

Article ID: 201391
DOI: 10.5586/aa/201391

Publication History

Received: 2024-07-19
Accepted: 2025-02-13
Published: 2025-06-26

Handling Editor

Alina Wiszniewska; University
of Agriculture in Krakow, Poland

Authors' Contributions

HM, IJ: Research concept and
design; SS: Collection and/or
assembly of data; HS, HM, SS:
Data analysis and
interpretation; HS: Writing the
article; HS, NS, IJ: Critical
revision of the article; NS: Final
approval of the article

Funding

This research was funded by
the Lebanese University in the
frame of a research project.

Competing Interests

No competing interests have
been declared.

Copyright Notice

© The Author(s) 2025. This is an
open access article distributed
under the terms of the [Creative
Commons Attribution License](#),
which permits redistribution,
commercial and
noncommercial, provided that
the article is properly cited.

ORIGINAL RESEARCH

Effect of mineral fertilization and native microbial preparations on productivity of Borlotti beans (*Phaseolus vulgaris*)

Hala Samaha^{1*}, Nidal Shaban², Sleiman Skaf³, Ihab Jomaa³,
Hassane Makhoul¹

¹ Earth and Life Sciences Department, L2GE, Lebanese University, Lebanon

² Agronomy Department, University of Forestry, Bulgaria

³ Lebanese Agricultural Research Institute, Lebanon

*To whom correspondence should be addressed. Email: halasamaha@hotmail.com

Abstract

In order to fully realize the potential of beneficial microorganisms in the agricultural system, there remains a huge research gap in understanding the factors shaping their functional activities in the rhizosphere. Recently, native microorganisms retrieved from healthy soils have shown a thriving effect in plant growth stimulation. This study aimed to compare the outcome of two commercially available native microbial products (namely LBM) on the growth and yield of *Phaseolus vulgaris* in an open-field experiment. The research hypothesis suggested that microbial inoculants could increase the efficiency of water use in beans, thereby leading to seed yield augmentation in the total absence of chemical fertilization. The results of the study indicated that plant growth and productivity parameters were clearly affected by the LBM application. Moreover, the organic fertilizer (OF) enhanced the effectiveness of microbial consortia significantly by increasing leaf area (89%), above ground biomass (56%), pod number per plant (50%), pod weight (73%), and seed mass (78%), compared to the control. This study highlights a rise in WUE by 44–54% subsequently to microbial treatments versus 6.7% recorded with the mineral fertilizer in normal irrigation conditions. The hypothesis of this study was positively confirmed by a high correlation between WUE and seed mass per plant ($r = 0.84$) or 1000-seed weight ($r = 0.61$). In conclusion, the co-incorporation of native microbial inoculants and organic amendments can help to meet the requirements of beans crops, thus allowing the replacement of mineral fertilizers while safeguarding the yields and quality of the final product (pods and seeds).

Keywords

microbial product; plant growth; beans; field trial; biofertilizer; beneficial microorganisms

1. Introduction

Common bean (*Phaseolus vulgaris* sp.) is one of the most popular crop legumes worldwide (Gonzalez & Paredes-Lopez, 2009). Besides its antioxidant activities, bean grains are packed with proteins, fibers, essential vitamins, and minerals such as potassium, vitamin B6, and folic acid (Cardador-Martínez, et al., 2002; Garden-Robinson & McNeal, 2013). Beans are considered the main staple food, especially in the Mediterranean countries, owing to their valuable abundance of plant-based proteins.

Recently, the increasing concerns for food security, particularly in Lebanon, have intensified the need for crop production with low management costs and higher yields, combined with a wide-ranging nutritional value. Hence, the common bean crop emerged with a huge potential and its production is boosted in most regions of the country. However, the cultivation of crop vegetables is one of the most demanding cropping systems

in terms of agrochemicals and pesticides (Zhao et al, 2005). Nowadays, the expansion of crop production in Lebanon relies basically on the over-consumption of chemical fertilizers (El-Alam et al., 2018) and high exploitation of agricultural lands. This mismanagement of fertilizer application can be harmful to the nature through contamination of water resources (Nehme et al., 2014) and soil salinization and to biodiversity by the appearance of resistant pathogens destabilizing the soil microbiota (Shukla et al, 2019; Thakur et al., 2020). A new relevant challenge related to water scarcity for irrigation has emerged (Condon, 2020), particularly in Mediterranean conditions (hot, dry summer and cool, wet winter), highlighting the need for further investigation towards innovative solutions. Given the global environmental variation and challenges, agricultural practices must be evolving to an eco-friendly and sustainable system. Therefore, an efficient use of existing land and available water, animal and green manures, off-farm organic wastes, and the use of bio-fertilization to maintain soil productivity are promoted.

Nowadays, microbial biofertilizers are receiving particular interest and are considered the best alternative to chemical fertilizers (Yadav et al, 2023). This may be attributed to the fact that most microbial products applied to crops stimulate plant growth and productivity. Microbial preparations may include a large variety of microorganisms, such as plant growth-promoting rhizobacteria (PGPR), mycorrhizal and non-mycorrhizal fungi, and bacterial endosymbionts, all of which adopt diverse mechanisms and strategies to promote plant development in normal conditions and enhance tolerance to biotic and abiotic stress (Ali et al., 2022).

In a previous study, a mixture of native microbial inoculants was identified, exerting stimulatory effects on vegetative growth and yield production in tomato and pepper (Makhlouf et al, 2023). The type of product used, called LBM, is a solution based on naturally assembled multi strain species which appeared effective in the total absence of chemical fertilizers and revealed higher outcomes when combined with organic compounds. Exploiting the efficacy of the combined microbial consortia-organic fertilizer is very promising in the development of plant biofertilizers. Although microbial products can provide an option to safeguard sustainable crop yields, farmers would not be able to adopt biofertilizers so easily. One of the major problems with the use of biofertilizers is that, in almost all cases, biofertilizers have been tested in greenhouse conditions, thus leading to different results when applied in field conditions. The beneficial effect of microbial inoculants on plants is known to be dependent on several factors, such as soil type and plant species (Ortiz & Sansinenea, 2022). Moreover, the literature does not provide adequate information since it is a new practice in the traditional agricultural system, especially in Lebanon. Therefore, there is a potential pathway to explore the feasibility of microbial biofertilizers applied on a large scale, mainly on the productivity of high-nutritive crop species.

In this regard, this study was performed to (1) investigate the effect of two commercially available LBM products on the productivity of Borlotti beans in an agro-ecological zone where beans are typically grown on farms, (2) explore an alternative fertilization strategy for beans under challenging environmental factors, and (3) have more insights into the mode of action of microbial biofertilizers related to their ability to ameliorate the water use by plants. The hypothesis was that the use of microbial consortia combined with organic compounds will improve plant nutrient acquisition, leading to productivity enhancement and amelioration of the outcome quality, thus allowing the replacement of mineral fertilization.

2. Material and methods

2.1. Trial establishment

The field experiment was conducted during the growing season of summer-autumn 2019 in the Lebanese Agricultural Research Institute, in Tal Amara station, located in central Bekaa, Lebanon. The average altitude of the site is 943 m above sea level, latitude 33°51'23" N and longitude 35°59'5" W. The climate in this region is typically Mediterranean, characterized by hot and dry summer (from June to October) and rainy winter. Long-run data indicate an average seasonal rain of 592 mm, with 95% of the rain occurring between November and March.

Long-run data indicate an average seasonal rain of 592 mm, with 95% of the rain occurring between November and March. The soil of the study field is clayey (60%), alkaline (pH 7.4) with low-organic content (0.07%); available macroelements include (mg kg^{-1} soil) phosphorus – 40, potassium – 311, and magnesium – 511.

The Borlotti bean cultivar (Dwarf variety) available in the market was used in this experiment. This variety is medium-early, very productive with a growing season of 75–80 days. Seeds of Borlotti beans (*Phaseolus vulgaris* L.) were sown on 14 August in the soil at a density of 24 plants per m^2 (8 sowing holes at 3 seeds per hole).

A drip irrigation system was adopted using JR Pipes (16 mm – Dripper spacing: 25 cm – Dripper Flow rate: 4 L/h) with flowmeters set in the system. The water amount requirement was determined based on the volumetric soil water content using Decagon sensors at 20 cm soil depth (Field capacity considered $F_c = 32\%$). The mineral fertilizer and microbial products were applied separately using Dosatron and Venturi Pumps.

2.2. Experimental design and treatments

The experimental design adopted in this study was the Randomized Completely Block Design (RCBD) with four replicates. The experimental plot had an area of 50 m^2 ($10 \times 5 \text{ m}$), consisting of 6 rows at a space of 50 cm between two rows. The total area of the experimental field was 1196 m^2 .

The experimental design including the different treatments is indicated in Table 1. The microbial inoculation was carried out using different products available commercially (NESCO, National Environmental Solution Company, Lebanon) and referred to as LBM, Lebanese beneficial microorganisms, (Makhlouf et al., 2023). In this field experiment, two microbial products (SR, NFPC) were used in addition to an organic fertilizer:

- SR (Soil Regenerator) with declared active ingredients: lactic acid bacteria, acetic acid bacteria, *Cyanobacteria*, phototrophic bacteria, symbiotic nitrogen-fixing bacteria, *Saccharomyces*, enzymes, organic acids, and minerals.
- NFPC (Nitrogen Fixator and Pest Controller): lactic acid bacteria, nitrogen-fixing bacteria, acetic acid bacteria, phototrophic bacteria, yeast, alcohol, enzymes, and organic metabolites.

Organic fertilizer (OF) based on chicken manure with 4% nitrogen, 3% phosphorus, and 3% potassium (quantified using the Kjeldahl method for N, the Oslen method for P, and flame photometry for K). The microbial products were used in the trial starting from 21 days after sowing (DAS) and were diluted following the supplier's instructions. SR was applied one time per week (0.4 L/m^2) with the irrigation water. 2.5 ml of NFPC were diluted in one liter of water; afterwards, using 400 ml sprayers, the diluted NFPC solution was applied on leaves at 10-day intervals. OF were spread on the soil two times (at 30 and 56 DAS) at the dosage of 0.24 kg/m^2 . The mineral fertilizer was applied once per week according to the recommended protocols for beans cultivation starting on 21 DAS. Phosphorus fertilization in the form of ammophos (nitrogen content 12%, phosphorus 61%) was applied at the dosage of $125 \text{ kg ha}^{-1} \text{ P}_2\text{O}_5$, and potassium fertilization in the form of potassium nitrate (potassium content 46%) was applied at dose of $350 \text{ kg ha}^{-1} \text{ K}_2\text{O}$. Nitrogen fertilizers in the form of urea (46%) were applied at 20 kg ha^{-1} .

Table 1 Experimental design.

Treatment	Product used
T1	Control – without application of fertilization
T2	Soil application of SR + foliar application of NFPC
T3	Soil application of SR + foliar application of NFPC + OF
T4	NPK (mineral fertilizer)

2.3. Harvest, biomass, and productivity

To estimate the leaf cover area, many pictures were taken at 70 DAS in each plot related to the treatments. The images were analyzed by the canopeo tool measuring the fractional green canopy cover (FGCC) as described by Patrignani & Ochsner (2015), and expressed in percentage.

The harvest of green pods took place at marketable maturity at 75 DAS for all treatments. The inner area of each plot was reserved to harvest sampling, while the remaining plot area was used for growth sampling. Nine plants were taken randomly from each plot at 2-day-intervals, grouped according to their treatment, and weighed to measure the biomass. The pod number, seed number, and 1.000-seed weight were determined. After determination of the mass of seeds for each treatment, their yield was obtained by extrapolation to productivity kg ha^{-1} . The pods and seeds were counted by plant, and the samples were oven-dried at 75°C for 48 h.

To analyze the pod quality, pods from 1 m^2 quadrates in three replicates were sampled randomly from each treatment using the procedure given in Foroud et al. (1993). The pods were categorized depending on their size and the number of seeds inside. Then, the parameters related to the pod number and the number of seeds per pod were determined and weighed in each category.

Water use efficiency representing the amount of biomass produced per unit of water used by a plant was calculated for the seed yield. The values were determined as a ratio of fresh seed yield per hectare to the total amount of water monitored by sensors during the experiment and were expressed in units of $\text{kg ha}^{-1} \text{ mm}^{-1}$.

2.4. Statistical analysis

The data were subjected to a one-factor analysis of variance ANOVA using the SAS package (SAS 2002) to evaluate the effect of the treatments on the studied parameters. The significance of differences between evaluated mean values was determined by the Tukey test at a significance level of 0.05.

3. Results

3.1. Growth parameters and water use efficiency

The results obtained from the leaf cover area analysis (Table 2, Figure 1) in the different treatments indicate that T2, T3, and T4 differed significantly from the control with no added fertilizer. The bean leaf area increased depending on the microbial preparation applied, from 22.2% in the control to 28.7% in T2 and 42.2% in T3. Overall, T3 produced the highest mean value of leaf covering, followed by T4 (mineral fertilizer) and T2, respectively. In parallel, a significant increase in the fresh mass of the aboveground part of plants was recorded in T3, about 56% higher than the control, whereas T2 and T4 did not show a significant effect on the value of fresh biomass. Likewise, a remarkable extension of the vegetative growth was detectable in

Table 2 Effect of treatments on *P. vulgaris* leaf area, biomass and water use efficiency.

Treatments	Leaf cover area (%)	Aboveground biomass (g.plant^{-1})	WUEi (kg. mm^{-1})
T1 (control)	22.25 ± 4.21^a	61.49 ± 9.35^a	5.97 ± 0.61^a
T2 (SR + NFPC)	28.75 ± 1.52^b	59.11 ± 7.42^a	9.25 ± 2.07^c
T3 (SR + NFPC+ OF)	42.25 ± 1.70^c	96.52 ± 8.23^b	8.62 ± 1.15^{bc}
T4 (NPK)	37.00 ± 3.05^d	71.31 ± 2.42^{aa}	6.37 ± 0.94^{ab}

The readings were expressed as mean $\pm$ SD ($n = 36$). In each column, values with the same letter are not significantly different (Tukey test, $p \geq 0.05$) when compared to control. WUE: water use efficiency. SR: soil regenerator, NFPC: nitrogen fixator and pests controller, OF: organic fertilizer, NPK: mineral fertilizer.



Figure 1 Effect of microbial treatments and mineral fertilizer on *P. vulgaris* plants. Pictures related to plots of beans plants were taken at 60 days after sowing. T1: control, T2: SR+NFPC, T3: SR+NFPC+OF, T4: NPK. SR: soil regenerator, NFPC: nitrogen fixator and pests controller, OF: organic fertilizer, NPK: mineral fertilizer.

T3 combined with a clear intensification in the color of leaves, as evidenced in the pictures of the Borlotti plants, compared to the other treatments (Figure 1).

Irrigation-related water use efficiency averaged $5.97 \text{ kg} \cdot \text{mm}^{-1}$ in the control, while the WUE of microbial treatments T2 and T3 were 54 and 44% higher, respectively. However, the chemical fertilizer (T4) had a WUE value of 6.7% higher than the control.

3.2. Pod yield and productivity

The microbial treatments and mineral fertilization had a significant positive effect on yield and seed productivity (Table 3). The highest values of yield, compared to the control, were noted in microbial treatment T3, reflected by an increase in the pod number and weight as well as seed production. The average number of pods per plant increased in the applied microbial product treatment by about 16% in T2 and 50% in

Table 3 Influence of chemical fertilizer and microbial preparation on *P. vulgaris* productivity.

Treatments	Pod number (nb.plant ⁻¹)	Pod weight (g.plant ⁻¹)	Seed mass (g.plant ⁻¹)	1000-seed weight (g)	Seed yield (t ha ⁻¹)
T1 (control)	6.87 ± 1.11^a	26.43 ± 4.4^a	9.64 ± 2.1^a	914.4 ± 41^a	1.87 ± 0.6^a
T2 (SR + NFPC)	8.00 ± 0.94^{ab}	35.6 ± 2.41^{ab}	13.88 ± 2.2^{ab}	1082.2 ± 88^b	3.00 ± 0.5^b
T3 (SR + NFPC + OF)	10.37 ± 1.55^c	45.89 ± 4.8^b	17.25 ± 3.2^b	1072.9 ± 93^b	4.12 ± 0.3^c
T4 (NPK)	9.50 ± 1.38^{cb}	37.29 ± 1.9^{ab}	12.74 ± 2.5^{ab}	972.6 ± 44^a	2.78 ± 0.6^b

The results were expressed as mean $\pm$ SD ($n = 36$). Values in each column followed by the same letter are not significantly different (Tukey test, $p \geq 0.05$) when compared to control. SR: soil regenerator, NFPC: nitrogen fixator and pests controller, OF: organic fertilizer, NPK: mineral fertilizer.

T3 versus an increase of 38% induced by the mineral fertilizer (T4). Similarly, the microbial product increased the mean pod weight per plant. For this parameter, T3 had the greatest effect with a 73% increase in the pod weight per plant, compared to the control, whereas T2 and T4 displayed comparable results with a 34 and 41% increase, respectively.

Following the pod weight augmentation due to the fertilizer and microbial treatments, the increase in productivity could be expected. As can be seen from the data related to this parameter, the microbial treatment induced a significant increase in the mass of seeds per plant by about 43 and 78% in T2 and T3, respectively, while the mineral fertilizer increased the value of this parameter by 32%. However, in terms of the weight of 1000 seeds of beans, both microbial applications showed similar results and exceeded the values obtained with the mineral fertilizer. The improvement of plant growth parameters under the microbial treatments was finally reflected by significant augmentation in total seed yield per hectare. The highest seed yield was obtained in T3 with an average of 4.12 t ha^{-1} , whereas T2 and T4 scored 3 and 2.78 t ha^{-1} , respectively.

3.3. Characterization of pods

The growth characteristics of pods and seeds were compared between untreated plants and those treated with the microbial product or the mineral fertilizer. The harvested pods were classified in two categories (Table 4): category 1 included bigger pods showing more than three beans per pod, while category 2 consisted of short pods with less than three beans in each. It is notable that the total seed number per m^2 was positively affected by the application of the microbial inoculants, thus higher values were obtained, compared to the control. The maximum increase in the total seed number was attained by T2 (79%) and T3 (84%) followed by the chemical fertilizer T4 (32%). This significant rise in the total seed number per m^2 was attributed to the higher number and weight of large pods (category 1) as well as the number of seeds in each. For this latter category, the highest values were registered in T2 (146 pods and 295 seeds per m^2) and T3 (140 pods and 301 seeds per m^2), compared to the control, whereas 114 pods and 187 seeds per m^2 were scored in the variant with the chemical fertilizer. Similarly, in category 1, T2 and T3 exhibited higher pod weight scoring 1108.6 and 1067.8 g.m^{-2} , respectively, differing statistically from the mineral fertilizer treatment (T4) and the control.

On the contrary, the category of small pods (category 2) did not show any significant difference between the treatments (T2, T3 and T4) in the pod number and number of seeds per pod, although the lowest values were obtained in the control. In this category of small pods, only the pod weight was significantly increased in both microbial treatments, compared to the control, and the chemical fertilizer, counting 1186 g.m^{-2} equally for T2 and T3.

Table 4 Categories of pods related to *P. vulgaris* seeds number recorded in each treatment.

Treatments	Category 1			Category 2			Total seeds number (nb.m ⁻²)
	Pod number (nb.m ⁻²)	Pod weight (g.m ⁻²)	Seed number (nb.m ⁻²)	Pod number (nb.m ⁻²)	Pod weight (g.m ⁻²)	Seed number (nb.m ⁻²)	
T1 (control)	81.6 ± 12 ^a	526.3 ± 20 ^a	140.0 ± 15 ^a	167.0 ± 22 ^a	636.3 ± 10 ^a	68.0 ± 16 ^a	208.0 ± 19 ^a
T2 (SR + NFPC)	146.3 ± 18 ^b	1108.6 ± 59 ^b	295.0 ± 33 ^b	204.0 ± 41 ^a	1186.9 ± 39 ^b	79.0 ± 14 ^a	374.0 ± 32 ^{ab}
T3 (SR + NFPC+ OF)	140.6 ± 9 ^b	1067.8 ± 84 ^b	301.6 ± 42 ^b	211.3 ± 32 ^a	1186.1 ± 18 ^b	82.0 ± 7 ^a	383.6 ± 56 ^b
T4 (NPK)	114.3 ± 7 ^{ab}	690.2 ± 75 ^a	187.6 ± 20 ^{ab}	224.3 ± 39 ^a	871.3 ± 57 ^{ab}	88.0 ± 10 ^a	276.0 ± 42 ^{ba}

The results were expressed as mean ± SD ($n = 24$). Values in each column followed by the same letter are not significantly different (Tukey test, $p \geq 0.05$) when compared to control. Category 1: pods containing ≥ 4 beans. Category 2: pods having ≤ 3 beans. SR: soil regenerator, NFPC: nitrogen fixator and pests controller, OF: organic fertilizer, NPK: mineral fertilizer.

3.4. Correlation relationships between yield components, morphological characteristics of plants, and water use efficiency

The results of the correlation between the yield elements and the morphological features of Borlotti plants are represented in Table 5. The crop yield is basically dependent on plant traits that improve carbon gain, such as greater biomass and leaf area, under the influence of environmental factors. Our results show a strong positive correlation between the seed yield and parameters of vegetative growth, such as above-ground biomass ($r = 0.66$) and leaf cover area ($r = 0.70$) in addition to pod weight ($r = 0.71$).

Table 5 Relationships between yields, morphological elements and water use efficiency.

Parameters	PN	SM	TSW	AB	LCA	WUE	PW	SY
PN	1.000	0.198	0.115	0.412	0.821 ***	0.077	0.636 **	0.520 *
SM		1.000	0.683 ***	0.616 **	0.255	0.844 ***	0.716 ***	0.444
TSW			1.000	0.368	0.293	0.617 **	0.601 **	0.489 *
AB				1.000	0.539 *	0.385	0.766 ***	0.667 **
LCA					1.000	0.151	0.572 *	0.701 **
WUE						1.000	0.446	0.348
PW							1.000	0.713 ***
SY								1.000

PN: pods number per plant, SM: seeds mass (g.plant^{-1}), TSW: 1000-seeds weight (g), AB: Aboveground biomass (g.plant^{-1}), LCA: leaf cover area (%), WUE: water use efficiency (kg.mm^{-1}), PW: pods weight (g.plant^{-1}), SY: seeds yield (t.ha^{-1}). Significance levels are represented as *** $p \leq 0.001$, ** $p \leq 0.01$, * $p \leq 0.05$.

A moderate correlation of the seed yield with the pod number ($r = 0.52$) and the 1000-seed weight ($r = 0.48$) was observed in this study. There were also high significant correlations of the pod weight with the pod number ($r = 0.63$), the aboveground biomass ($r = 0.76$), the seed mass ($r = 0.71$), and the 1000-seed weight ($r = 0.60$), while a moderate correlation was observed with the leaf canopy cover ($r = 0.57$).

This study also found high correlations between the water use efficiency and the seed mass per plant ($r = 0.84$) and the 1000-seed weight ($r = 0.61$).

4. Discussion

In recent years, considerable attention has been focused on the potential of microbial inoculums in promoting plant growth and productivity in normal and stressful conditions (Ali et al., 2022). To this point, specific microbial strains have been considered in many studies, used singly or even combined with other strains and/or NPK fertilizers (Yeremko et al., 2024; Marques et al., 2022). Although some expected outcomes are being generated, many inconsistent results have been obtained, as the microbial stimulant effect might differ between crop species and in different environmental conditions (Rouphael et al., 2015; Kunicki et al., 2010). Microorganisms found in the rhizosphere offer a valuable resource for microbial biostimulants. Exploiting such natural strain combinations to improve plant biomass and yield has been confirmed using tomato and green pepper plants (Makhlouf et al., 2023). The above-mentioned strains, so-called LBM, hold an agricultural prospective, since the mixture maintains the natural interactions between microorganisms assisting their establishment in the inoculated soil, thereby leading to greater biomass and yield. Despite the successful contribution of LBM in promoting plant health and productivity, the underlying mechanism that impacts their ability to increase nutrient uptake is still unclear. Furthermore, there remains a major research gap in establishing a standardized protocol for economically important crops, principally in the Mediterranean regions. Globally,

it still the main challenge faced by the vast majority of microbial biostimulants (Yadav et al, 2023; Ortiz & Sansinenea, 2022).

In this study, a positive effect of LBM treatments on *Phaseolus vulgaris* growth was reported. The microbial consortia contributed to a significant increase in plant biomass (for T3) and the leaf cover area in T2 and T3 (Table 2). For both factors, the outcome was amplified when organic compounds were incorporated (T3). Our results are consistent with those obtained in tomato and green pepper where a significant increase in the shoot length and the number of leaves were registered consequently to LBM treatments (Makhlouf et al, 2023). A similar effect was also confirmed in other studies where the application of *Trichoderma herzianum* ALL42 was associated with higher shoot biomass in *P. vulgaris* (Pereira et al., 2014) and spinach (Ozbay et al., 2018). In the study of Yeremko et al. (2024), the inoculum with microbial preparations had a positive effect on the formation of pea leaf area. Furthermore, our results indicated that LBM consortia had a great impact on plant vigor, reflected by an intensification of pigmentation producing dark green leaves, compared to the control, which indicates an enhancement in the photosynthetic activity. Likewise, other authors demonstrated a significant change in the chlorophyll content of *P. vulgaris* leaves after treatment with a microbial inoculum (Marques et. al., 2022). Taken together, the expansion of leaf area and boost in photosynthetic efficiency reported in this study could be explained by an adequate availability of nutrients to the plants, which stimulate the growth process. Minerals such as nitrogen and phosphorus can stimulate the root system activity and production of leaves (Choudhary et al, 2008; Yeremko et al, 2024). It is known that common bean has low tolerance to low soil fertility (Singh et al, 2003), and its productivity relies on external N supply due to its poor capacity of fixing atmospheric nitrogen (Martínez-Romero, 2003). In this study, the amelioration of beans growth associated with the microbial incorporation in the absence of mineral supply (NPK) indicates that the microbial strains had a beneficial effect on soil fertility. As a function of higher microbial activity in inoculated soils, a rise in the availability of organic compounds occurs and can benefit plant nutrient solubility and uptake (Rembialkowska, 2007; Adediran et al., 2007). Moreover, organic amendments have been shown as effective means of improving water retention capacity in semiarid regions (Zhao et al, 2018; Li et al, 2019). In this regard, it is worth highlighting the significant rise in WUE by 44–54% subsequently to the microbial LBM treatments, whereas 6.7% amelioration only was recorded in the NPK fertilizer treatment (Table 2). This could be due to the amelioration of the soil structure (by increasing macropores) after the microbial incorporation, improving water potential and enhancing water and nutrient absorption by plants (Naseem and Bano, 2014). Consequently, a vigorous leaf system can be developed, allowing a substantial improvement of plant yield by assisting seed sets and grain filling (Gao & Lynch, 2016). In agreement with this hypothesis, our results showed a significant positive correlation between the leaf covering surface and seed yield ($r = 0.7$) or the number of pods per plant ($r = 0.82$) as well as between biomass and seed yield ($r = 0.66$) or seed mass ($r = 0.61$). The tight relationship between the yield and the structural components of bean plants was also confirmed in other studies (Shaban, 2021). On the other hand, a strong correlation ($r = 0.84$) was obtained between WUE and the seed mass parameters. WUE, which is known as the ratio of carbon gains to water losses, displays high plasticity to environmental factors. To maintain production in the face of climate change, improved WUE associated with growth maintenance is today a key target (Condon, 2020). Consistent with our results, water use efficiency and tolerance towards drought conditions were shown to be improved by inoculating plants with microbial plant biostimulants (Batool et al. 2020; Begum et al., 2022).

The current study showed a significant impact of microbial application on beans productivity and gain in yield-related parameters. Both microbial treatments T2 and T3 displayed a substantial influence on the yield evidenced by a rise in the pod number and weight, seed mass, 1000-seed weight, and seed yield per hectare (Table 3). Approximately similar results were obtained between the NPK fertilizer and T2 treatments with a significant increase, compared to the control variants. Furthermore, the combination of the microbial inoculum with organic compounds (T3) proved to be the most effective for all the yield parameters and produced the highest seed yield

(4.12 t ha⁻¹). These findings could be most probably attributed to the fact that improving soil fertility by microbial inoculation generates greater biomass and extended leaf area, which in turn promotes the rise in the production of photo-assimilates that were further directed to the seeds. This could explain the higher total seed number per m² obtained in T2 and T3, compared to the mineral fertilizer variant or the control. Indeed, an augmentation in the supply of organic materials to reproductive organs during their formation stimulates plant productivity (Symanowicz et al., 2017). This was confirmed in this study, where the highest numbers of pods per plant and total seed numbers were recorded in T3. In a study carried out by Marques et al. (2022), similar results were found in bean plants treated with a microbial product, where the authors obtained a 34% increase in the pod number, compared to the control, and showed a significant increase in all productivity parameters. Russo et al. (2020) reported also a significant elevation in pod productivity in *P. vulgaris*, compared to the control, following the application of bio-inputs.

It is notable that the microbial treatment of the bean plants may have contributed to the significant increase in pod morphology; consequently, the number of large-sized pods (category 1) per m² had the highest values in T2 and T3, compared to the mineral fertilizer and the control treatments (Table 4). In turn, this was reflected in the greater number of high quality seeds per m² from this category, compared to the mineral fertilizer variant. This trend was not evident in the category of small pods, where the control treatment produced the lowest values, but there were no significant differences between the treatments. The findings are consistent with other studies whose authors found significant changes in the dimensions of *P. vulgaris* pods following microbial inputs (Marques et al., 2022, Russo et al., 2020). A significant increase in the length of *V. fava* pods was also obtained by using biostimulants (Ali, 2019). Remarkably, the parameters related to the pod category exhibited similar values between T2 and T3, suggesting that the microbial inoculants applied favored the modifications in the pod size rather than the organic supply. These findings strongly indicate once again that, under microbial treatments, plants had the chance to efficiently use their available resources in the process of biomass establishment, seed formation, and grain filling (Poorter et al., 2012). It has been reported that nitrogen has a significant effect on bean pod loading and yields (Chekanai et al., 2018). Furthermore, additional plant nitrogen nutrition can extend the vegetative growth and increase the level of nutrient supply to the seed during the maturation phase (Laghari et al., 2016).

However, in this study, the pod morphology values obtained with the NPK fertilizer are lower than those obtained with the use of the microbial products, suggesting that the efficacy of the microbial inoculants may reside in modulating soil fertility, as indicated before, facilitating the mineral (nitrogen) and organic uptake by roots. This could explain the highest values in the total productivity parameters, such as the total seed yield per hectare, obtained by combining microbial inoculants with organic compounds (T3).

5. Conclusion

In general, it can be concluded that the application of microbial products in the bean crop exerted significant positive effects on the morphological features of plants (leaf area, aboveground biomass), productivity, and pod morphology as well as yield components and WUE. Overall, it can be assumed that an equal yield outcome can be obtained using natural microbial strains (in T2), compared to the mineral fertilizer, with a significantly greater influence on the studied parameters when combining organic matters with microbial inoculates (T3).

The co-incorporation of microbial inoculums and organic compounds seems to improve soil water/nutrient uptake from subsoil, most probably by ameliorating soil fertility and symbiotic relationships in the rhizosphere facilitated by a vigorous root system. Consequently, the leaf area development is enhanced and the photosynthetic characteristics are improved giving an extensive boost to the crop yield by assisting seed sets and grain filling. The promotion of vegetative growth traits may translate to enhanced WUE and grain yield of *P. vulgaris* crops. The risk of crop yield deficit caused by water limitation is nowadays a real threat. Development of economical

methods displaying high throughput while improving crop WUE is an on-going challenge. In this regard, the incorporation of native microbial consortia combined with organic compounds into the farming system could represent a promising strategy to investigate on a large scale. This study highlights prospects for consideration of microbial consortia combined with organic compounds in the replacement of chemical fertilizers without penalizing yields and quality of the final products (pods and seeds).

Data Availability Statement

The data presented in this study are available on request from the corresponding author.

Acknowledgments

Authors would like to thank LARI (Lebanese Agricultural Research Institute) for administrative support and for providing the laboratory premises and field to carry out assays.

References

- Adediran, J.A., Taiwo, L.B., Akande, M.O., Sobulo, R.A., & Idowu, O.J. (2007). Application of organic and inorganic fertilizer for sustainable maize and cowpea yields in Nigeria. *Journal of Plant Nutrition*, 27, 1163–1181. <https://doi.org/10.1081/PLN-120038542>
- Ali, S., Moon, Y.S., Hamayun, M., Aaqil Khan, M., Bibi, K., & Lee, I.J. (2022). Pragmatic role of microbial plant biostimulants in abiotic stress relief in crop plants. *Journal of Plant Interactions*, 17(1), 705–718. <https://doi.org/10.1080/17429145.2022.2091801>
- Ali, S.T. (2019). Effect of cultivars, plant spacing and Al-gamix on growth and green yield of broad bean (*Vicia faba* L.). *The Journal of the University of Duhok*, 22, 204–220.
- Batool, T., Ali, S., Seleiman, M.F., Naveed, N.H., Ali, A., Ahmed, K., Abid, M., Rizwan, M., Shahid, M.R., & Alotaibi, M. (2020). Plant growth promoting rhizobacteria alleviates drought stress in potato in response to suppressive oxidative stress and antioxidant enzymes activities. *Scientific Reports*, 10, 1–19. <https://doi.org/10.1038/s41598-020-73489-z>
- Begum, N., Wang, L., Ahmad, H., Akhtar, K., Roy, R., Khan, M.I., & Zhao, T. (2022). Co-inoculation of arbuscular mycorrhizal fungi and the plant growth-promoting rhizobacteria improve growth and photosynthesis in tobacco under drought stress by up-regulating antioxidant and mineral nutrition metabolism. *Microbial Ecology*, 83, 971–988. <https://doi.org/10.1007/s00248-021-01815-7>
- Cardador-Martínez, A., Loarca-Piña, G., & Oomah, B.D. (2002). Antioxidant activity in common beans (*Phaseolus Vulgaris* L.). *Journal of Agriculture and Food Chemistry*, 50, 6975–6980. <https://doi.org/10.1021/jf020296n>
- Chekanai, V., Chikowo, R., & Vanlauwe, B. (2018). Response of common bean (*Phaseolus Vulgaris* L.) to nitrogen, phosphorus and rhizobia inoculation across variable soils in Zimbabwe. *Agriculture, Ecosystems & Environment*, 266, 167–173. <https://doi.org/10.1016/j.agee.2018.08.010>
- Choudhary, M.I., Adu-Gyamfi, J.J., Saneoka, H., Tran, N.T., Suwa, R., Kanai, S., El-Shemy, H.A., Lightfoot, D. A., & Fujita, K. (2008). The effect of phosphorus deficiency on nutrient uptake, nitrogen fixation and photosynthetic rate in mashbean, mungbean and soybean. *Acta Physiologiae Plantarum*, 30(4), 537–544. <https://doi.org/10.1007/s11738-008-0152-8>
- Condon, A.G. (2020). Drying times: plant traits to improve crop water use efficiency and yield. *Journal of Experimental Botany*, 71(7), 2239–2252. <https://doi.org/10.1093/jxb/eraa002>
- El-Alam, I., Verdin, A., Fontaine, J., Laruelle, F., Chahine, R., Makhoul, H., & Sahraoui, A.L.H. (2018). Ecotoxicity evaluation and human risk assessment of an agricultural polluted soil. *Environmental Monitoring and Assessment*, 190, article 738. <https://doi.org/10.1007/s10661-018-7077-5>
- Foroud, N., Mundel, H.H., Saindon, G., & Entz, T. (1993). Effect of level and timing of moisture stress on soybean yield components. *Irrigation Science*, 13, 149–155. [https://doi.org/10.1016/0378-4290\(93\)90062-R](https://doi.org/10.1016/0378-4290(93)90062-R)
- Gao, Y., & Lynch, J.P. (2016). Reduced crown root number improves water acquisition under water deficit stress in maize (*Zea mays* L.). *Journal of Experimental Botany*, 67, 4545–4557. <https://doi.org/10.1093/jxb/erw243>
- Garden-Robinson, J., & McNeal, K. (2013). Nutrition, health benefits, preparation and use in menus. North Dakota State University Extension Service, North Dakota, USA.

- Gonzalez, E., & Paredes-Lopez, O. (2009). Hard-to-cook phenomenon in common beans—*Critical Reviews in Food Science and Nutrition*, 33, 227–286.
<https://doi.org/10.1080/10408399309527621>
- Kunicki, E., Grabowska, A., Sekara, A., & Wojciechowska, R. (2010). The effect of cultivar type, time of cultivation, and biostimulant treatment on the yield of spinach (*Spinacia oleracea* L.). *Folia Horticulturae*, 22, 9–13. <https://doi.org/10.2478/fhort-2013-0153>
- Laghari, U.A., Shah, A.N., Kandhro, M.N., Zia-ul-Hassan, Jamro, G.M., & Talpur, K.H. (2016). Growth and yield response of five elite grass pea (*Lathyrus sativus* L.) genotypes to varying levels of potassium. *Sarhad Journal of Agriculture*, 32, 218–222. <https://doi.org/10.17582/journal.sja/2016.32.3.218.222>
- Li, Z., Qiu, K., Schneider, R.L., Morreale, S.J., & Xie, Y. (2019). Comparison of microbial community structures in soils with woody organic amendments and soils with traditional local organic amendments in Ningxia of Northern China. *Peer Journal*, 7, article e6854. <https://doi.org/10.7717/peerj.6854>
- Makhlouf, H., Mouawad, C., Thabit Shaban, N., Aad, J., Sassine, L., & Samaha, H. (2023). Evaluation of the growth promoting effect of native microbial community under field conditions. *Acta Agrobotanica*, 76, article 168485. <https://doi.org/10.5586/aa/168485>
- Marques, B., Bahcevandziev, K., De Melo, P. C., Critchley, A. T. (2022). The influence of applications of bio-Inputs derived from macroalgae and bacteria on a *Phaseolus vulgaris* L. crop. *Frontiers in Bioscience* (Elite Ed), 14(3), 21. <https://doi.org/10.31083/j.fbe1403021>
- Martínez-Romero, E. (2003). Diversity of rhizobium-*Phaseolus Vulgaris* Symbiosis: Overview and Perspectives. *Plant and Soil*, 252, 11–23. <https://doi.org/10.1023/A:1024199013926>
- Naseem, H., & Bano, A. (2014). Role of plant growth-promoting rhizobacteria and their exopolysaccharide in drought tolerance of maize. *Journal of Plant Interactions*, 9, 689–701. <https://doi.org/10.1080/17429145.2014.902125>
- Nehme, N., Haydar, C., Koubaissay, B., Fakih, M., Awad, S., Yaacoub, A., Toufaily, J., Villeras F., & Hamieh, T. (2014). The distribution of heavy metals in the lower river basin, Lebanon. *Physics Procedia*, 55, 456–463. <https://doi.org/10.1016/j.phpro.2014.07.066>
- Ortiz, A., & Sansinenea, E. (2022). The role of beneficial microorganisms in soil quality and plant health. *Sustainability*, 14, article 5358. <https://doi.org/10.3390/su14095358>
- Ozbay N., Ergun M., & Demirkiran A.R. (2018). Effect of a commercial microbial fertilizer Sim Derma (*Trichoderma harzianum*, Kuen 1585) on germination, growth and yield of spinach. *Turkish Journal of Agricultural and Natural Sciences*, 5(4), 482–491. <https://doi.org/10.30910/turkjans.471290>
- Patrignani, A. & Ochsner, T. (2015). Canopeo: A Powerful New Tool for Measuring Fractional Green Canopy Cover. *Agronomy Journal*, 107(6), 2312–2320. <https://doi.org/10.2134/agronj15.0150>
- Pereira, J.L., Queiroz, R.M.L., Charneau, S.O., Felix, C.R., Ricart, C.A.O., Lopes Da Silva, F., Steindor, A.S., Ulhoa, C.J., & Noronha, E.F. (2014). Analysis of *Phaseolus vulgaris* response to its association with *Trichoderma harzianum* (ALL-42) in the presence or absence of the phytopathogenic fungi *Rhizoctonia solani* and *Fusarium solani*. *Plos One*, 9, 1–23. <https://doi.org/10.1371/journal.pone.0098234>
- Poorter, H., Buhler, J., Van Dusschoten, D., Climent, J., Postma, J.A. (2012). Pot size matters: A meta-analysis of the effects of rooting volume on plant growth. *Functional Plant Biology*, 39, 839–850. <https://doi.org/10.1071/FP12049>
- Rembiałkowska, E. (2007). Quality of plant products from organic agriculture. *Journal of Science and Food Agriculture*, 87, 2757–2762. <https://doi.org/10.1002/jsfa.3000>
- Rouphael, Y., Franken, P., Schneider, C., Schwarz, D., Giovannetti, M., Agnolucci, M., De Pascale, S., Bonini, P., & Colla, G. (2015). Arbuscular mycorrhizal fungi act as biostimulants in horticultural crops. *Scientia Horticulturae*, 196, 91–108. <https://doi.org/10.1016/j.scienta.2015.09.002>
- Russo, G.R., Teixeira, N.T., Arrud, A.C., & Tomaz Junior, D.J. (2020). Prolina e polihidroxibutirato no cultivo de feijoeiro (*Phaseolus vulgaris* L.). *Brazilian Journal of Animal and Environmental Research*, 3, 1807–1823. (In Portuguese).
- Shaban, N. (2021). Correlation and path analysis of interaction between snap beans yield and its components with crop management. *Trakia Journal of Sciences*, 19(1), 86–96. <https://doi.org/10.15547/tjs.2021.01.014>
- Shukla, P.S., Mantin, E.G., Adil, M., Bajpai, S., Critchley, A.T., & Prithiviraj, B. (2019). Ascophyllum nodosum-based biostimulants: Sustainable applications in agriculture for the stimulation of plant growth, stress tolerance, and disease management. *Frontiers in Plant Science*, 10, article 655. <https://doi.org/10.3389/fpls.2019.00655>
- Singh S.P., Terán H., Muñoz C.G., Osorno J.M., Takegami J.C., & Thung M.D.T. (2003) Low soil fertility tolerance in landraces and improved common bean genotypes. *Crop Science*, 43, 110–119. <https://doi.org/10.2135/cropsci2003.1100a>

- Symanowicz, B., Kalembsa, S., Becher, M., Toczko, M., & Skwarek, K. (2017). Effect of varied levels of fertilization with potassium on field pea yield and content and uptake of nitrogen. *Acta Scientiarum Polonorum Agricultura*, 16, 163–173.
- Thakur, N., Kaur, S., Tomar, P., Thakur, S., & Yadav, A.N. (2020). Chapter 15 – Microbial biopesticides: Current status and advancement for sustainable agriculture and environment. In Rastegari, A.A., Yadav, A.N., Yadav N. (Eds.) *New and Future Developments in Microbial Biotechnology and Bioengineering*. Elsevier, Amsterdam.
- Yadav A., Yadav K., & Ahmad R. (2023). Rhizosphere engineering for semiarid tropics: prospects and bottlenecks. *Academia Biology*, 1. <https://doi.org/10.20935/AcadBiol6075>
- Yeremko, L., Hanhur, V., & Staniak, M. (2024). Effect of mineral fertilization and seed inoculation with microbial preparation on seed and Protein yield of Pea (*Pisum sativum* L.). *Agronomy*, 14, 1004. <https://doi.org/10.3390/agronomy14051004>
- Zhao, D., Reddy, K.R., Kakani, V.G., & Reddy, V.R. (2005). Nitrogen deficiency effects on plant growth, leaf photosynthesis, and hyperspectral reflectance properties of sorghum. *European Journal of Agronomy*, 22, 391–403. <https://doi.org/10.1016/j.eja.2004.06.005>
- Zhao L.-L., Li L.-S., Cai H.-J., Shi X.-H., & Zhang C. (2018). Organic amendments influence soil water depletion, root distribution, and water productivity of summer maize in the guanzhong plain of northwest China. *Water*, 10(11), 1640. <https://doi.org/10.3390/w10111640>