

## Original article

# Effects of Microbial Fertiliser on Yield and Quality of Curly Lettuce Grown in Pots

Ufuk Uçan 👵 a, \*, Halil Demir 🕩 a & Hasan Kayhan Yalçi 📵 b

#### **Abstract**

Microbial fertilisers containing various microorganisms that are in a symbiotic relationship within each other and with plant roots have positive effects on plant growth, development and flowering. They can be used in conventional, organic and sustainable agriculture systems and reduce chemical fertiliser consumption. In this study conducted under greenhouse conditions, the effects of Herasim microbial fertiliser on yield and some quality characteristics of lettuce were investigated. Caipira (Lactuca sativa var. crispa) lettuce cultivar was used as plant material. In pot trials, 70% peat + 30% perlite mixture was used as a growing medium. A total of 9 different treatments were included in the research: Control (C), 100% Chemical Fertilisation (100% CF), 50% Chemical Fertilisation + Microbial Fertilisation (50% CF + MF), 100% Chemical Fertilisation + Microbial Fertilisation (100% CF), 50% Chemical Fertilisation + Microbial Fertilisation (100% CF + MF), Immersion + Chemical Fertilisation (I + CF), Microbial Fertilisation alone (MF), 50% Chemical Fertilisation + Microbial Fertilisation + Foliar Microbial Fertilisation (50% CF + MF + FMF). Head height (cm), root collar diameter (mm), number of leaves (number/plant), leaf colour (L\*, a\* and b\*), chlorophyll (SPAD), soluble solids (%SS), pH, total and marketable yield (g/plant) criteria were examined. The total and marketable yield results obtained from 100% CF + MF, I + CF, 75% CF + MF, 75% CF + MF + FMF and 50% CF + MF + FMF treatments were the highest and very similar to that of 100% CF treatment. In addition, similar or better results were obtained for the same treatments in terms of lettuce growth, colour, chlorophyll and SS criteria.

Keywords: Biofertiliser, Chlorophyll, Colour, Microbial Fertilisation, Lettuce, Quality.

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Ucan Ufuk is an doctoral candidate in the Department of Horticulture at Akdeniz University in Antalya, Türkiye. He has lived, worked, and studied in Antalya, Türkiye.

Email: ucanufuk@gmail.com

<sup>&</sup>lt;sup>a</sup> Department of Horticulture, Faculty of Agriculture, Akdeniz University, Türkiye

<sup>&</sup>lt;sup>b</sup> Herasim Agricultural Products Livestock and Environmental Sciences Ltd. Co., Türkiye

<sup>\*</sup> Corresponding author:

## INTRODUCTION

According to the data of TUIK, vegetable production in Turkey in 2023 is determined as 31.8 million tonnes. The most produced vegetable is tomato with 13.3 million tons, followed by watermelon with 3.2 million tons and onion with 2.6 million tons. The production of leafy vegetables, including lettuce, is over 2 million tonnes and lettuce production is 577 thousand tonnes (TÜİK 2024). Lettuce, one of the annual cool-climate vegetables whose leaves are mostly consumed (Aćamović-Đoković et al. 2011; Colonna et al. 2016) as salads and processed vegetables, is of great importance for human nutrition due to its rich vitamins, minerals, and antioxidants. Raw lettuce contains more nutrients than processed lettuce (Aćamović-Đoković et al. 2011).

One of the most significant challenges facing vegetable cultivation in our country is the relatively low yield per unit area, which varies depending on the region and on the species being cultivated (Aksoy & Altındişli, 1998). Since the beginning of the 20th century, chemical fertilisers, hormones and pesticides have been used extensively to increase yields (Aksoy, 1999). In today's intensive modern agriculture, the main objective is to achieve the maximum yield. However, the potential damage to the soil and the environment caused by chemical fertilisers is often overlooked, as highlighted by Parr et al. (1994). Chemicals lead to deterioration of soil structure and biological balance in the soil. Therefore, alternative organic inputs for agriculture are being investigated to regain the lost fertility and biological balance, and to grow crops that will bring benefit to environment and human health. Biofertilisers or microbial fertilisers are among these alternative products with significant potential.

Microbial fertilisers are organic products that generally contain microorganisms obtained from plant roots or root zones (Chen, 2006; Gupta & Sen, 2013). In other words, microbial fertilisers are products that contain different types of microorganisms that can mobilize elements in the soil that cannot be used by plants (Muraleedharan et al. 2010). Cocktail mixtures known as Effective Microorganisms consisting of lactic acid bacteria, phototrophic bacteria and yeast (Goessler & Kuehnelt, 2002) are used in plant cultivation, are physiologically compatible with each other and can coexist by working in a symbiotic relationship. There are studies suggesting that beneficial microorganisms can improve soil quality, plant growth and yield (Yamada & Xu, 2001). In addition, yeasts produce hormones and enzymes that promote plant cell and root division. The different microorganisms in the cocktail mixtures complement each other and establish a symbiotic relationship with the plant roots in the soil.

Plants grow extremely well in soils where effective microorganisms live (Sun et al. 2014). Plant growth promoting bacteria (PGPR) such as rhizobacteria are widely used as biofertilisers for many crops as well as vegetables (Gravel et al. 2007). Such organisms increase the availability of nutrients to the plant by reducing ethylene levels in plants or increasing the production of plant growth regulators such as indole-3-acetic acid (IAA). Physiological mechanisms also change the morphology of roots, facilitating root growth and increasing nutrient uptake (Vessey, 2003; Antoun & Prévost 2006;

Richardson et al. 2009). Microorganisms are also known to be capable of developing defences against both biotic and abiotic stress factors (Pieterse et al. 2014; Prasanna et al. 2014; Triveni et al. 2015). Beneficial microorganisms not only suppress soil-borne pathogens but also increase the decomposition of organic materials and the availability of plant nutrients and important organic compounds to plants (Singh et al. 2003). They have significant positive effects on improving soil fertility, plant growth, flowering, fruit development and ripening (Lévai et al. 2006). They can be used in conventional, organic and integrated farming systems to reduce the consumption of chemical fertilisers and pesticides and minimize their negative effects on the ecosystem (Molla et al. 2012). Microorganisms activate nutrients (macro and micro elements) and support plant growth (Colla et al. 2010; Wang et al. 2022).

Leafy vegetable production largely depends on the availability of NO3, which is the most important N source (Colla et al. 2011). Excessive N use can reduce leaf quality because it causes nitrate accumulation in the leaves of vegetables with a short vegetation period such as lettuce (Awaad et al. 2016). Pseudomonas, Bacillus, Lactobacillus, Paenibacillus and Pantoea bacteria are root bacteria that stimulate plant growth (Chen et al. 1996; Fálico et al. 2000; Luz, 2000; Pal et al. 2000; Wall, 2000). Research has focused particularly on the genera Pseudomonas and Bacillus (Compant et al. 2005). It has been reported that many Pseudomonas species promote rapid growth by affecting seed germination, growth and root development, and Bacillus megaterium helps in the solubilization of P and K in soil (Wu et al. 2012; Keshavarz Zarjani et al. 2013; Sindhu et al. 2016; Zhao et al. 2019) and promotes plant growth (Zou et al. 2010; Zhou et al. 2016; Korir et al. 2017). In one study, it was found that biofertilisers enriched with inorganic fertilisers played an important role in the growth and yield of tomatoes and reduced inorganic fertiliser costs by 50% (Haque et al. 2012). As an alternative approach, NPK fertilisers were reduced by 75% by the use of microbial fertilisers in bean cultivation (Chauhan & Bagyaraj, 2015). According to Zhao et al. (2021) B. megaterium, in combination with conventional fertilization increased cucumber yield by 11.8%-15.2%. They also found that cucumber yield and quality were not negatively affected when P and K were reduced from conventional fertilization. In another study, microorganisms significantly increased the weight of red lettuce and contributed to the highest vitamin C concentration (Stojanović et al. 2020).

This research was conducted to determine the effects of a microbial fertiliser Herasim, which contains a cocktail of different beneficial bacteria, on the growth, quality features and yield of curly lettuce grown in pots.

#### **MATERIALS and METHODS**

#### Materials

# Research Area and Growing Medium

This research was carried out by growing curly lettuce in pots in a glass greenhouse located at Akdeniz University, Türkiye. Before planting the seedlings, the pots (7 L) were filled with a mixture of 30% perlite and 70% peat. The salt content of the peat used is very low, the grain size is between 0-7 mm and the pH value is 6.

#### Plant Material

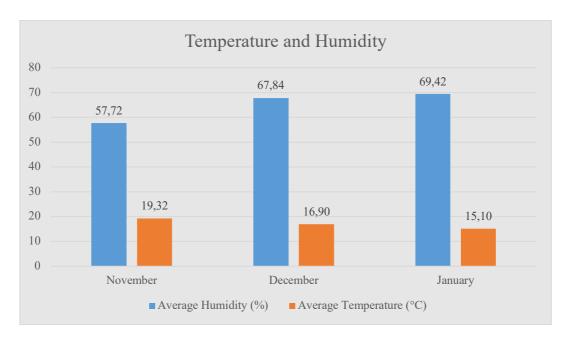
Caipira curly lettuce variety was used as plant material in the research and seedlings were purchased from a seedling company. Caipira curly lettuce variety is suitable for open fields, greenhouses, vertical farming and hydroponic systems. The head structure of Caipira, which is a curly, dark green lettuce variety, is homogeneous; the leaves are thick, juicy and crisp. The variety, which is suitable for spring and early autumn production in the open field in temperate coastal regions, is also suitable for late autumn, winter and early spring cultivation under cover. Especially in cold periods, due to the high number of leaves, it looks as if it has reached the harvest stage, which facilitates its sale. The maturity period of the variety varies between 50-60 days in hot periods and 70-90 days in cool periods. The average head weight at the harvest stage is between 750-1100 g. Caipira lettuce variety is resistant to the 16th-26th, 28th and 32nd strains of lettuce mildew, to lettuce mosaic virus and aphids.

#### Microbial Fertiliser Used in The Research

The microbial fertiliser used in the experiment is a product called 'Herasim Microbial Fertiliser' belonging to Herasim Agricultural Products Livestock and Environmental Sciences Ltd. Co. Herasim Microbial Fertiliser contains *Lactobacillus lactis*, *Lactobacillus rhamnosus*, *Lactobacillus plantarum*, *Lactobacillus acidophilus*, *Lactobacillus casei*, *Rhodopseudomonas palustris*, *Bacillus subtilis bacteria and Kluyveromyces lactis* and *Saccharomyces cerevisiae* yeasts. It also contains organic carob molasses and water filtered by activated carbon and sand filters. Herasim microbial fertiliser contains  $1x10^7$  cfu/ml total number of live microorganisms.

# Climatic Characteristics of The Research Area

A data logger was placed in the research greenhouse to record humidity and temperature values. After the seedlings were planted, the average humidity was recorded as 58.72% in November, 68.84% in December, and 70.42% in January. The average temperature was measured as 19.52 °C in November, 17.10 °C in December, and 15.26 °C in January. The humidity and temperature values obtained are shown in Figure 1.



**Figure 1.** Humidity and temperature values recorded in the research greenhouse.

## Properties of Irrigation Water Used in The Research

Well water has been used as irrigation water in the Faculty of Agriculture Research Area. Table 1 gives the pH and hardness of the irrigation water used in the research.

Table 1. Analysis of irrigation water

Parameters*	Value	Unit
рН	7.50	
EC (25 °C)	6.36	μmhos cm <sup>-1</sup>
Bicarbonate (HCO <sub>3</sub> -)	5.44	meq 1 <sup>-1</sup>
Calcium (Ca <sup>+2</sup> )	4.63	meq l <sup>-1</sup>
Magnesium (Mg <sup>+2</sup> )	1.07	meq l <sup>-1</sup>

<sup>\*</sup>Irrigation water characteristics were used together with climate characteristics in another study conducted in the same greenhouse.

## Method

# **Experiment Design and Treatments**

Within the scope of the research, 9 different treatments were included together with the control treatment. The study was carried out according to the random plots experimental design with 3 replications. Lettuce seedlings were transplanted as one seedling for each pot on 04.11.2022. Chemical and microbial fertiliser applications were applied for lettuce plants in pots by calculating per plant. Chemical fertiliser treatments were calculated per plant in accordance with the recommended doses of 6.5 kg da<sup>-1</sup> N, 2.5 kg da<sup>-1</sup> P<sub>2</sub>O<sub>5</sub>, 6 kg da<sup>-1</sup> K<sub>2</sub>O as suggested by Aybak (2002). Chemical fertiliser was

applied once a week on a certain day. Microbial fertiliser treatments to growing media were applied every 15 days on a certain day. Foliar microbial fertiliser treatments were applied once a week on a certain day. The treatments and their abbreviations are given in Table 2.

**Table 2.** Treatments and abbreviations included in the research

Treatments	Abbreviations
Control	С
100% Chemical Fertilisation	100% CF
50% Chemical Fertilisation+ Microbial Fertilisation	50% CF + MF
75% Chemical Fertilisation+ Microbial Fertilisation	75% CF + MF
100% Chemical Fertilisation+ Microbial Fertilisation	100% CF + MF
Immersion + Chemical Fertilisation	I + CF
Only Microbial Fertilisation	MF
50% Chemical Fertilisation+ Microbial Fertilisation + Foliar Microbial Fertilization	50% CF + MF + FMF
75% Chemical Fertilisation+ Microbial Fertilisation + Foliar Microbial Fertilization	75% CF + MF + FMF

## Application of Microbial Fertiliser

Before lettuce seedlings were planted, a solution was prepared by mixing 1 litre of microbial fertiliser (1 L/60 L) in 60 litres of water. Except for the lettuce seedlings to be planted in the control and chemical fertiliser alone treatments, the lettuce seedlings to be planted in the pots of the other treatments were planted after being immersed in the prepared solution for 30 min. After the seedlings were planted, the first irrigation was performed. One week after planting, microbial fertiliser and chemical fertiliser applications were started according to the research plan.

During the growing period, microbial fertiliser was applied with the irrigation water once every 15 days as 1 litre Herasim (1 L/60 L) per 60 litres of water. Microbial fertiliser was also applied weekly by foliar spraying as 1 litre of Herasim in 120 litres of water (1 L/120 L).

## Criteria Examined in Curly Lettuce

Curly lettuce plants that reached harvest maturity were harvested on 25 January 2023. The parameters measured and analysed in lettuce plants are given below. Total chlorophyll content was determined by SPAD 502 chlorophyll meter from the leaves of curly lettuce plants before harvesting. Harvested lettuce plants were crushed with a juicer and the TSS value was determined as % with a Hanna HI 96801 Model digital refractometer. Harvested lettuce plants were crushed with a juicer the pH of these juices was measured using a Hanna HI 2002-02 Model pH meter.

Colour measurements were taken as L\*, a\* and b\* on the third leaves from the outside of the harvested plants using the Minolta CR 400 Model colour chromameter, and the C (Chroma) and h<sup>o</sup> (hue) angle values were calculated using the \*a and \*b values.

C: 
$$\sqrt{(a^2+b^2)}$$

The L\*, a\* and b\* colour parameters show the colour values that the human eye can perceive, and L\* shows the changes in the brightness of the colour, reaching a maximum value as it approaches 100, which is called white. Of the colour values, a\* shows the changes from green to red, and b\* shows the changes from yellow to blue. Increasing negative or positive values of the measured values indicate darkening of the colour, positive values of a\* indicate red, negative values indicate green, and in the same way positive values of b\* indicate yellow and negative values indicate blue. The hue angle calculated using the determined colour values indicates 0=red, 90=yellow, 180=green and 270=blue (Siomos et al. 2002; Madeira et al. 2003; Demir et al. 2023).

## Statistical Analysis

The study was conducted according to the random plots trial design with 3 replications. The statistical analysis of the results was carried out with SAS 2009 Package Program and JPM Pro 17, an SAS program.



Figure 2. Lettuce plants in pot trials before harvest

## **RESULTS and DISCUSSION**

The effects of microbial fertiliser applications on lettuce head length (cm), root collar diameter (mm), number of leaves (number/plant), total soluble solids (TSS; %) and pH in lettuce juices are given in Table 3.

**Table 3.** Effects of microbial fertiliser applications on head length, root collar diameter, leaf number, TSS and pH of lettuce

Treatments	Head Length (cm)	Root Collar Diameter (mm)	Leaf Number (leaves/plant)	TSS (%)	pН
С	12.47 c*	14.92	39.00 b*	5.07 a*	6.23
100% CF	18.33 a	15.02	42.00 ab	4.20 b	6.21
50% CF + MF	17.03 ab	15.71	41.00 ab	4.03 b	6.22
75% CF + MF	19.30 a	15.43	42.00 ab	3.95 b	6.19
100% CF + MF	18.27 a	15.76	44.00 ab	3.87 b	6.17
I + CF	18.17 a	16.38	46.33 a	3.90 b	6.15
MF	14.33 bc	15.57	39.67 b	4.42 b	6.27
50%  CF + MF + FMF	19.53 a	15.51	46.00 a	3.90 b	6.31
75%  CF + MF + FMF	18.13 a	16.83	43.33 ab	3.97 b	6.25
LSD%5	2.7746	ns	5.8963	0.5836	ns

<sup>\*</sup>Differences between values not indicated with the same letter are significant at p<0.05 level.

ns: Not Significant

Different microbial fertiliser applications had significant effects on head length, leaf number and TSS, but no significant effects on root collar diameter and pH (p<0.05). The shortest plants in terms of head length were measured in the C plot, followed by and MF and 50% CF+MF plots. The highest values were obtained from 50% CF + MF + FMF, 75% CF + MF, 100% CF, 100% CF+ MF, I + CF and 75% CF + MF + FMF treatments. Root collar diameter values varied between 16.83-14.92 cm and there were no significant differences between treatments. When the number of leaves was compared in different treatments, the plants with the lowest number of leaves were obtained in the C and MF treatments, while there were no significant differences in between the other treatments, but more leaves were obtained compared to the C and MF treatments. Total soluble solids content was highest in C and all other treatments were in the same group with lower values. There were no significant differences between the measured pH values and they varied between 6.31-6.15.

Microbial fertiliser applications combined with inorganic fertiliser did not negatively affect the growth characteristics of lettuce and even showed an improving effect. These results are consistent with previous studies. For example, Demir et al. (2023) reported similar results in terms of head length, leaf number and TSS of lettuce with the use of a microbial fertiliser in combination with chemical fertilisers. In addition, contrary to this study, it was reported that the combination of microbial and chemical fertilisers was effective on root collar diameter and pH. Miskoska-Milevska et al. (2018) found that

commercial Slavol containing the microorganisms *Azotobacter chroococcum*, *Azotobacter vinelandii*, *Derxia sp.*, *B. megaterium*, *B. licheniformis* ve *B. subtilis* increased leaf length and width when sprayed on cauliflower leaves. On the contrary, Rabiei et al. (2020) reported that *Azospirillum brasilense* and *Azotobacter chroococcum* did not affect the length of coriander whose leaves were consumed. Polat et al. (2004) reported that TSS was in the range of 3.86-4.06% in head lettuce and 4.40-4.60% in cos type lettuce and pH was in the range of 5.99-6.06% in head lettuce and 5.89-5.94% in cos type lettuce. These values are compatible with our study. Özbay et al. (2010) found that *T. harzianum* bacteria increased the TSS of rocket by 13% and cress by 11%.

The effects of microbial fertiliser applications on L\*, C and ho colour values and total chlorophyll (SPAD) content of lettuce are shown in Table 4.

**Table 4.** Effects of microbial fertiliser applications on L\*, C and ho colour values and total chlorophyll of lettuce

Treatments	L*	С	hº	Chlorophyll
С	66.74 a*	43.26 a*	116.25 c*	24.60 de*
100% CF	59.68 bc	40.93 cd	118.95 ab	31.47 a
50% CF + MF	58.39 cd	42.06 abc	119.24 a	26.53 cde
75% CF + MF	56.68 cd	40.61 cd	119.70 a	30.07 ab
100% CF + MF	57.53 cd	41.66 abcd	119.24 a	27.83 bcd
I + CF	59.96 bc	41.28 bcd	119.25 a	30.80 ab
MF	63.68 ab	42.96 ab	117.84 b	23.90 e
50%  CF + MF + FMF	58.08 cd	40.95 cd	118.79 ab	29.50 abc
75%  CF + MF + FMF	55.15 d	40.16 d	119.41 a	31.07 ab
LSD%5	4.4066	1.7997	1.2615	3.3217

<sup>\*</sup>Differences between values not indicated with the same letter are significant at p<0.05 level.

Statistically significant differences were found between treatments in terms of colour criteria and chlorophyll content (p<0.05). The highest L\* value in lettuce leaves was measured in C and MF. The lowest L\* value was determined in 75% CF + MF + FMF, followed by 75% CF + MF, 100% CF + MF, 50% CF + MF + FMF and 50% CF + MF plots. The highest C value indicating leaf colour saturation was obtained in the C and MF, while the lowest values were obtained in the 75% CF + MF + FMF, 75% CF + MF, 100% CF and 50% CF + MF + FMF treatments. The ho angle, which expresses the changes in the colour of the leaves, was highest in 75% CF + MF, 75% CF + MF + FMF, I + CF, 50% CF + MF, 100% CF + MF and 50% CF + MF + FMF treatments. The lowest ho value was determined in C. Green colour is an important morphological trait in lettuce, the leaves of which are consumed as a vegetable. When the colour values obtained were examined from this point of view, it can be said that greener lettuce was harvested in 50% CF + MF, 75% CF + MF, 100% CF + MF, I + CF, 50% CF + MF + FMF and 75% CF + MF + FMF treatments. When the total chlorophyll content was evaluated according to the treatments, the highest values were obtained in the plants in 100% CF, 75% CF + MF + FMF, I +

CF and 75% CF + MF plots. The treatments with the lowest chlorophyll content were in the MF and C plots. It was also determined that the chlorophyll contents were generally in parallel with the colour values.



**Figure 3.** 75% CF + MF + FMF treatment plants that have reached the harvest stage.

Ucok et al. (2019) showed in their research with different organic fertilisers that L\* in curly lettuce varied between 56.23–59.43, C varied between 37.19–38.82, and h° varied between 115.56–117.61. Contrary to these, Sönmez et al. (2017) found that organic fertilisers did not affect these colour values. The colour values determined in our research are consistent with these studies. Demir et al. (2023), who investigated the effects of microbial fertilisers on lettuce, reported quite similar results to our research in terms of the lettuce colour and chlorophyll changes.

The effects of different microbial fertiliser applications on total (g/plant) and marketable (g/plant) yield of lettuce are shown in Table 5.

**Table 5.** Effects of different microbial fertiliser applications on total and marketable yield of curly lettuce.

Treatments	Total yield (g/plant)	Marketable yield (g/plant)
С	138.33 c*	125.00 c*
100% CF	270.00 ab	246.67 ab
50% CF + MF	253.33 b	235.00 b
75% CF + MF	280.00 ab	260.00 ab
100% CF + MF	281.67 ab	266.67 ab
I + CF	296.67 a	276.67 a
MF	146.67 с	140.00 c
50%  CF + MF + FMF	278.33 ab	261.67 ab
75% CF + MF + FMF	286.67 a	261.67 ab
LSD%5	31.622	32.43

<sup>\*</sup>Differences between values not indicated with the same letter are significant at p<0.05 level.

Different treatments significantly affected total and marketable lettuce yield (p<0.05). The highest total yields were found in I + CF (296.67 g/plant), 75% CF + MF + FMF (286.67 g/plant), followed by 100% CF + MF (281.67 g/plant), 75% CF + MF (280.00 g/plant), 50% CF + MF + FMF (278.33 g/plant) and 100% CF (270.00 g/plant) treatments. The lowest yield values were determined in C (138.33 g/plant) and MF (146.67 g/plant) treatments. The highest values in terms of marketable yield were obtained from I + CF (276.67 g/plant), 100% CF + MF (266.67 g/plant), 75% CF + MF + FMF (261.67 g/plant), 50% CF + MF + FMF (261.67 g/plant), 75% CF + MF (260.00 g/plant) and 100% CF (246.67 g/plant) treatments. Similar to the total yield, the lowest marketable yields were again determined in C (125.00 g/plant) and MF (140.00 g/plant) treatments.

When the results were evaluated in terms of yield; the I + CF treatment, which gave the highest value in total yield, provided 114.5% more yield compared to C and provided 102.3% more yield compared to MF. When this application (I + CF) was compared to 100% CF, 9.9% more total yield was obtained. The 75% CF + MF + FMF treatment, provided the highest yield although the chemical fertiliser dose was reduced by 25%; this yield is 107.2% higher than C; 95.5% higher than MF and 6.2% higher than the yield obtained with 100% CF.

There are many studies supporting the view that microbial fertilisers increase the yield and reduce chemical fertiliser consumption. Demir et al. (2023) found the highest total yield in lettuce in the microbial fertiliser, "immersion + chemical fertilisation," and in the "chemical fertilisation (CF) + microbial fertiliser (MF)" applications, while in the same study, the highest first class yield in cucumber was obtained in 50% CF + MF application. When the researchers compared the results obtained, they reported that savings could be achieved on chemical fertilisers by introducing accompanying microbial fertilisation. These results are similar to our research. Vejan et al. (2016) reported that yield was increased between 7% and 33% with the use of PGPR. Stojanović et al. (2020) concluded that some microorganisms were effective on lettuce yield. There are other studies with similar results on saving on chemical fertiliser. For example, Chauhan and Bagyaraj (2015) found that NPK in beans could be reduced by microbial inoculation. Kafi et al. (2021) determined that the recommended NPK dose in cucumber could be reduced by 30% by utilizing some beneficial bacteria.

The positive and negative proportional effects related to the criteria examined within the scope of the research are shown with Pearson correlation coefficients (Table 6). Average values were used in the criteria examined.

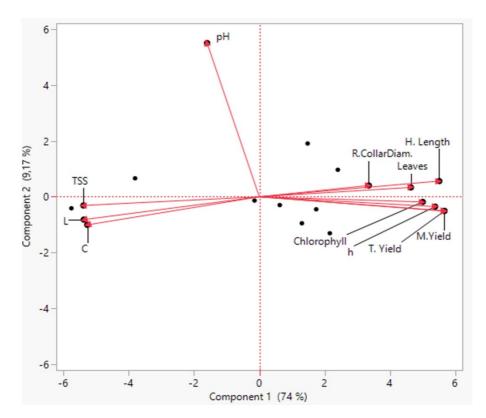
**Table 6.** Pearson correlation coefficients related to the criteria examined in the study.

Parameters	L	С	hº	Chlorophyll	Total Yield	Market. Yield	Head Length	Root Collar Diam.	Leaves Num.	TSS	рН
L	1,000										
С	0,888**	1,000									
hº	-0,935**	-0,817**	1,000								
Chlorophyll	-0,732*	-0,935**	0,722*	1,000							
Total Yield	-0,898**	-0,884**	0,902**	0,877**	1,000						
Market. Yield	-0,897**	-0,867**	0,907**	0,855**	0,998**	1,000					
Head Length	-0,897**	-0,893**	0,904**	0,835**	0,946**	0,953**	1,000				
Root Collar Diam.	-0,585ns	-0,486 <sup>ns</sup>	0,537ns	0,392 <sup>ns</sup>	0,502 <sup>ns</sup>	0,501 <sup>ns</sup>	0,356 <sup>ns</sup>	1,000			
Leaves Num.	-0,609ns	-0,652ns	0,607 <sup>ns</sup>	0,699*	0,807**	0,823**	0,772*	0,525ns	1,000		
TSS	0,904**	0,761*	-0,952**	-0,661 <sup>ns</sup>	-0,897**	-0,916**	-0,919**	-0,571 <sup>ns</sup>	-0,766*	1,000	
рН	0,134 <sup>ns</sup>	0,095 <sup>ns</sup>	-0,320	-0,268 <sup>ns</sup>	-0,351 <sup>ns</sup>	-0,349	-0,160 <sup>ns</sup>	-0,136 <sup>ns</sup>	-0,157 <sup>ns</sup>	0,20 <sup>ns</sup>	1,000

<sup>\*</sup>Significant at p<0.05 level; \*\*Significant at p<0.001 level; ns: Not Significant

There was a strong positive relationship between L and C and TSS among the examined criteria, while there was a strong negative relationship between L and ho, total yield, marketable yield and head length. The negative relationship between L and chlorophyll was moderate. The significant relationships between C and other criteria were negative except for TSS (positive). Among the relationships between ho and other important criteria, only TSS was negative. There was a strong positive relationship between chlorophyll and total yield, marketable yield, head length, and a moderately significant positive relationship with the number of leaves. While there was a strong positive relationship between total yield and marketable yield, head length and number of leaves, there was a strong negative relationship with TSS. The relationships between the examined criteria can be seen in Table 6.

The principal component analysis conducted by considering the criteria examined in the research is shown in Figure 4, and the load matrices are shown in Table 7.



**Figure 4.** Principal component analysis results of the parameters examining the effects of microbial fertiliser applications.

According to the principal component analysis results, Figure 4 shows two components. The first principal component represented 74%, the second principal component 9.17% and the total variation 83.17%. All analysed components and factor loadings can be seen in Table 7.

**Table 7.** Load matrices of the examined criteria

Parameters	Prin1	Prin2	Prin3	Prin4	Prin5
L	-0,93390	-0,14187	-0,05527	0,28158	0,05680
С	-0,91533	-0,17470	0,15446	-0,01020	0,31765
h <sup>o</sup>	0,93532	-0,06099	0,03716	-0,31904	0,05801
Chlorophyll	0,86920	-0,03195	-0,25100	0,23750	-0,33756
Total Yield	0,98398	-0,08892	-0,08128	0,04446	0,02241
Marketable Yield	0,98470	-0,08739	-0,06865	0,04093	0,07833
Head Length	0,95707	0,09793	-0,23209	-0,04192	0,12400
Root Collar Diameter	0,58232	0,07031	0,78301	0,07432	-0,19250
Leaves Number	0,80591	0,05831	0,08860	0,50667	0,28424
TSS	-0,93574	-0,05393	-0,11003	0,15663	-0,27603
рН	-0,27789	0,95777	-0,04517	0,01212	0,03662

The most important traits constituting the first main component were marketable yield, total yield, head length, TSS, ho colour value, L\* and C colour values, chlorophyll and leaf number according to

factor loadings. In the second main component, only the most important factor loading was pH, while the third component was root collar diameter.

#### **Conclusions**

This research was carried out in pots to determine the effects of a microbial fertiliser called Herasim containing different beneficial bacteria in a cocktail form on the growth, quality and yield characteristics of lettuce. The results of the study were significant. First of all, it was determined that Herasim microbial fertiliser had no negative effects on lettuce growth characteristics, quality criteria, leaf colours and yield. It was determined that basically similar results were obtained with 100% CF in terms of the criteria examined, and even some healing effects of microbial fertiliser appeared when the chemical fertiliser dose was reduced. For example, greener lettuce leaves were harvested by applying microbial fertilisation at reduced chemical fertiliser doses.

In addition to different chemical fertiliser doses, higher yield values were obtained with microbial fertilisers applied in comparison to that of control and microbial fertiliser only applications. Despite the reduced NPK doses, it is noteworthy that similar yield results were obtained with 100% CF alone with the addition of microbial fertiliser. In this respect, 75% CF + MF and 75% CF + MF + FMF and 50% CF + MF + FMF trials are important in terms of fertiliser savings. The increase in yield by spraying microbial fertiliser on the leaves was also found to be remarkable.

In the I + 100% CF application, where the microbial fertiliser was applied to lettuce by dipping only before planting, the highest total yield (296.67 g/plant) and marketable yield (276.67 g/plant) were obtained and it was shown that the plant could develop better due to better nutrient uptake by the plant roots at an early stage in this application.

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#### REFERENCES

- Aćamović-Djoković, G., Pavlović, R., Mladenović, J., & Djurić, M. (2011). Vitamin C content of different types of lettuce varieties. *Acta Agric Serb*, *16*(32), 83-89.
- Aksoy, U., Altındişli, A. (1998). Ekolojik (Organik, Biyolojik) Tarım. ETOD, 125s, İzmir.
- Aksoy, U. (1999). Ekolojik tarımdaki gelişmeler. Ekolojik Tarım, Ekolojik Tarım Organizasyonu Derneği, Emre Basımevi, İzmir.
- Antoun, H., & Prévost, D. (2006). Ecology of plant growth promoting rhizobacteria. *PGPR: Biocontrol and biofertilization*, 1-38.

- Awaad, M. S., Badr, R. A., Badr, M. A., & Abd-elrahman, A. H. (2016). Effects of different nitrogen and potassium sources on lettuce (Lactuca sativa L.) yield in a sandy soil. *Eurasian Journal of Soil Science*, 5(4), 299-306.
- Aybak H.Ç. 2002. *Salata/Marul Yetiştiriciliği*. ISBN: 975-8377-20-5, (sayfa sayısı 96), İstanbul: Hasad Yayıncılık (in Turkish).
- Chauhan, H., & Bagyaraj, D. J. (2015). Inoculation with selected microbial consortia not only enhances growth and yield of French bean but also reduces fertilizer application under field condition. *Scientia Horticulturae*, 197, 441-446.
- Chen, Y., Mei, R., Lu, S., Liu, L., & Kloepper, J. W. (1996). The use of yield increasing bacteria (YIB) as plant growth promoting rhizobacteria in Chinese agriculture. In R. S. Uthkede, and W. K. Gupta (Eds.), Management of soil borne diseases (pp. 164–184). Ludhiana: Kalyani Publishers.
- Chen, J. H. (2006, October). The combined use of chemical and organic fertilizers and/or biofertilizer for crop growth and soil fertility. In *International workshop on sustained management of the soil-rhizosphere system for efficient crop production and fertilizer use* (Vol. 16, No. 20, pp. 1-11). Land Development Department Bangkok Thailand.
- Colla, G., Suárez, C. M. C., Cardarelli, M., & Rouphael, Y. (2010). Improving nitrogen use efficiency in melon by grafting. *HortScience*, 45(4), 559-565.
- Colla, G., Rouphael, Y., Mirabelli, C., & Cardarelli, M. (2011). Nitrogen-use efficiency traits of miniwatermelon in response to grafting and nitrogen-fertilization doses. *Journal of Plant Nutrition and Soil Science*, 174(6), 933-941.
- Colonna, E., Rouphael, Y., Barbieri, G., & De Pascale, S. (2016). Nutritional quality of ten leafy vegetables harvested at two light intensities. *Food chemistry*, 199, 702-710.
- Compant, S., Duffy, B., Nowak, J., Clément, C., & Barka, E. A. (2005). Use of plant growth-promoting bacteria for biocontrol of plant diseases: principles, mechanisms of action, and future prospects. *Applied and environmental microbiology*, 71(9), 4951-4959.
- Demir, H., Yalçi, H. K., & Katgici, A. (2023). Ameliorative effects of microbial fertiliser on yield and quality parameters of curly lettuce and cucumber with fertiliser saving. *Folia Horticulturae*, 35(1), 91-106.
- Fálico, L., García, B., Sillon, M., & Visintin, G. (2000). Bacillus sp. and growth promotion in soybean. In *Proceedings 5th International PGPR Workshop* (p. 40).
- Goessler, W., & Kuehnelt, D. (2002). Analytical methods for the determination of arsenic and arsenic compounds in the environment. *Environmental chemistry of arsenic*, 27-50.
- Gravel, V., Antoun, H., & Tweddell, R. J. (2007). Growth stimulation and fruit yield improvement of greenhouse tomato plants by inoculation with Pseudomonas putida or Trichoderma atroviride: possible role of indole acetic acid (IAA). *Soil biology and Biochemistry*, 39(8), 1968-1977.
- Gupta, A., & Sen, S. (2013). Role of biofertilisers and biopesticides for sustainable agriculture, scholar. google. com.
- Haque, M. M., Ilias, G. N. M., & Molla, A. H. (2012). Impact of Trichoderma-enriched biofertilizer on the growth and yield of mustard (Brassica rapa L.) and tomato (Solanum lycopersicon Mill.).

- Kafi, S. A., Arabhosseini, S., Karimi, E., Koobaz, P., Mohammadi, A., & Sadeghi, A. (2021). Pseudomonas putida P3-57 induces cucumber (Cucumis sativus L.) defense responses and improves fruit quality characteristics under commercial greenhouse conditions. *Scientia Horticulturae*, 280, 109942.
- Yamada, K., & Xu, H. L. (2001). Properties and applications of an organic fertilizer inoculated with effective microorganisms. *Journal of Crop production*, *3*(1), 255-268.
- Keshavarz Zarjani, J., Aliasgharzad, N., Oustan, S., Emadi, M., & Ahmadi, A. (2013). Isolation and characterization of potassium solubilizing bacteria in some Iranian soils. *Archives of Agronomy and Soil Science*, 59(12), 1713-1723.
- Korir, H., Mungai, N. W., Thuita, M., Hamba, Y., & Masso, C. (2017). Co-inoculation effect of rhizobia and plant growth promoting rhizobacteria on common bean growth in a low phosphorus soil. *Frontiers in plant science*, *8*, 141.
- Lévai, L., Veres, S. Z., Makleit, P., Marozsán, M., & Szabó, B. (2006, February). New trends in plant nutrition. In *Proceedings of 41st Croatian and 1st International Symposium on Agriculture* (pp. 435-436).
- Luz, W. C. (2000, October). Plant growth promoting rhizobacteria in graminicolous crops in Brazil. In *Fifth International PGPR Workshop* (Vol. 29).
- Madeira, A.C., Ferreira, A., de Varennes, A., and Vieira, M.I. (2003). SPAD Meter Versus Tristimulus Colorimeter to Estimate Chlorophyll Content and Leaf Colour in Sweet Pepper. *Communications in Soil Science and Plant Analysis*, 34(17-18), 2461-2470.
- Miskoska-Milevska, E., Najdenovska, O., Popovski, Z., & Dimovska, D. (2018). The influence of the microbiological fertilizer–Slavol on cauliflower growth. *Romanian Biotechnological Letters*, 2(2), 13511-13516.
- Molla, A. H., Manjurul Haque, M., Amdadul Haque, M., & Ilias, G. N. M. (2012). Trichoderma-enriched biofertilizer enhances production and nutritional quality of tomato (Lycopersicon esculentum Mill.) and minimizes NPK fertilizer use. *Agricultural Research*, 1, 265-272.
- Muraleedharan, H., Seshadri, S., & Perumal, K. (2010). Biofertilizer (phosphobacteria). *Shri AMM Murugappa Chettiar Research Centre. Taramani, Chennal-600113*.
- Özbay, N., Emrebas, N., and Akinci, S. (2010). Effects of a microbial fertilizer (Trichoderma hertzian, Kuen 1585) on growth and yield of arugula (Eruca vesicaria subsp. sativa) and garden cress (Lepidium sativum) grown in soilless culture. The Journal of Agricultural Faculty of Ege University, Special Issue, 268–274, ISSN 1018-8851.
- Pal, K. K., Dey, R., Bhatt, D. M., & Chauhan, S. M. (2000, October). Plant growth promoting fluorescent pseudomonads enhanced peanut growth, yield and nutrient uptake. In *Proceedings of the Fifth International PGPR Workshop* (Vol. 29).
- Sun, P. F., Fang, W. T., Shin, L. Y., Wei, J. Y., Fu, S. F., & Chou, J. Y. (2014). Indole-3-acetic acid-producing yeasts in the phyllosphere of the carnivorous plant Drosera indica L. *PloS one*, *9*(12), e114196.
- Parr, J. F., Hornick, S. B., & Kaufman, D. D. (1994). Use of microbial inoculants and organic fertilizers in agricultural production.
- Pieterse, C. M., Zamioudis, C., Berendsen, R. L., Weller, D. M., Van Wees, S. C., & Bakker, P. A. (2014). Induced systemic resistance by beneficial microbes. *Annual review of phytopathology*, 52(1), 347-375.

- Polat, E., Onus, A. N., and Demir, H. (2004). Atik Mantar Kompostunun Marul Yetiştiriciliğinde Verim ve Kaliteye Etkisi. *Akdeniz University Journal of the Faculty of Agriculture*, *17*(2), 149-154.
- Prasanna, R., Sood, A., Ratha, S. K., & K. Singh, P. (2014). Cyanobacteria as a "green" option for sustainable agriculture. *Cyanobacteria: an economic perspective*, 145-166.
- Rabiei, Z., Hosseini, S. J., Pirdashti, H., & Hazrati, S. (2020). Physiological and biochemical traits in coriander affected by plant growth-promoting rhizobacteria under salt stress. *Heliyon*, 6(10).
- Richardson, A. E., Barea, J. M., McNeill, A. M., & Prigent-Combaret, C. (2009). Acquisition of phosphorus and nitrogen in the rhizosphere and plant growth promotion by microorganisms.
- Sindhu, S. S., Parmar, P., Phour, M., & Sehrawat, A. (2016). Potassium-solubilizing microorganisms (KSMs) and its effect on plant growth improvement. *Potassium solubilizing microorganisms for sustainable agriculture*, 171-185.
- Singh, D., Chand, S., & Anvar, M. (2003). Effect of organic and inorganic amendment on growth and nutrient accumulation by isabgol (Plantago ovata) in sodic soil under greenhouse conditions.
- Siomos, A. S., Papadopoulou, P. P., Niklis, N. D., & Dogras, C. C. (2000, October). Quality of romaine and leaf lettuce at harvest and during storage. In *II Balkan Symposium on Vegetables and Potatoes 579* (pp. 641-646).
- Sönmez, İ., Kalkan, H., Demir, H., Külcü, R., Yaldiz, O., & Kaplan, M. (2017). Mineral composition and quality parameters of greenhouse-grown lettuce (Lactuca sativa L.) depending on fertilization with agricultural waste composts. *Acta Scientiarum Polonorum Hortorum Cultus*, 16(3), 85-95.
- Stojanović, M., Radović, I., Zuza Prastalo, M., Jovanovic, Z., Moravcevic, D., Cvijanović, G., & Savic, S. (2020). The productivity and quality of Lactuca sativa as influenced by microbiological fertilisers and seasonal conditions. *Zemdirbyste-Agriculture*, 107(4), 345-352.
- Triveni, S., Prasanna, R., Kumar, A., Bidyarani, N., Singh, R., & Saxena, A. K. (2015). Evaluating the promise of Trichoderma and Anabaena based biofilms as multifunctional agents in Macrophomina phaseolina-infected cotton crop. *Biocontrol Science and Technology*, 25(6), 656-670.
- TÜİK 2024. Bitkisel Üretim İstatistikleri, 2023. https://data.tuik.gov.tr/Bulten/Index?p=Bitkisel-Uretim-Istatistikleri-2023-49535.
- Uçok, Z., Demir, H., Sönmez, İ., & Polat, E. (2019). Farklı organik gübre uygulamalarının kıvırcık salatada (Lactuca sativa L. var. crispa) verim, kalite ve bitki besin elementi içeriklerine etkileri. *Mediterranean Agricultural Sciences*, 32, 63-68.
- Vejan, P., Abdullah, R., Khadiran, T., Ismail, S., & Nasrulhaq Boyce, A. (2016). Role of plant growth promoting rhizobacteria in agricultural sustainability—a review. *Molecules*, 21(5), 573.
- Vessey, J. K. (2003). Plant growth promoting rhizobacteria as biofertilizers. Plant and soil, 255, 571-586.
- Wall, L. G. (2000, October). Consequences of an overview on PGPR work in Argentina: The field should be wider. In *Fifth International PGPR Workshop* (Vol. 29).
- Wang, G., Ren, Y., Bai, X., Su, Y., & Han, J. (2022). Contributions of beneficial microorganisms in soil remediation and quality improvement of medicinal plants. *Plants*, 11(23), 3200.
- Wu, F., Wan, J. H. C., Wu, S., & Wong, M. (2012). Effects of earthworms and plant growth–promoting rhizobacteria (PGPR) on availability of nitrogen, phosphorus, and potassium in soil. *Journal of Plant Nutrition and Soil Science*, 175(3), 423-433.

- Zhao, Y., Zhang, M., Yang, W., Di, H. J., Ma, L., Liu, W., & Li, B. (2019). Effects of microbial inoculants on phosphorus and potassium availability, bacterial community composition, and chili pepper growth in a calcareous soil: a greenhouse study. *Journal of Soils and Sediments*, 19, 3597-3607.
- Zhao, Y., Mao, X., Zhang, M., Yang, W., Di, H. J., Ma, L., ... & Li, B. (2021). The application of Bacillus Megaterium alters soil microbial community composition, bioavailability of soil phosphorus and potassium, and cucumber growth in the plastic shed system of North China. *Agriculture, Ecosystems & Environment*, 307, 107236.
- Zhou, C., Ma, Z., Zhu, L., Xiao, X., Xie, Y., Zhu, J., & Wang, J. (2016). Rhizobacterial strain Bacillus megaterium BOFC15 induces cellular polyamine changes that improve plant growth and drought resistance. *International Journal of Molecular Sciences*, 17(6), 976.
- Zou, C., Li, Z., & Yu, D. (2010). Bacillus megaterium strain XTBG34 promotes plant growth by producing 2-pentylfuran. *The Journal of Microbiology*, 48, 460-466.