient cycling (Ghimire et al., 2023). Drought disrupts the balance of soil microbial communities, reduces nutrient availability, and impairs soil structure (Bogati & Walczak, 2022). For sugar beet, these conditions can lead to reduced root growth, lower sucrose content, and diminished crop quality, highlighting the need for solutions that improve water use efficiency and sustain soil fertility (Mubarak et al., 2016).

Mycorrhizal fungi play a crucial role in forming symbiotic relationships with plant roots, boosting the plants' ability to absorb water and nutrients. These fungi are especially effective in helping plants acquire nutrients like phosphorus, which are not easily available in soil, thus promoting healthier plant growth (Mickan et al., 2016). By expanding root surface areas and optimizing soil water availability, these fungi also help plants survive better under water stress (Behrooz et al., 2019). Certain arbuscular mycorrhizal fungi species, such as *Glomus intraradices*, *Glomus mosseae*, *Glomus fasciculatum*, and *Glomus etunicatum*, have proven effective in water-limited conditions (Begum et al., 2022; Rahimzadeh & Pirzad, 2017).

Similarly, rhizosphere bacteria support plant health by releasing growth-promoting substances and enhancing soil microbial activity (Mickan et al., 2019; Sheteiwy et al., 2021). For instance, *Bradyrhizobium japonicum* can promote plant growth through nitrogen fixation and even shows benefits under water stress conditions (Laranjeira et al., 2021). These bacteria also increase microbial diversity and activity in the soil, contributing to sustainable soil ecosystems (Silva et al., 2023).

Biological treatments, such as mycorrhizal fungi and rhizosphere bacteria, have shown potential in enhancing soil quality and plant performance under abiotic stress conditions (Wahab et al., 2023). These microbial agents improve soil structure, increase nutrient bioavailability, and stimulate plant growth by enhancing root architecture and stress tolerance (Ranjan et al., 2024). Previous studies have demonstrated their effectiveness in various crops, but limited research has focused on sugar beet under water-limited conditions.

The mechanisms through which these biological amendments operate include promoting root elongation, enhancing nutrient uptake efficiency, and improving soil moisture retention (Ullah et al., 2021). For example, arbuscular mycorrhizal fungi facilitate phosphorus uptake, while *Bradyrhizobium japonicum* contributes to nitrogen fixation, which is particularly beneficial under nutrient-deficient and drought conditions (Chang et al., 2017). Additionally, these treatments can induce systemic resistance in plants, helping them manage abiotic stresses more effectively.

This study aims to evaluate how effectively mycorrhizal and bacterial treatments can mitigate the harmful effects of water stress on sugar beet yield and soil microbial activity. Sugar beet, a highly valuable crop, requires innovative strategies to improve its performance under drought due to its particular sensitivity. To this end, the research will focus on a mycorrhizal mixture (Mikostar BTH-100) containing *Glomus intraradices, Glomus mosseae, Glomus fasciculatum,* and *Glomus etunicatum,* along with *Bradyrhizobium japonicum,* under varying water stress conditions.

This research addresses a critical gap in the literature regarding the role of microbial treatments in improving sugar beet resilience under drought conditions. By conducting a two-year field experiment under different irrigation levels, this study will provide valuable insights into the practical applications of biotechnological methods in sustainable agriculture and water management. The findings are expected to contribute to the development of more resilient and productive agricultural systems, particularly in semi-arid regions facing water scarcity.

An economic analysis of mycorrhiza and bacteria applications compared to the control group was not included in this study. However, considering the potential of these applications to enhance yield and water use efficiency, future studies focusing on economic analyses are recommended. Such analyses could provide valuable insights into the cost-effectiveness of biological treatments for farmers and contribute to broader agricultural sustainability.

#### **MATERIALS and METHODS**

## Characteristics of the experimental field

The research was conducted in the R&D work area of Harran University Faculty of Agriculture during the 2018 and 2019 growing seasons.

Harran Plain is under the influence of the continental climate characteristics of the Southeastern Anatolia region and the Mediterranean climate. Summers in the plain are hot and dry, and winters are cold and rainy. The climate data of the research area for many years, 2018 and 2019, are given in Table 1.

Table 1. Climate characteristics of the research area *Çizelge 1. Araştırma alanının iklim özellikleri* 

|                                      | Years | April | May  | June | July | August | September | October |
|--------------------------------------|-------|-------|------|------|------|--------|-----------|---------|
| May Town (°C)                        | 2018  | 32.1  | 36.3 | 43.1 | 43.2 | 42.2   | 41.5      | 34.2    |
| Max. Temp. (°C)                      | 2019  | 26.8  | 40.3 | 44.1 | 42.3 | 45.8   | 39.5      | 36.2    |
| Min. Temp.                           | 2018  | 9.3   | 12.2 | 16.2 | 21.2 | 20.8   | 17.7      | 9.3     |
| (°C)                                 | 2019  | 5.9   | 10.1 | 18.5 | 19.7 | 20.7   | 15.9      | 11.3    |
| Ave. Temp.                           | 2018  | 19.9  | 23.0 | 28.6 | 31.9 | 32.2   | 28.8      | 21.6    |
| (°C)                                 | 2019  | 14.4  | 25.2 | 30.7 | 31.7 | 32.8   | 27.9      | 22.9    |
| Ave. Relative humidity (%)           | 2018  | 38.4  | 50.1 | 36.6 | 34.2 | 33.6   | 31.3      | 45.6    |
| Ave. Relative number (%)             | 2019  | 67.0  | 35.8 | 30.6 | 29.6 | 29.3   | 30.3      | 44.9    |
| Ave. Wind Speed (m s <sup>-1</sup> ) | 2018  | 1.3   | 1.2  | 2.0  | 2.0  | 1.8    | 1.5       | 1.1     |
| Ave. Willa Speed (iii s )            | 2019  | 1.2   | 1.6  | 1.8  | 2.0  | 1.8    | 1.5       | 1.2     |
| Painfall (mm)                        | 2018  | 35.8  | 64.5 | 10.1 | 0.0  | 0.0    | 2.2       | 39.4    |
| Rainfall (mm)                        | 2019  | 97.4  | 7.3  | 8.9  | 0.0  | 0.0    | 0.2       | 45.1    |

The soil characteristics of the research area belong to the Ikizce series, formed from colluvial parent materials and consisting of medium-deep soils with nearly level slopes. The entire profile contains a high clay content. The soil pH varies between 7.4 and 7.8 at the surface, and while the organic matter content is about 1.1% at the surface, it decreases to approximately 0.8% in deeper layers (Dinç et al., 1988). Soil samples representing the experimental site were analyzed at the GAP Agricultural Research Institute laboratory. They are presented physical properties in Table 2.

Table 2. Physical properties of trial area soils Çizelge 2. Deneme alanı topraklarının fiziksel özellikleri

| Depth<br>(cm) | Field<br>capacity<br>(g g <sup>-1</sup> ) | Wilting<br>point<br>(g g <sup>-1</sup> ) | Bulk<br>density<br>(g cm <sup>-3</sup> ) | Sand<br>(%) | Clay<br>(%) | Silt<br>(%) | рН   | EC<br>(dS m <sup>-1</sup> ) | Org.<br>Matter<br>(%) | Lime<br>(%) |
|---------------|---|--|--|-------------|-------------|-------------|------|-----------------------------|-----------------------|-------------|
| 0-30          | 0.280                                     | 0.167                                    | 1.37                                     | 20.40       | 54.00       | 25.60       | 7.85 | 0.67                        | 2.33                  | 7.90        |
| 30-60         | 0.288                                     | 0.174                                    | 1.39                                     | 20.40       | 52.00       | 27.60       | 7.94 | 0.59                        | 1.31                  | 9.50        |
| 60-90         | 0.296                                     | 0.182                                    | 1.40                                     | 20.30       | 53.50       | 26.20       | 7.92 | 0.63                        | 1.42                  | 9.90        |

The irrigation water requirement was met from the Atatürk Dam, located through the middle of the field and providing a discharge capacity of 200 m³ s⁻¹, as well as from the Mardin-Ceylanpınar (MC) conveyance channel. In the experimental area, irrigation was applied using a drip irrigation system composed of Ø32 manifold and Ø16 lateral pipes. The irrigation water used in the study was sourced from the Atatürk Dam and the Mardin-Ceylanpınar (MC) conveyance channel. The water quality was analyzed, showing a pH of 7.2 and an EC of 0.54 dS/m, classified as C2S1 according to standard irrigation water quality criteria. These values indicate moderate salinity with a low risk of sodium hazards, suitable for agricultural use under proper management practices.

## Plant type used in the research

In this study, a certified genetic monogerm variety of sugar beet from Türkşeker was used. Images reflecting the state of sugar beet plants during the experiment are presented in Figure 1.

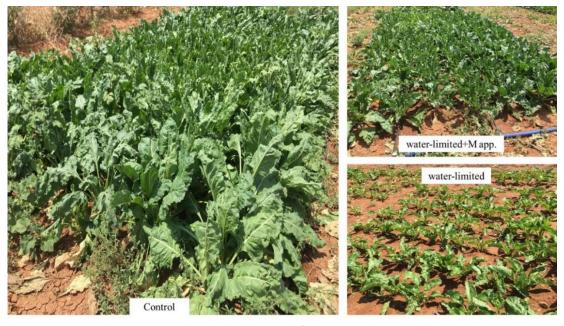


Figure 1. Some images from trial parcels Şekil 1. Deneme parsellerinden bazı görüntüler

During the course of the research, pesticide treatments were carried out to prevent and reduce possible disease and pest occurrences. Prior to sugar beet sowing, Pyramin Super herbicide, containing an active substance of 520 g L<sup>-1</sup> Chloridazon, was applied to manage weed and harmful vegetations.

As for fertilization, prior to sowing, 6-8 kg da<sup>-1</sup> of ammonium sulfate and 8-10 kg da<sup>-1</sup> of TSP (triple superphosphate) were applied at the base. Additionally, before the first hoeing, an additional 6-8 kg da<sup>-1</sup> of ammonium sulfate was applied.

## Field design and treatments

The experiment was conducted in a randomized block design with split plots, featuring three main factors, three sub-levels, and three replications. The main factors consisted of Mycorrhiza, Bacteria, and a control (no treatment), while the sub-factors were the different irrigation water levels (I1: 33%, I2: 66%, and I3: 100%). The sugar beet seeds were sown using a planter adjusted to a 40 cm inter-row spacing and 40 cm intra-row spacing, on March 30, 2018 and April 15, 2019, respectively.

In order to determine the soil moisture contents in the research area, samples were collected from depths of 0-30 cm, 30-60 cm, and 60-90 cm. Afterwards, these samples were placed in an oven at 105°C for 24 hours, and later their moisture content was measured following Peterson et al. (1982).

In this research, a drip irrigation system was utilized. During the initial irrigation, enough water was applied to bring the soil moisture up to field capacity, and for the following three weeks, equal amounts of irrigation water were supplied to all treatments, allowing the seedling root system to properly develop. Once the plants adapted to the field conditions, the different irrigation treatments were initiated.

To calculate the irrigation water amount, the open-water surface evaporation method described by James et al. (1982) was employed, and Equation 3.1 (provided below) was used.

$$I=A* [_Ep] *k_cp*P$$
 (3.1)

In Equation 3.1, I is the amount of irrigation water applied to the plot (L), A is the plot area (m²), Ep is the cumulative Class A Pan evaporation over the irrigation interval (mm), kcp is the chosen Pan coefficient, and P is the percentage of soil cover (%). In this study, the P value was taken as 100%. In the experimental plots, the first irrigation was performed when the available water in the 90 cm soil profile decreased to about 35%, and then enough irrigation water was applied to bring the soil moisture back up to field capacity. For subsequent irrigations, Equation 3.1 was used and the irrigation interval was set at 4 days. During the subject irrigations, the applied irrigation water quantities were measured and controlled through water meters.

In determining seasonal plant water consumption values regarding the subjects included in the research. James et al. (1982) water balance equation (3.2) method was applied.

$$[ET] _a=I+P+C_r-(R_f+D_P)\pm\Delta S$$
 (3.2)

In Equation 3.2, ETa represents the crop water consumption (mm), I is the irrigation water (mm), P is the effective rainfall (mm), Cr is the capillary rise (mm), Dp is the deep percolation (mm), Rf is the surface runoff losses (mm), and ΔS refers to the moisture variation in the soil profile (mm). Since the research area does not suffer from deep drainage or salinity issues, and considering that drip irrigation was used, neither capillary rise from groundwater nor surface runoff occurs. Therefore, Cr, Rf, and Dp values were not included in the calculations. The amount of water retained in the soil profile (mm) was determined by the difference in moisture content measured at the beginning and the end of the crop growth period.

In this research, a mycorrhizal mixture called Mikostar BTH-100 was used, containing *Glomus intraradices, Glomus mosseae, Glomus fasciculatum*, and *Glomus etunicatum* strains, along with a soybean bacterium identified as strain

110. The mycorrhiza application was done to the seeds at the time of sowing, and similarly, the bacterial inoculation was also performed by treating the seeds during planting. Thus, both mycorrhiza and bacterial treatments were applied directly onto the sugar beet seeds at sowing.

## Soil and plant analyses

In soil samples representing the experimental field, pH and salinity were determined by the dilution method (U.S. Salinity Laboratory Staff, 1954), while organic matter content was measured through titration with iron sulfate (Walkley & Black, 1934), and lime content via the Scheibler calcimeter (Çağlar, 1949). Microbial analyses included the assessment of carbon dioxide (CO<sub>2</sub>) production, dehydrogenase (DHA) activity, and microbial biomass carbon (MBC), which are critical indicators of soil microbial activity and health. CO₂ production was measured using the Isermayer (1952) method, where soil samples were incubated under controlled conditions, and the CO₂ released was absorbed in an alkaline solution, providing an indicator of microbial respiration and overall soil microbial activity. Dehydrogenase activity (DHA) was determined following Thalman (1967), where a substrate such as triphenyltetrazolium chloride (TTC) was added to soil samples, and its reduction by dehydrogenase enzymes produced a colored compound. The intensity of this color, measured spectrophotometrically, correlated with the microbial oxidative activity in the soil. Microbial biomass carbon (MBC) was assessed using the chloroform fumigation-extraction method proposed by Öhlinger (1993), which involved comparing the amount of carbon extracted from fumigated and non-fumigated soil samples to estimate the biomass of living microorganisms in the soil, offering insights into microbial population and nutrient cycling capacity. The heads of the plants in the harvest plots were taken and their yield per decare was determined (Ermiş, 1998). The sugar content in sugar beet was determined according to the equation given by Reinefeld et al. (1974) at the end of harvest.

 $PSR=P-[0.343(Na+K)+0.094x+\alpha N+0.29]$ 

Accordingly:

PSR: Purified sugar rate

P: Polar (%)

K: Potassium (mmol / 100 g rooth)

αN: Amino Nitrogen (mmol / 100 g rooth)

#### Statistical analysis

The statistical analysis was conducted using the split-plot ANOVA approach within the SPSS statistical package program, as the experiment followed a randomized block design with split plots. A two-way ANOVA was performed to evaluate the main effects of applications (A) and irrigation levels (B), as well as their interaction (A  $\times$  B). To determine significant differences among treatment means, Duncan's multiple range test was applied as a post-hoc analysis. In response to reviewer suggestions, tables have been revised to enhance the clarity of data presentation and facilitate a more comprehensive interpretation of the results.

#### **RESULTS and DISCUSSIONS**

## Yield

The results indicate that mycorrhiza and bacterial applications had positive impacts on sugar beet yield under water-stress conditions. Data obtained in 2018 and 2019 (Table 3A) clearly show that both treatments improved yields at all irrigation levels when compared to the control group.

In 2018, it was observed that yield losses increased as irrigation levels decreased. Nevertheless, mycorrhiza and bacterial applications proved particularly effective in mitigating these reductions at the lowest irrigation level (I<sub>1</sub>).

Under 33% irrigation, the mycorrhizal treatment provided about 30.7% higher yield than the control, while the bacterial application increased yield by approximately 21.6%. At the moderate irrigation level (I<sub>2</sub>), the influence of both mycorrhiza and bacteria on yield became more evident, with mycorrhiza producing the highest yield. Under full irrigation conditions (I<sub>3</sub>), both treatments significantly boosted yields, and mycorrhiza again performed better than the bacterial treatment. Overall, in 2018, mycorrhiza applications achieved the highest average yield (7820.87), followed by bacterial treatments (7343.89), while the control group recorded the lowest yield (6267). Although similar trends were observed in 2019, there was a slight decrease in overall yield values, possibly attributable to varying climatic conditions or annual changes in soil characteristics. Under the low irrigation level (I<sub>1</sub>) in 2019, mycorrhiza and bacterial applications increased yields by approximately 25.8% and 22.6%, respectively, compared to the control. At the moderate irrigation level (I2), mycorrhiza once again delivered the highest yield (8066.67), followed by the bacterial application (7619.33). Under full irrigation conditions (I<sub>3</sub>), mycorrhiza achieved the top yield (10130.00), while the bacterial treatment also demonstrated a substantial increase (9589.00). Considering average values, mycorrhiza applications (7708.56) continued to produce the highest yields in 2019, followed by bacterial treatments (7337.00), whereas the control group recorded the lowest yield values (6251.22). In both years, crop yield increased as irrigation levels rose, highlighting the essential role of water in supporting plant growth. However, under limited irrigation conditions, the yield-boosting effects of mycorrhiza and bacteria were even more evident. The differences in yield between the two years can likely be attributed to varying environmental conditions. Yet, considering the results from both years, it is clear that mycorrhiza applications were more effective than bacterial treatments. Both approaches, however, significantly enhanced plant performance under water stress.

Research has consistently shown that mycorrhizal fungi improve water and nutrient absorption, enabling plants to better tolerate water stress and achieve higher yields under both full and limited irrigation conditions (Augé, 2001; Smith & Read, 2010). Similarly, bacterial treatments, especially those using PGPR strains, help plants adapt to water stress by producing growth-promoting hormones and fixing nitrogen, thereby increasing water use efficiency (Vurukonda et al., 2016). Under reduced irrigation (I<sub>1</sub> and I<sub>2</sub>), both mycorrhiza and bacteria provided higher yields than the control group, with particularly strong results in 2019.

Full irrigation ( $I_3$ ) amplified the positive effects of mycorrhiza and bacteria in both years by optimizing water and nutrient efficiency, which led to significant yield improvements. Mycorrhiza applications stood out for their ability to enhance phosphorus uptake and root development, contributing to higher yields (Yang et al., 2018; Miransari, 2010). Bacteria, on the other hand, mitigated the adverse effects of water stress under limited irrigation ( $I_1$  and  $I_2$ ) by improving osmotic regulation and promoting root growth, which also resulted in increased yields (Bhattacharyya & Jha, 2012).

These findings underscore the potential of biological applications, particularly the combination of mycorrhiza and bacteria, to play a key role in sustainable agricultural practices by improving crop performance and resilience under varying irrigation conditions.

## Sugar content

When examining the effects of the study on sugar beet sugar content, it becomes evident that both mycorrhiza and bacterial treatments increased sugar percentages compared to the control group. The results obtained in 2018 (Table 3B) show that under low irrigation conditions ( $I_1$ ), mycorrhiza and bacterial applications notably raised sugar percentages. Mycorrhiza treatment recorded 16.16%, which is about 6.6% higher than the control (15.16%), while the bacterial application reached 15.63%, offering a more modest improvement. Under moderate irrigation ( $I_2$ ), the bacterial treatment achieved the highest sugar content (17.81%), though mycorrhiza (15.98%) also exhibited a significant increase compared to the control (14.48%). Under full irrigation conditions ( $I_3$ ), the control group presented the lowest sugar content (13.85%), whereas the bacterial treatment's effect (17.48%) stood out

Table 3. Sugar beet yield and sugar beet sugar ratios (%) in 2018 and 2019 in the study their grouping according to the Duncan test

Çizelge 3. Araştırmada 2018 ve 2019 yılı şeker pancarı verimi ve şeker pancarı şeker oranları (%) Duncan testine göre gruplandırılması

| Tuestusente  | Sugar beet yiel | d [(kg da <sup>-1</sup> ) (A)] | Sugar beet sugar ratios [(%) (B)] |        |  |
|--------------|-----------------|--------------------------------|-----------------------------------|--------|--|
| Treatments   | 2018            | 2019                           | 2018                              | 2019   |  |
| С            | 6267c           | 6251.22c                       | 14.5c                             | 14.49c |  |
| M            | 7820.87a        | 7708.56a                       | 15.49b                            | 15.49b |  |
| В            | 7343.89b        | 7337.00b                       | 16.97a                            | 16.93a |  |
| P (Factor A) | **              | **                             | **                                | **     |  |
| 11           | 4677.44c        | 4549.67c                       | 15.64b                            | 15.58b |  |
| 12           | 7383.33b        | 7378.67b                       | 16.09a                            | 16.13a |  |
| 13           | 9371.00a        | 9368.44a                       | 15.22c                            | 15.20c |  |
| P (Factor B) | **              | **                             | **                                | **     |  |
| C-I1         | 3982.33ı        | 3917.33h                       | 15.16e                            | 15.10f |  |
| C-12         | 6496.33f        | 6450.00f                       | 14.48f                            | 14.59g |  |
| C-13         | 8322.33c        | 8386.33c                       | 13.85g                            | 13.77ı |  |
| M-I1         | 5206.33g        | 4929.00g                       | 16.16c                            | 16.11c |  |
| M-I2         | 8069.33d        | 8066.67d                       | 15.98c                            | 15.96d |  |
| M-I3         | 10187.00a       | 10130.00a                      | 14.33f                            | 14.41h |  |
| B-I1         | 4843.67h        | 4802.67g                       | 15.63d                            | 15.52e |  |
| B-I2         | 7584.33e        | 7619.33e                       | 17.81a                            | 17.85a |  |
| B-I3         | 9603.67b        | 9589.00b                       | 17.48b                            | 17.43b |  |
| P (A*B)      | **              | **                             | **                                | **     |  |

C: Control, M: Mycorrhiza application, B: Bacteria application, I1: 33%, I2: 66%, and I3: 100%,

prominently. Looking at the year 2019 (Table 3B), the results exhibited similar trends, though there was a general decrease in sugar content values. Under low irrigation ( $I_1$ ), mycorrhiza application achieved 16.11%, about 6.7% higher than the control (15.10%), while the bacterial treatment provided a more balanced increase at 15.52%. At the moderate irrigation level ( $I_2$ ), bacteria once again produced the highest sugar percentage (17.85%), while mycorrhiza (15.96%) still significantly surpassed the control (14.59%). Under full irrigation ( $I_3$ ), mycorrhiza (14.41%) showed an improvement compared to the control (13.77%), yet bacteria (17.43%) obtained the highest sugar ratio at this irrigation level as well.

Even though the overall trends were similar between years, small differences in absolute values were noticeable. In 2019, sugar percentages were slightly lower across all treatments, likely due to shifts in environmental conditions. The inverse relationship between irrigation level and sugar content in the control group was evident in both years. Under water-limited conditions, the ability of mycorrhiza and bacteria to enhance sugar content emphasizes the agricultural importance of these biotechnological approaches.

When the results are considered as a whole, bacterial treatments were found to be more effective than mycorrhiza applications in increasing sugar content, especially at low and moderate irrigation levels. Although mycorrhiza showed positive effects, its improvements were generally more modest compared to bacterial treatments. The marked decline in sugar content under low irrigation levels in the control group more clearly illustrates the potential of mycorrhiza and bacteria to alleviate adverse environmental conditions.

These findings indicate that mycorrhiza and bacterial applications could serve as effective strategies for enhancing sugar beet production, particularly in regions where water resources are limited. The significance of these biotechnological approaches has once again been underlined in terms of sustainable agriculture and water management.

<sup>\*\*</sup> Significant at the 1% level. \* Significant at the 5% level. ns: Not significant.

This study examined the effects of mycorrhiza (M) and bacterial (B) applications on the sugar content of sugar beet under different irrigation levels ( $I_1$ ,  $I_2$ ,  $I_3$ ). The two-year data (2018 and 2019) showed that both mycorrhiza and bacterial treatments increased sugar content compared to the control group (C). The highest sugar content in both years was achieved under the moderate irrigation level ( $I_2$ ) with the bacterial application (2018: 17.81%; 2019: 17.85%). Mycorrhiza also enhanced sugar content at all irrigation levels relative to the control, although its effect was slightly less pronounced than that of the bacteria.

Mycorrhizal fungi are known to play a crucial role in regulating plant carbon metabolism and promoting sugar synthesis (Wu et al., 2019; Augé, 2001). Research has shown that these fungi enhance sugar content by facilitating phosphorus uptake and supporting carbohydrate metabolism (Smith & Read, 2010). Similarly, bacterial treatments, particularly those involving PGPR strains, have been found effective in increasing sugar content. This is often attributed to their ability to fix nitrogen and produce growth hormones (Bhattacharyya & Jha, 2012; Vurukonda et al., 2016).

When analyzing the results by irrigation levels, sugar content was observed to be higher under limited irrigation conditions ( $I_1$  and  $I_2$ ) and lower under full irrigation ( $I_3$ ). This phenomenon can be explained by the plant's natural response to water stress, which involves accumulating carbohydrates to stabilize osmotic pressure (Koch, 2004). Numerous studies suggest that both mycorrhiza and bacteria play a supportive role in these osmotic regulation processes, helping to increase sugar content even under less favorable water conditions (Miransari, 2014).

Overall, the findings indicate that mycorrhiza and bacterial applications are particularly effective in increasing sugar content under moderate irrigation conditions ( $I_2$ ). These treatments also enhance plant quality and productivity, even when water availability is limited, making them valuable tools for sustainable agriculture in water-scarce environments.

#### Soil microbial analysis results (CO<sub>2</sub>, DHA ve MBC)

The results related to soil microbial respiration, reflected by  $CO_2$  emissions, show that mycorrhiza and bacterial applications substantially enhanced microbial activity compared to the control group. In 2018 (Table 4A), it was observed that  $CO_2$  emissions increased across all treatments as irrigation levels rose. Under the low irrigation level (I<sub>1</sub>), the control group (C) recorded a  $CO_2$  emission of only 1.93, whereas the mycorrhizal (M) treatment raised this value by about 189%, reaching 5.6. In the bacterial (B) treatment,  $CO_2$  emission climbed to 10.1, corresponding to roughly a 423% increase compared to the control.

Under moderate irrigation conditions ( $I_2$ ),  $CO_2$  emissions measured 3.17 in the control, 8.5 in the mycorrhiza-treated soil, and 11.63 in the bacteria-treated soil. These findings confirm that both treatments effectively boost microbial activity. At full irrigation ( $I_3$ ), the bacterial treatment achieved the highest  $CO_2$  emission (13.97), followed by mycorrhiza at 8.93. The control group's value remained low at 3.83.

In 2019 (Table 4A),  $CO_2$  emission values were generally higher than the previous year, suggesting that environmental conditions may have stimulated an increase in microbial activity. Under the low irrigation level ( $I_1$ ), both mycorrhiza and bacterial treatments led to a substantial rise in  $CO_2$  emissions compared to the control. While the control recorded 3.93, the mycorrhizal treatment raised this value by about 127% (reaching 8.93), and the bacterial treatment enhanced it by approximately 240% (reaching 13.37).

At the moderate irrigation level ( $I_2$ ), the mycorrhizal treatment pushed  $CO_2$  emissions up to 11.5, and the bacterial treatment held it steady at 13.3. Meanwhile, the control remained at a mere 3.93. Under full irrigation ( $I_3$ ), the bacterial treatment hit the highest  $CO_2$  emission level at 16.8, while the mycorrhizal treatment, reaching 11.6, also showed effectiveness in boosting microbial activity. The control group, on the other hand, stayed at 5.1.

Table 4. Sugar beet soil respiration (CO<sub>2</sub>), DHA and MBC results for 2018 and 2019 in the study their grouping according to the Duncan test

Çizelge 4. Çalışmada 2018 ve 2019 yılı şeker pancarı toprak solunum (CO<sub>2</sub>) sonuçları

| Treatments   | CO <sub>2</sub><br>(mg C 100g <sup>-1</sup> ds.24 h)(A) |        | DI<br>(µg TPF 10 <sup>-1</sup> | IA<br>gds. 24h)(B) | MBC<br>(mg MBC 100g <sup>-1</sup> kt)(C) |         |  |
|--------------|---|--------|--------------------------------|--------------------|--|---------|--|
| <del>-</del> | 2018  | 2019   | 2018                           | 2019               | 2018                                     | 2019    |  |
| С            | 2.98c   | 4.32c  | 158.45c                        | 160.45c            | 27.32c                                   | 28.43c  |  |
| M            | 7.68b   | 10.68b | 236.85b                        | 243.69b            | 162.79b                                  | 167.91b |  |
| В            | 11.90a  | 14.49a | 360.64a                        | 364.11a            | 206.81a                                  | 215.47a |  |
| P (Factor A) | **  | **     | **                             | **                 | **                                       | **      |  |
| 11           | 5.88b   | 8.74b  | 180.26c                        | 183.63c            | 117.99c                                  | 123.11c |  |
| 12           | 7.77a   | 9.58b  | 238.41b                        | 243.51b            | 131.93b                                  | 136.69b |  |
| 13           | 8.91a   | 11.17a | 337.27a                        | 341.11a            | 147.01a                                  | 152.01a |  |
| P (Factor B) | **  | **     | **                             | **                 | **                                       | **      |  |
| C-I1         | 1.93  | 3.93   | 137.60f                        | 140.53f            | 23.74d                                   | 24.92d  |  |
| C-I2         | 3.17  | 3.93   | 170.27ef                       | 171.59ef           | 29.29d                                   | 30.59d  |  |
| C-13         | 3.83  | 5.1    | 167.49ef                       | 169.25ef           | 28.94d                                   | 29.76d  |  |
| M-I1         | 5.6   | 8.93   | 211.89bcd                      | 215.18de           | 124.01c                                  | 129.92c |  |
| M-12         | 8.5   | 11.5   | 237.90cd                       | 247.12cd           | 162.28b                                  | 165.83b |  |
| M-13         | 8.93  | 11.6   | 260.76bc                       | 268.16bc           | 202.08a                                  | 207.99a |  |
| B-I1         | 10.1  | 13.37  | 191.30de                       | 195.18e            | 206.22a                                  | 214.49a |  |
| B-I2         | 11.63   | 13.3   | 307.06b                        | 311.24b            | 204.21a                                  | 213.66a |  |
| B-I3         | 13.97   | 16.8   | 583.57a                        | 585.91a            | 210.00a                                  | 218.27a |  |
| P (A*B)      | ns  | ns     | **                             | **                 | **                                       | **      |  |

C: Control, M: Mycorrhiza application, B: Bacteria application, I1: 33%, I2: 66%, and I3: 100%,

Although overall trends were similar between years, the general increase in  $CO_2$  emissions in 2019 across all groups underscores the potential of mycorrhiza and bacteria to stimulate microbial activity. While mycorrhiza provided notable improvements in microbial activity, bacterial treatments exhibited an even more pronounced effect. These findings once again highlight the potential of mycorrhiza and bacteria to improve soil microbial health and enhance soil fertility.

Dehydrogenase activity (DHA), considered an indicator of soil microbial activity, was found to be significantly increased by mycorrhiza and bacterial applications compared to the control. In 2018 (Table 4B), under low irrigation conditions (I<sub>1</sub>), the control recorded a DHA value of 137.60, while mycorrhiza raised it by about 54% to 211.89, and bacteria enhanced it by approximately 39% to 191.30. These outcomes indicate that both treatments can improve microbial activity even under limited water availability.

<sup>\*\*</sup> Significant at the 1% level. \* Significant at the 5% level. ns: Not significant.

At the moderate irrigation level ( $I_2$ ), a more pronounced rise in DHA was observed. The control's DHA measured 170.27, whereas mycorrhiza pushed this figure up to 237.9, and the bacterial treatment reached 307.06, providing the highest value. Under full irrigation ( $I_3$ ), the control maintained a DHA level of 167.49, while mycorrhiza increased it to 260.76, and bacteria soared to 583.57, clearly outperforming the control. These findings highlight that bacterial treatments were notably more effective at boosting microbial activity under full irrigation conditions.

At the end of the study, soil samples were collected to examine the microbial effectiveness of the treatments, and CO<sub>2</sub>, DHA, and MBC analyses were performed on the soils. The two-year results indicated that mycorrhiza and bacteria applications significantly increased soil CO<sub>2</sub> emissions compared to the control group (C). The highest CO<sub>2</sub> emission values in both years were obtained with bacterial application under full irrigation conditions (I<sub>3</sub>). Although the mycorrhiza application also enhanced CO<sub>2</sub> emissions at all irrigation levels, its effect remained lower than that of the bacteria.

Mycorrhizal fungi are widely recognized for their role in supporting soil respiration and contributing to the carbon cycle through their symbiotic relationships with plants (Bender et al., 2016; Wang et al., 2016). Research has shown that mycorrhiza aids in CO<sub>2</sub> emissions by breaking down organic matter in the soil and boosting root activity (Drigo et al., 2008; Wu et al., 2014). Similarly, bacterial applications, particularly those involving PGPR strains, enhance soil microbial activity and carbon metabolism, leading to increased CO<sub>2</sub> emissions (Jia et al., 2020).

When examined across irrigation levels, both mycorrhiza and bacterial treatments demonstrated their highest effectiveness in optimizing CO<sub>2</sub> emissions under full irrigation (I<sub>3</sub>). This is likely due to the increased microbial activity when water availability is not a limiting factor (Wang et al., 2018). Under lower irrigation levels (I<sub>1</sub>), bacterial applications were found to enhance CO<sub>2</sub> emissions by promoting carbon metabolism and root respiration. Studies suggest that even under water-stressed conditions, bacteria play a crucial role in maintaining the soil's biological processes by improving carbon metabolism (Sun et al., 2022).

These findings highlight the significant contributions of mycorrhiza and bacteria to soil carbon metabolism and their relationship to varying irrigation levels. Their role in the carbon cycle underscores the importance of incorporating biological treatments into sustainable agricultural practices, particularly in addressing challenges related to water availability and soil health.

The 2019 data (Table 4B) largely followed similar trends to those observed in 2018, although a slight increase in absolute values was noticeable. Under low irrigation ( $I_1$ ), the control group's DHA measured 140.53, while mycorrhiza raised it to 215.18 and the bacterial treatment brought it up to 195.18. At the moderate irrigation level ( $I_2$ ), the control registered a DHA value of 171.59, which rose to 247.12 with mycorrhiza and 311.24 under bacterial treatment. Under full irrigation ( $I_3$ ), DHA for the control stood at 169.25, while mycorrhiza moved this figure up to 268.16, and bacteria pushed it to 585.91.

When considering both years together, bacterial treatments increased DHA levels more than mycorrhiza did. Although mycorrhiza applications also provided a clear improvement compared to the control, their impact remained somewhat lower than that of bacteria. As irrigation levels rose, DHA values generally climbed, and bacterial treatments were particularly notable for producing very high DHA readings under full irrigation conditions. Differences between the two years may be linked to environmental variations, since all groups showed slightly higher DHA levels in 2019.

In conclusion, both mycorrhiza and bacterial treatments effectively enhance soil microbial activity, with bacterial treatments exhibiting the strongest effects under full irrigation. These findings reaffirm that mycorrhiza and bacteria hold potential as sustainable tools for improving soil biological health in agricultural systems.

According to the study's findings, both mycorrhiza and bacterial applications significantly increased the microbial biomass carbon (MBC) values in the soil. Data obtained in 2018 (Table 4C) showed that the control group (C) had low MBC values at all irrigation levels, while mycorrhiza (M) and bacteria (B) treatments notably raised these values. At the lowest irrigation level (I<sub>1</sub>), the control group recorded an MBC value of 23.74, whereas the mycorrhiza

application boosted this figure by approximately 422%, reaching 124.01. Under the bacterial treatment, MBC was measured at 206.22, about 768% higher than the control.

At the moderate irrigation level ( $I_2$ ), the control had an MBC value of 29.29, which mycorrhiza elevated to 162.28, indicating an increase of about 454%. Still, the bacterial application, at 204.21, surpassed mycorrhiza's value. Under full irrigation conditions ( $I_3$ ), the control's MBC stood at 28.94, while mycorrhiza and bacteria achieved much higher readings of 202.08 and 210.00, respectively.

One of the important indicators examined is DHA activity, which reflects the effectiveness of the applied treatments. The highest DHA values in both years were obtained through bacterial treatments under full irrigation ( $I_3$ ). While mycorrhiza also increased DHA activity, it was not as effective as the bacterial applications.

ehydrogenase activity (DHA) is a vital indicator of soil microbial activity, playing a significant role in the breakdown of organic matter and serving as a measure of soil quality (Xu et al., 2022). Studies show that mycorrhizal fungi enhance soil microbial diversity and activate enzymes, which in turn boost DHA levels (Cao et al., 2021; & Mummey 2006). Similarly, bacterial treatments, particularly those using PGPR strains, accelerate microbial processes by increasing soil enzyme activity, leading to higher DHA levels (Rawat et al., 2020; Mohamed et al., 2020).

Analysis by irrigation levels revealed that DHA levels were highest under full irrigation ( $I_3$ ), indicating that mycorrhiza and bacteria perform best in moist soil conditions (Hu et al., 2024). While DHA levels decreased under reduced irrigation ( $I_1$  and  $I_2$ ), both mycorrhiza and bacteria still managed to improve DHA levels compared to the control group. This suggests that even under less-than-optimal water conditions, microbial treatments can effectively enhance soil health and microbial activity.

These findings emphasize the potential of mycorrhiza and bacteria applications to maintain and improve soil quality, making them valuable tools for sustainable agriculture, especially in environments with varying water availability.

In 2019 (Table 4C), similar trends were observed, yet there was a general increase in MBC values for all groups. This suggests that yearly variations in environmental conditions may have had a positive effect on microbial activity. At the lowest irrigation level ( $I_1$ ), the control group's MBC value was measured at 24.92, while the mycorrhiza treatment raised it to 129.92, indicating about a 421% increase. Under the bacterial treatment, MBC reached 214.49, corresponding to a 760% rise compared to the control.

At the moderate irrigation level ( $I_2$ ), the control's MBC was 30.59, which increased to 165.83 with mycorrhiza, and 213.66 with bacteria. Under full irrigation conditions ( $I_3$ ), the control measured 29.76, whereas with mycorrhiza this climbed to 207.99, and with bacteria it reached 218.27.

Overall, both mycorrhiza and bacterial applications significantly enhanced MBC values compared to the control, showing similar patterns in both years. Bacterial treatments provided higher MBC levels than mycorrhiza across all irrigation regimes, with the difference becoming more pronounced under full irrigation. The persistently low MBC levels in the control group clearly demonstrate the capacity of mycorrhiza and bacteria to support microbial biomass formation.

The increase observed in MBC values between years could be linked to variations in environmental conditions and the improvement of the soil's microbial structure. These findings suggest that mycorrhiza and bacterial applications play an essential role in enhancing soil microbial health, indicating their substantial potential for sustainability in agricultural practices.

Microbial Biomass Carbon (MBC), a key indicator of microbial activity, reflects the size and activity of soil microbial communities. Mycorrhizal fungi, through their symbiotic relationships with plant roots, are known to enhance soil microbial diversity and play a critical role in the carbon cycle (Hartmann et al., 2015; Braghiere et al., 2021). By promoting phosphorus uptake and increasing root activity, mycorrhiza contributes significantly to elevated MBC levels (Treseder & Lennon, 2015). Similarly, bacterial applications boost microbial biomass by fixing nitrogen and

releasing root exudates, further enriching soil microbial communities (Sarma, 2024; Kuzyakov & Blagodatskaya, 2015).

When assessed across irrigation levels, MBC was highest under full irrigation (I<sub>3</sub>), where sufficient moisture conditions enhanced microbial activities and facilitated more efficient organic matter mineralization (Schimel, 2023). Under restricted irrigation (I<sub>1</sub> and I<sub>2</sub>), MBC levels were lower, but both mycorrhiza and bacterial treatments still improved MBC compared to the control group. This indicates that even under water-limited conditions, microbial applications effectively support soil biological activity.

These findings highlight the role of microbial treatments in maintaining and enhancing soil microbial communities, particularly in the face of water stress. The ability of mycorrhiza and bacteria to sustain MBC levels under varying irrigation conditions underscores their potential as essential tools for sustainable agricultural practices.

Table 5. Irrigation data for sugar beet in 2018 and 2019 in the study *Çizelge 5. Araştırmada 2018 ve 2019 yılı şeker pancarı sulama verileri* 

| Vanular          | IW<br>(mm) |      | E    | T <sub>c</sub> | Verim<br>(kg da <sup>-1</sup> ) |          |  |
|------------------|------------|------|------|----------------|---------------------------------|----------|--|
| Konular          |            |      | (m   | ım)            |                                 |          |  |
|                  | 2018       | 2019 | 2018 | 2019           | 2018                            | 2019     |  |
| C-I <sub>1</sub> | 495        | 472  | 517  | 486            | 3982.33                         | 3917.33  |  |
| C-I <sub>2</sub> | 890        | 843  | 911  | 859            | 6496.33                         | 6450.00  |  |
| C-I <sub>3</sub> | 1298       | 1226 | 1280 | 1234           | 8322.33                         | 8386.33  |  |
| M-I <sub>1</sub> | 495        | 472  | 529  | 490            | 5206.33                         | 4929.00  |  |
| $M-I_2$          | 890        | 843  | 934  | 857            | 8069.33                         | 8066.67  |  |
| M-I <sub>3</sub> | 1298       | 1226 | 1286 | 1248           | 10187.00                        | 10130.00 |  |
| B-I <sub>1</sub> | 495        | 472  | 526  | 493            | 4843.67                         | 4802.67  |  |
| B-I <sub>2</sub> | 890        | 843  | 927  | 869            | 7584.33                         | 7619.33  |  |
| B-I <sub>3</sub> | 1298       | 1226 | 1310 | 1246           | 9603.67                         | 9589.00  |  |

C: Control, M: Subject treated with mycorrhiza, B: Subject treated with bacteria, I<sub>1</sub>: Subject where 33% of the irrigation water need is met, I<sub>2</sub>: Subject where 66% of the irrigation water need is met, I<sub>3</sub>: Subject where 100% of the irrigation water need is met. Subject

When examining crop water consumption values for 2018 and 2019, it becomes evident that mycorrhiza and bacterial applications enhanced both water use and yield at all irrigation levels compared to the control. In both years, under low irrigation conditions, mycorrhiza and bacteria optimized water consumption and produced marked yield increases over the control. Mycorrhiza treatments, in particular, demonstrated greater effectiveness in improving water use efficiency, presenting the highest yield values under full irrigation. Although bacterial treatments followed mycorrhiza in terms of yield enhancement, they still provided significant improvements over the control group throughout both years. When comparing the two years, slightly lower irrigation and crop water consumption values were noted in 2019, which may be attributed to shifts in environmental conditions. These findings highlight that mycorrhiza and bacteria applications serve as strong, sustainable tools for improving yield and water use efficiency under water-limited scenarios.

In conclusion, this study highlights the significant potential of mycorrhiza and bacterial applications in enhancing sugar beet yield and soil microbial activity under varying water stress conditions. The findings indicate that both mycorrhiza and bacteria improve plant performance by optimizing water consumption and promoting soil microbial health. Mycorrhiza applications, in particular, demonstrated superior effectiveness, especially under full irrigation conditions, achieving the highest yield and microbial activity levels while maintaining efficient water use. Bacterial applications, while slightly less effective than mycorrhiza, also significantly enhanced both yield and soil microbial parameters compared to the control group.

The results emphasize that mycorrhiza and bacteria can mitigate the negative effects of water stress, particularly under limited irrigation scenarios, by improving plant water use efficiency and stimulating microbial activity in the soil. These outcomes underline the potential of these biological treatments to support sustainable agricultural practices, especially in regions where water resources are constrained.

Future research could explore the long-term impacts of combining mycorrhiza and bacteria applications under different environmental conditions and cropping systems. Additionally, further studies on optimizing application rates and understanding the interactions between these biological agents and soil properties could provide deeper insights into their broader applicability. Overall, this study reinforces the role of mycorrhiza and bacteria as valuable tools in achieving higher productivity and sustainability in agriculture under water-limited conditions.

This study aimed to address critical gaps in traditional agricultural practices, particularly concerning water use efficiency, soil microbial activity, and sustainable crop yield under stress conditions. Conventional methods often result in suboptimal water management and limited soil health improvements. By integrating biological treatments such as mycorrhiza and bacteria, this research demonstrated significant advancements in enhancing soil microbial diversity and promoting efficient water use, thereby offering practical solutions where traditional practices fall short.

While no specific economic analysis was conducted, the results suggest potential benefits for sustainable agriculture. Further research incorporating economic evaluations and exploring the long-term effects of these treatments under varying environmental conditions is recommended.

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## STATEMENT OF CONFLICT OF INTEREST

Authors have declared no conflict of interest.

## **AUTHOR'S CONTRIBUTIONS**

In the manuscript, Ali SARIOĞLU contributed to writing the manuscript, conducting field studies, and performing analyses. Sabri AKIN contributed to field studies, analyses, statistical analysis, and proofreading. Mehmet ŞİMŞEK contributed to the evaluation of the analyses and proofreading. Cengiz KAYA also contributed to the evaluation of the analyses and proofreading.

## STATEMENT OF ETHICS CONSENT

Ethical approval is not required as this article does not contain any studies with human or animal subjects.

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