



RESEARCH ARTICLE

Response of saffron to some bacteria and mycorrhiza

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Abstract

This study examines the impact of various bacterial types and mycorrhizal fungi on the growth and chemical composition of saffron (*Crocus sativus* L.) cultivated in calcareous soils of Iraq. Inoculation with *Azospirillum brasilense* increased daughter corm weight by 58 % compared with the control, followed by *Pseudomonas aeruginosa* (29 %). At the same time, *Bacillus megaterium* and *Azotobacter chroococcum* reduced growth. Combined bacterial inoculation resulted in a modest 12 % increase, while *Glomus mosseae* alone slightly decreased daughter corm weight (6 %). The interaction of *G. mosseae* with *A. brasilense* produced the highest improvement (65 %). The number of new corms doubled with *P. aeruginosa* but declined under mixed bacterial treatments due to competition. Parent corm dry weight rose with *A. brasilense* (9 %) and *A. chroococcum* (8 %), while *B. megaterium* reduced it by 38 %. Mycorrhizal inoculation enhanced parent corm dry weight by 40 %, with an additional 21 % increase when combined with bacteria. Leaf biochemical analysis showed higher nitrogen, phosphorus and iron levels under microbial treatments, with *G. mosseae* notably enhancing phosphorus (5.39 mg/g). Potassium content was unaffected by microbial interactions. These results demonstrate that biofertilizers improve saffron growth, nutrient uptake and metabolite accumulation, supporting its successful and economically viable cultivation in calcareous soils.

Keywords: bacterial inoculation; biofertilizers; mycorrhizal fungi; saffron corm growth; secondary metabolites

Introduction

Crocus sativus L., or saffron, is a small perennial plant of the Iridaceae family. It is among the most valuable medicinal and culinary plants worldwide. After drying, its stigmas are used as a spice, flavouring and natural colouring. In medicine, saffron is applied as a sedative, anti-inflammatory and treatment for coughs and asthma (1). Its distinct aroma and colour have made it highly prized in cuisine (2). The main quality factors are its secondary metabolites: crocin (colour), picrocrocin (flavour) and safranal (aroma) (3). Plants naturally host diverse microorganisms, known as the plant microbiome (4, 5). These microbes play vital roles in plant growth, development and stress tolerance. Plant growth-promoting bacteria (PGPB) enhance yield through nutrient uptake, nitrogen fixation (6), stress reduction, siderophore production, phosphate solubilization and phytohormone biosynthesis. They also suppress pathogens by producing allelochemicals (7-9). Most studied PGPB belong to the Firmicutes and Proteobacteria phyla. Their use in agriculture is increasing as an eco-friendly alternative to chemical fertilizers, herbicides and pesticides (10, 11).

Microbial inoculants have shown strong effects on saffron growth. *Curtobacterium herbarum* Cs10 increased flower number, filament length and productivity (13). Rhizobacterial strains such as *Brevibacterium frigoritolerans* (AIS-3), *Alcaligenes faecalis* subsp. *phenolicus* (AIS-8) and *Bacillus aryabhatai* (AIS-10) improved cormlet growth and yield (14). Other bacterial strains boosted leaf area, cormlet number and corm weight (15). Plant microbiomes also affect phenotypic variation, foliage growth, flowering time and

stigma yield (16). In addition, *Bacillus* strains such as *B. thuringiensis* DC1, *B. megaterium* VC3 and *B. amyloliquefaciens* DC8 reduced corm rot and improved root and shoot growth, flower number and cormlet weight (17). Biofertilizers further increase saffron yield. The best results were obtained with *Azospirillum* + VAM, followed by *Azotobacter* + Vermicompost (18). Arbuscular mycorrhizal fungi (AMF), especially *Glomus* sp., alone or with *Pseudomonas fluorescens*, improved bulb size, corm traits, carbohydrates, proteins and nitrogen assimilation (19-21). Mycorrhizal inoculation also promoted early flowering, stronger root and leaf growth, higher daughter corm numbers and enhanced apocarotenoid biosynthesis (22-24). Stigma yield increased to 3.498 kg/ha under AMF treatment, with higher fresh and dry weights, chlorophyll and sugar content (25, 26).

In Iraq, saffron cultivation faces challenges due to calcareous soils with low nutrient availability and high pH. Conventional fertilizers often fail to meet crop needs. Microbial inoculants may improve corm growth, nutrient uptake and metabolite production in these conditions, but their potential has not been fully studied. Objectives of this study included Evaluate the effects of different bacterial inoculants on saffron corm growth and development. This study was undertaken to assess the role of *Glomus mosseae* mycorrhizal fungi in improving corm traits and biochemical composition and test biofertilizers as eco-friendly alternatives for saffron production in calcareous soils. This study aims to provide a sustainable approach for saffron cultivation in Iraq, improving both yield and quality while reducing dependence on synthetic fertilizers.

Materials and Methods

Plant material and trial layout

The Turkish corms of the saffron cultivar were studied in a pot experiment as a goal crop. This cultivar was purchased from a private farm. Trial pots were prepared by mixing the cultivated media of peatmoss and sandy loam soil in a 1:1 ratio. The pots had a size of 15 kg, with dimensions of 15 cm (diameter) and 20 cm (height). Three corms were planted in each pot in November 15, 2023, thrived until May 15, 2024, when they were harvested. Two factors were randomly distributed underwith RCBD with three replicates. The first factor was corms inoculated with bacterial species, namely *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium*, *Pseudomonas aeruginosa* and a mixture of these bacterial species. The second factor was the incorporation of soil with/without mycorrhiza, specifically *Glomus mosseae*. The control treatment included without any addition. All crop practices were done as required and necessary. The fertilisation was applied using NPK 10-55-10-TE (MgO) fertilizer, originally from Iraq (Table 1). Fig 1. showed the experimental layout.

Table 1. Main and trace elements in NPK 10-55-10-TE(MgO) fertilizer

Element	Symbol	Concentration
Total nitrogen	N	10 %
Total phosphorus	P ₂ O ₅	55 %
Total potassium	K ₂ O	10 %
Total magnesium	MgO	5 %
Iron	Fe-EDTA	400ppm
Copper	Cu-EDTA	150ppm
Manganese	Mn-EDTA	250ppm
Zinc	Zn-EDTA	250ppm
Boron	B	100ppm
Molybdenum	Mo10ppm	

ppm: parts per million; EDTA: Ethylenediaminetetraacetic acid

Preparation of Bacterial Fertilizer

The isolates obtained from the microbiology laboratories at the Desert Studies Centre, University of Anbar, were cultured in liquid Nutrient Broth medium for each bacterial genus. The medium was prepared and sterilised and 50 mL of this medium was placed in a 100 mL conical flask. The selected bacterial isolate for each genus was inoculated into the flask using a sterile loop and incubated at 28°C for two days. Subsequently, four 1 L

conical flasks containing liquid nutrient broth medium were sterilized in an autoclave at 121°C and 15 psi for 20 min, followed by cooling. Ten millilitres of bacterial culture from each type were added to the conical flasks containing the medium. The flasks were placed in a shaking incubator two days before planting. The number of cells for each type of bacteria was 10 million cells per millilitre for *Azospirillum brasilense*, 8 million for *Bacillus megaterium*, 2.5 million for *Azotobacter chroococcum* and 3 million for *Pseudomonas aeruginosa*. For each liter of fertilizer, we prepared Arabic gum by dissolving 20 g in 80 mL of distilled water in a 250 mL flask. The gum powder was gradually added to the flask while stirring continuously to enhance adhesion. The seeds to be inoculated were soaked in the liquid inoculum for 30 minutes to ensure adhesion, as per standardized protocol (27). The fertilizer mixture containing the four bacterial inoculants was prepared in the field and added to the pots.

Preparation of Mycorrhizal Fungal Inoculum

The mycorrhizal fungus *Glomus mosseae*, which includes spores, infected roots and dry soil, was grown by planting maize seeds in plastic pots filled with 7 kg of sterilized soil. Fourteen grams of the fungus were placed about 5 cm deep under the top layer of soil in the pots. Fourteen grams of the inoculum were added beneath the soil surface layer in the pots at a depth of approximately 5 cm. After two months of germination, the aerial parts of the plants were removed and the soil and roots were chopped into small pieces. These pieces were then placed in sterile plastic bags and stored in a cool, dry place until used as an inoculum. Samples were examined under a microscope to confirm root colonization by the mycorrhizal fungus after staining (28). In the saffron experiment, we added 50 grams of soil containing the fungal inoculum to each pot. Estimating the total phosphorus, nitrogen concentration, total potassium content and iron was done as per the standardised protocol AOAC (29).

Data statistics

OriginPro 2025b software was undertaken to analyze data means. Means of traits had compared via Tukey test under $p \leq 0.5$. furthermore, correlation plot for traits were applied by mentioned software (30).



Fig. 1. Experimental layout of bacteria and mycorrhiza on saffron (*Crocus sativus* L.)

Results

Corm growth traits

Total corm dry weight

The results in Fig. 2 indicate the effect of adding different types of bacteria on the structural saffron corm weight individually. Some bacterial species increased the weight, with the *A. brasilense* treatment showing a 58 % increase compared to the control, reaching 8.23 g per pot. *P. aeruginosa* ranked second with a 29 % increase. In contrast, *B. megaterium* had a negative impact, reducing the weight to 3.37 g per pot compared to the control (3.49 g per pot). The most significant weight reduction was observed with *A. chroococcum*, resulting in a weight of 2.61 g per pot, which is over 25 % lower than the control. The combined bacterial treatment resulted in a 12 % increase in weight compared to the control.

Cormlet dry weight

Adding the Mycorrhizal fungus *Glomus mosseae* negatively impacted the plants, lowering the structural corm weight to 3.26 g per pot, which is more than 6 % less than the control weight of 3.49 g per pot. The results indicated that adding bacteria together with fungi had no significant effect, keeping the structural corm weight at 3.98 g per pot (fig. 3, table 2). However, we observed significant differences in corm weight when we added individual bacterial species along with *G. mosseae*. *A. brasilense* increased the weight by approximately 65 % compared to fungi alone, while *P. aeruginosa* increased the weight by about 36 %. *B. megaterium*

Table 2. Effect of some bacteria and mycorrhiza on cormlet dry weight of saffron (*Crocus sativus* L.)

Bacteria	Mycorrhiza		Average
0	0	1	
	3.49	3.26	3.375
<i>A. brasilense</i>	8.23	9.27	8.75
<i>A. chroococcum</i>	2.61	2.457	2.533
<i>B. megaterium</i>	3.37	4.62	3.995
<i>P. aeruginosa</i>	4.92	5.08	5
Mixed bacteria	3.98	3.98	3.98
LSD _{0.5}	0.6091		
Average	4.433	4.778	
LSD _{0.5}	0.2487		0.4307

LSD – Least significant difference

followed with a 29 % increase, whereas, *A. chroococcum* caused a weight reduction of more than 24 %.

Number of cormlets

Fig. 4 and Table 3 illustrate that the individual addition of *P. aeruginosa* increased the number of cormlets to 6, marking a 100 % rise. Other bacterial treatments showed an increase of over 66 % compared to the control. Conversely, the combined bacterial treatment had a negative effect, resulting in a 66 % decrease in the number of structural corms from 3 to 1. This decrease was likely due to competition for nutrients, which ultimately led to a reduction in production. The addition of mycorrhizal fungi did not affect the number of structural corms, which remained at 3 in both treated and untreated conditions. The mix of fungi and different types of bacteria didn't really change the number of corms. Still, there was a small increase with *A. brasilense* (+ 1 corm) and small decreases with *A. chroococcum* (-2 corms), *B. megaterium* (-1 corm) and *P. aeruginosa* (-1.5 corms). The addition of *G. mosseae* with all four bacterial species together did not alter corm numbers, as competition among the added organisms prevented any significant change.

Parent corm dry weight

Fig. 5 and Table 4 show that the dry weight of mother saffron corms significantly increased from 12.67 g to 13.90 g per pot with *A. brasilense*, a 9 % rise. *A. chroococcum* followed with an 8 % increase, while *P. aeruginosa* showed no significant difference from the control. However, *B. megaterium* significantly reduced

Table 3. Effect of some bacteria and mycorrhiza on cormlet number of saffron (*Crocus sativus* L.)

Bacteria	Mycorrhiza		Average
	0	1	
0	3	3	3
<i>A. brasilense</i>	5	6	5.5
<i>A. chroococcum</i>	5	3	4
<i>B. megaterium</i>	5	4	4.5
<i>P. aeruginosa</i>	6	4.5	5.25
Mixed bacteria	1	1	1
LSD _{0.5}	NS		
Average	4.17	3.58	
LSD _{0.5}	NS		1.214

LSD – Least significant difference; NS – Non-significant

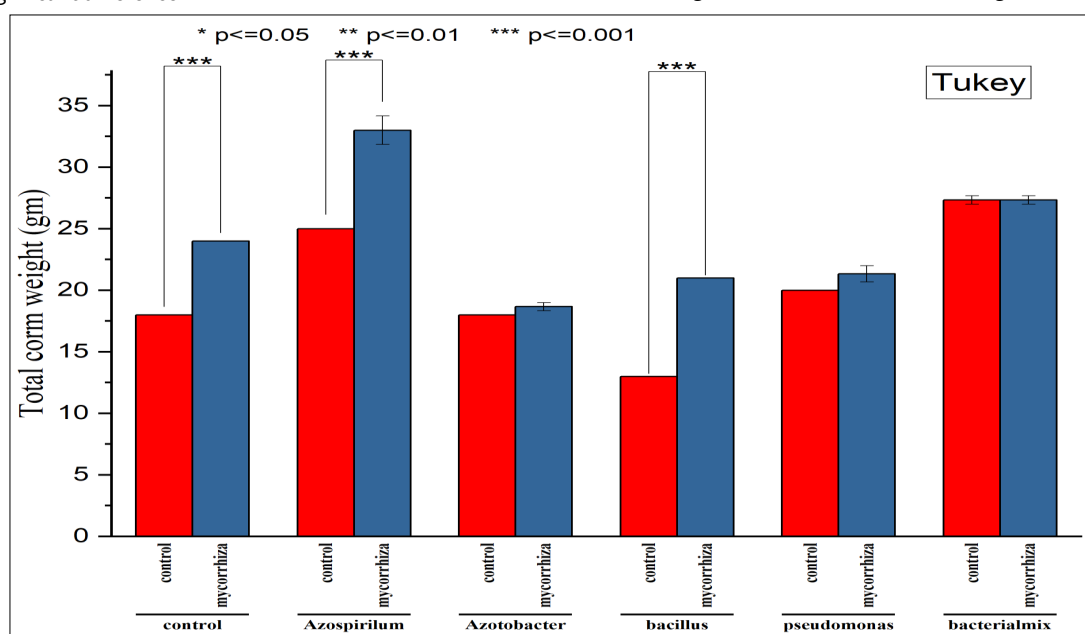


Fig. 2. Effect of some bacteria and mycorrhiza on total dry weight of saffron (*Crocus sativus* L.) corm (g). (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

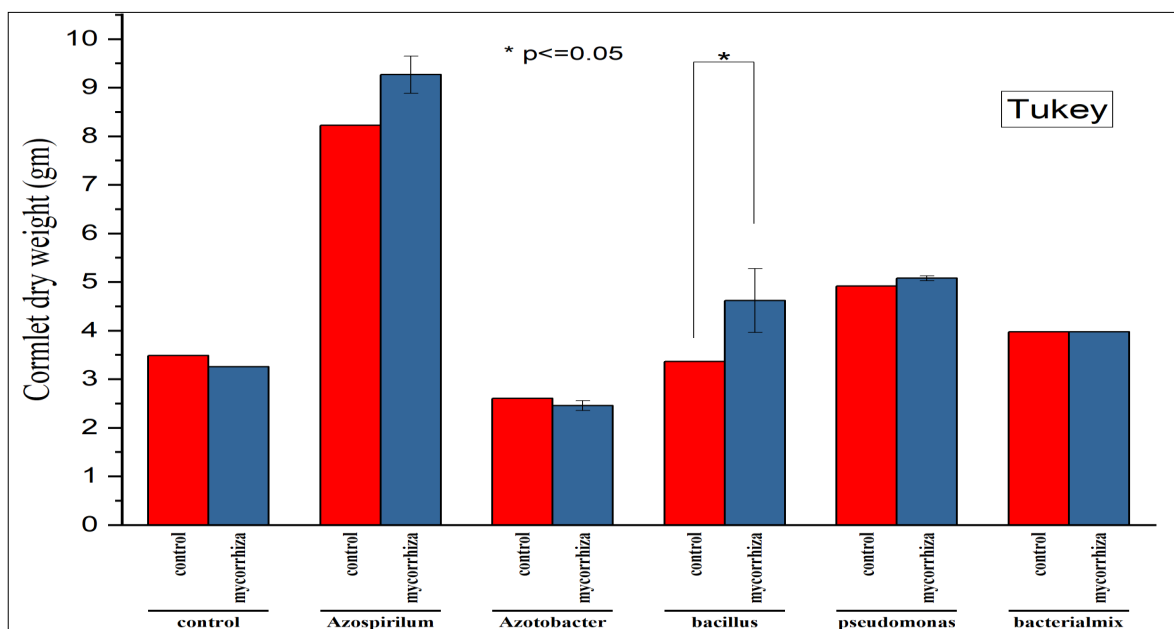


Fig. 3. Effect of some bacteria and mycorrhiza on cormlet dry weight (g) of saffron (*Crocus sativus* L.) (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

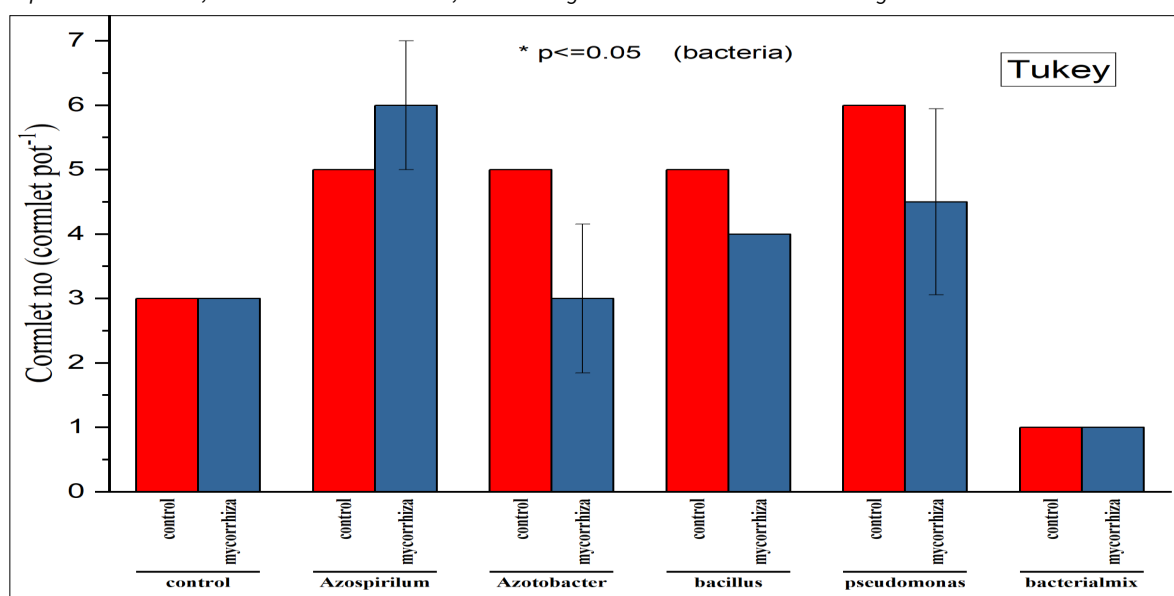


Fig. 4. Effect of some bacteria and mycorrhiza on cormlet number of saffron (*Crocus sativus* L.) corms. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

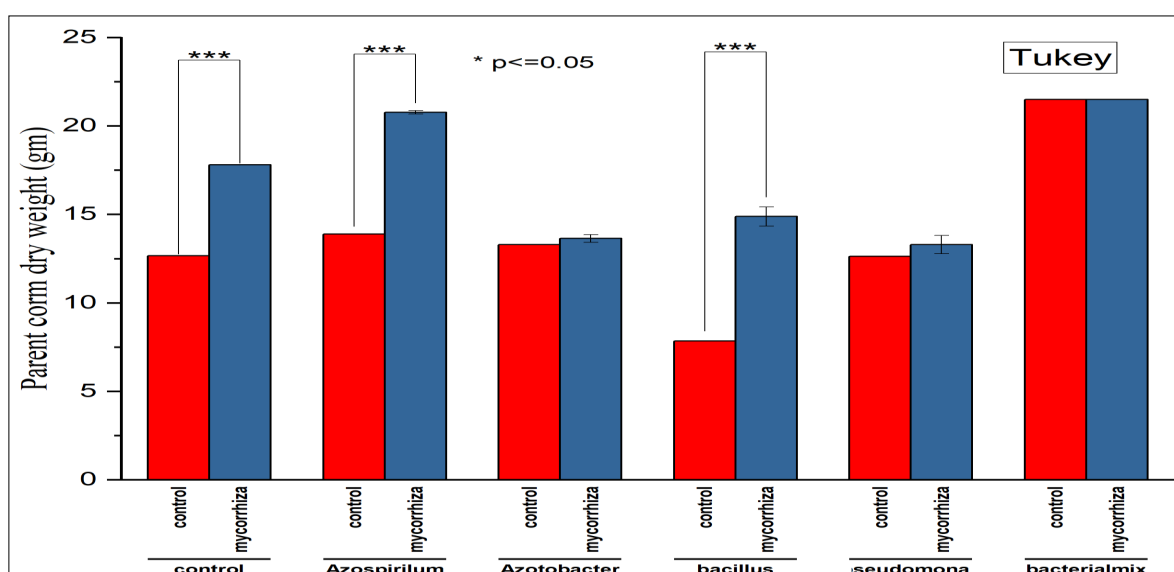


Fig. 5. Effect of some bacteria and mycorrhiza on parent dry weight (g) of saffron (*Crocus sativus* L.) corm. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

Table 4. Effect of some bacteria and mycorrhiza on parent dry weight of saffron (*Crocus sativus* L.) corm

Bacteria	Mycorrhiza		Average
	0	1	
0	12.67	17.81	15.24
<i>A. brasilense</i>	13.9	20.777	17.338
<i>A. chroococcum</i>	13.3	13.647	13.473
<i>B. megaterium</i>	7.85	14.89	11.37
<i>P. aeruginosa</i>	12.63	13.3	12.965
Mixed bacteria	21.5	21.5	21.5
LSD _{0.5}	0.6253		
Average	13.642	16.987	
LSD _{0.5}	0.2553		0.4422

LSD - Least significant difference

corm weight by 38 %. The combined bacterial treatment increased the dry weight of mother corms by approximately 70 %. Mycorrhizal fungi significantly enhanced the dry weight of mother corms by 40 % compared to untreated plants. We observed a significant 16 % increase when we added *A. brasilense* along with the fungi. The highest weight gain was observed with the combined bacterial and fungal treatment, resulting in a 21 % increase. However, when fungi were mixed with each type of bacteria separately, notable weight losses were observed: 23 % with *A. chroococcum*, 16 % with *B. megaterium* and 25 % with *P. aeruginosa*.

Parent corm number

Fig. 6 and Table 5 show that the different bacterial treatments, whether used alone or together, did not significantly change the number of parent saffron corms, regardless of the presence of mycorrhizal fungi. The application of biofertilizer had different effects on parent corms compared to structural corms. Fig. 6 shows that the different bacterial treatments, whether used alone or in combination, did not significantly affect the number of mother saffron corms, regardless of the presence of mycorrhizal fungi.

Table 5. Effect of some bacteria and mycorrhiza on parent number of saffron (*Crocus sativus* L.) corm.

Bacteria	Mycorrhiza		Average
	0	1	
0	2	2	2
<i>A. brasilense</i>	2	2.5	2.25
<i>A. chroococcum</i>	2	2	2
<i>B. megaterium</i>	1	1.333	1.167
<i>P. aeruginosa</i>	2	1.333	1.667
Mixed bacteria	2	2	2
LSD _{0.5}	NS		
Average	1.833	1.861	
LSD _{0.5}	N.S.		0.3023

LSD - Least significant difference; NS - Non-significant

Leaf dry weight

Table 6 and Fig. 7 shows significant differences in the dry weight of mother leaves with individual bacterial treatments, except for *B. megaterium*, which significantly reduced the weight. However, the mixed bacterial treatment significantly increased the dry weight of the leaves. *Azospirillum* increases leaves dry weight. Moreover, the combination of bacteria and mycorrhiza maximizes leaf dry weight, especially when bacteria are mixed with mycorrhiza and *Azospirillum* is combined with mycorrhiza. Whereas, inoculation of soil with isolated as *B. subtilis*, *B. anthracis*, *B. cereus*, *B. megaterium*, *Bacillus* sp., *Paenibacillus*, *Pseudomonas fluorescens*, *P. putida*, *Escherichia coli*, *Pectobacterium* sp. and *Pantoea* sp. did not affect the leaf dry weight of the cultivated saffron.

Table 6. Effect of some bacteria and mycorrhiza on dry weight of saffron (*Crocus sativus* L.) leaves

Bacteria	Mycorrhiza		Average
	0	1	
0	1.1	0.78	0.94
<i>A. brasilense</i>	1.28	1.7	1.49
<i>A. chroococcum</i>	0.9	0.933	0.917
<i>B. megaterium</i>	0.6	1.24	0.92
<i>P. aeruginosa</i>	1.16	0.92	1.04
Mixed bacteria	1.53	1.53	1.53
LSD _{0.5}	0.3454		
Average	1.095	1.184	
LSD _{0.5}	N.S.		0.2442

LSD - Least significant difference; NS - Non-significant

Biochemical elements of saffron leaves

Nitrogen

Fig. 8 and Table 7 show that the amount of nitrogen in leaves increased significantly with each type of bacteria used alone and in combination, except for *P. aeruginosa*, which did not differ substantially from the control. The combined bacterial treatment significantly increased nitrogen content. The addition of *G. mosseae* significantly increased nitrogen levels compared to untreated plants. However, when fungi were added with specific bacterial species, all treatments showed significant increases in nitrogen levels except for *B. megaterium*, which lowered nitrogen levels compared to the control. The combined bacterial treatment significantly enhanced nitrogen concentration in leaves, both with and without fungi. This is likely because bacterial species collectively fulfil the saffron's nitrogen requirements and fungi do not contribute to the nitrogen supply. This leads to the observed results.

Table 7. Effect of some bacteria and mycorrhiza on nitrogen of saffron (*Crocus sativus* L.) leaves

Bacteria	Mycorrhiza		Average
	0	1	
0	17.2	17.6	17.4
<i>A. brasilense</i>	17.5	17.767	17.633
<i>A. chroococcum</i>	17.467	17.633	17.55
<i>B. megaterium</i>	17.6	17.467	17.533
<i>P. aeruginosa</i>	17.3	17.967	17.633
Mixed bacteria	17.8	17.8	17.8
LSD _{0.5}	0.1242		
Average	17.478	17.706	
LSD _{0.5}	0.0507		0.0878

LSD - Least significant difference

Phosphorus

Fig. 9 and Table 8 demonstrate the significant impact of individual bacterial treatments on phosphorus concentration in saffron leaves, as well as the combined bacterial treatment. The fungal species *G. mosseae* significantly increased phosphorus concentration, recording the highest level (5.39 mg g⁻¹) compared

Table 8. Effect of some bacteria and mycorrhiza on phosphorus of saffron (*Crocus sativus* L.) leaves

Bacteria	Mycorrhiza		Average
	0	1	
0	4.8	5.3	5.05
<i>A. brasilense</i>	5	5.267	5.133
<i>A. chroococcum</i>	5.3	5.333	5.317
<i>B. megaterium</i>	5.2	5.4	5.3
<i>P. aeruginosa</i>	5.3	5.633	5.467
Mixed bacteria	5.4	5.4	5.4
LSD _{0.5}	NS		
Average	5.167	5.389	
LSD _{0.5}	0.0935		0.1620

LSD - Least significant difference; NS - Non-significant

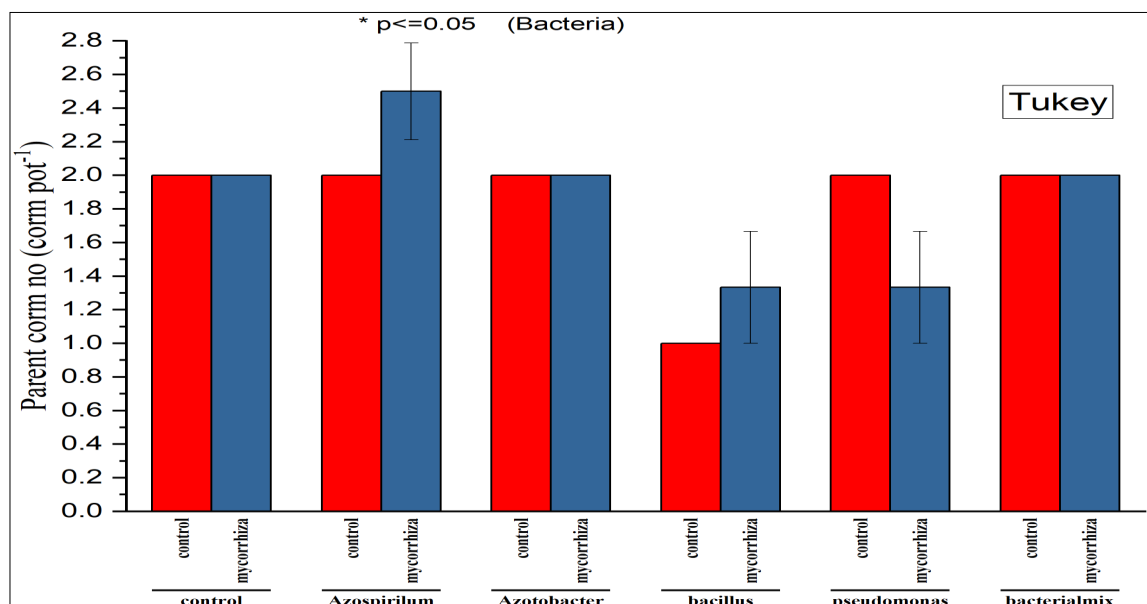


Fig. 6. Effect of some bacteria and mycorrhiza on parent number of saffron (*Crocus sativus* L.) corms. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

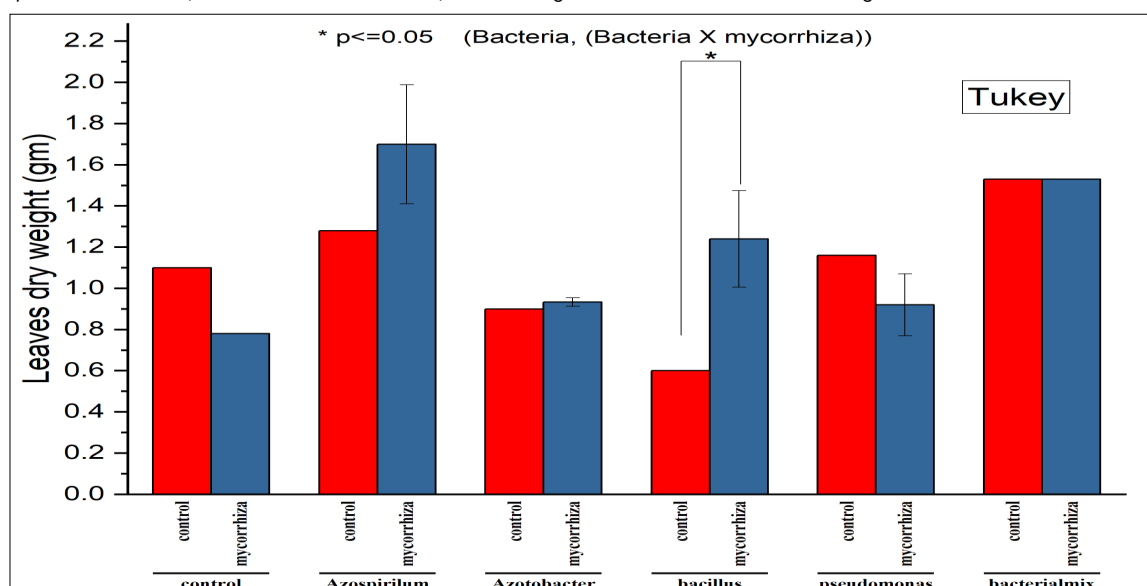


Fig. 7. Effect of some bacteria and mycorrhiza on the dry weight of saffron (*Crocus sativus* L.) leaf. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

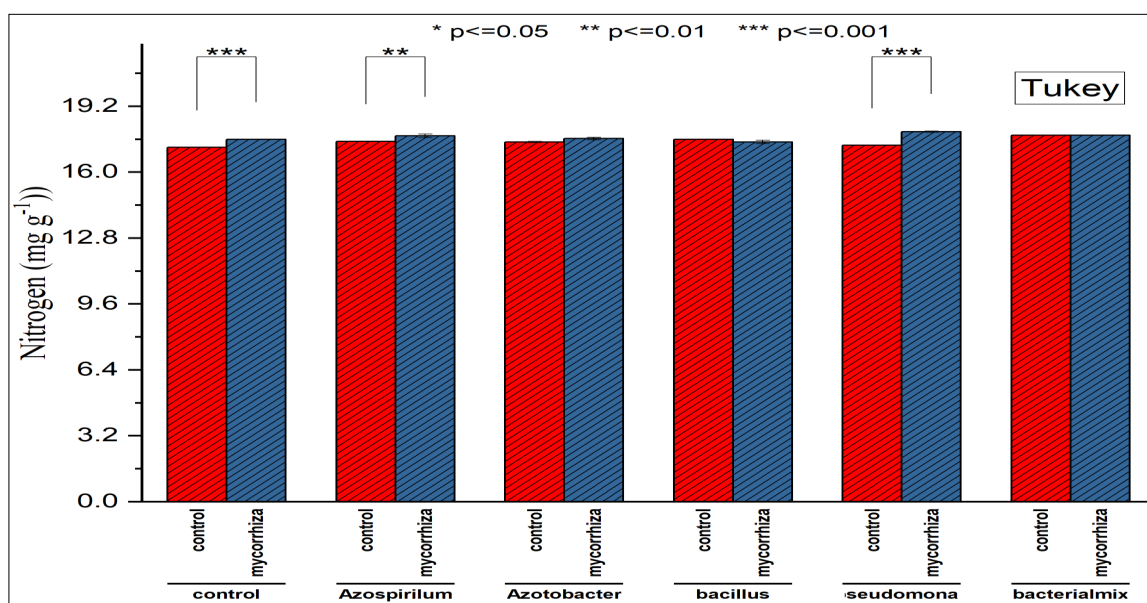


Fig. 8. Effect of some bacteria and Mycorrhiza on nitrogen of saffron (*Crocus sativus* L.) leaves. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

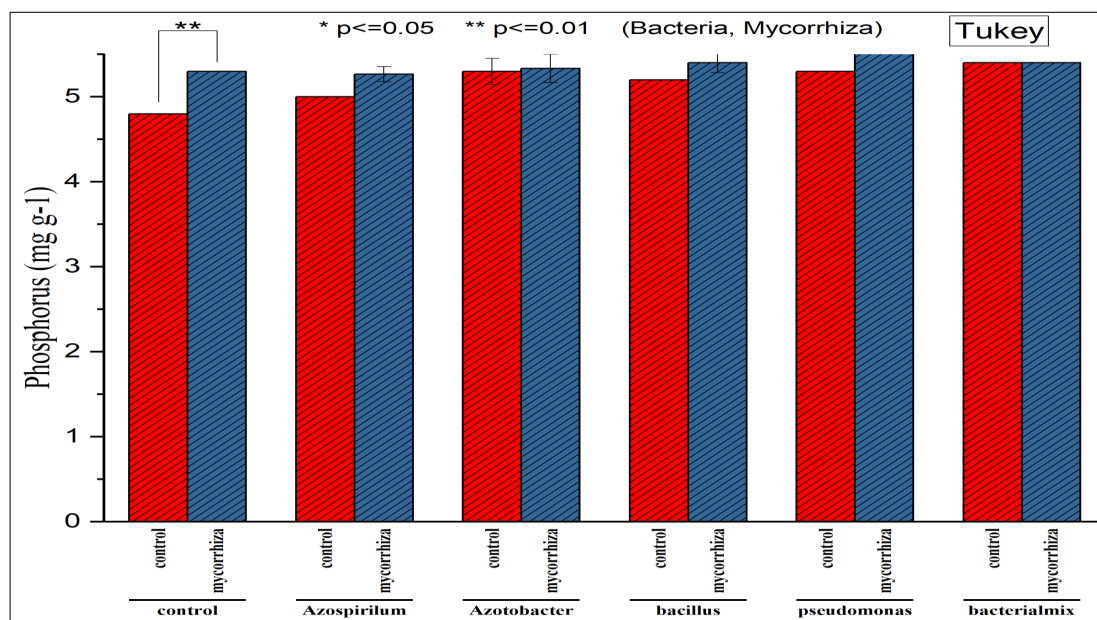


Fig. 9. Effect of some bacteria and mycorrhiza on phosphorus of saffron (*Crocus sativus* L.) leaves. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

to untreated plants (5.17 mg/g). However, the interaction between bacteria and fungi had no significant effect on phosphorus concentration.

Potassium

Fig. 10 and Table 9 illustrate the effectiveness of different bacterial treatments in enhancing potassium levels in saffron leaves, with a notable increase observed with the combined bacterial treatment (16.80 mg/g). The individual fungal treatment also had a significant effect. However, the interaction between fungi and individual bacterial species, as well as the combined bacterial treatment, was not significant.

Table 9. Effect of some bacteria and mycorrhiza on the potassium of saffron (*Crocus sativus* L.) leaves

Bacteria	Mycorrhiza		Average
	0	1	
0	16.4	16.5	16.45
<i>A. brasilense</i>	16.3	16.6	16.45
<i>A. chroococcum</i>	16.5	16.567	16.533
<i>B. megaterium</i>	16.4	16.667	16.533
<i>P. aeruginosa</i>	16.6	16.833	16.717
Mixed bacteria	16.8	16.8	16.8
LSD _{0.5}	NS		
Average	16.5	16.661	
LSD _{0.5}	0.0935		0.1300

LSD – Least significant difference; NS – Non-significant

Iron

Bacterial treatments significantly increased iron concentration in saffron leaves (Fig. 11 and Table 10). All treatments had significant effects. Similarly, fungi significantly increased iron concentration, both alone and in combination with individual or mixed bacterial treatments. Bacteria and fungi improve plant health by enhancing root and vegetative cell division and improving photosynthesis, leading to healthier plants.

Correlation analysis

Correlation analysis (Fig. 12) showed strong positive relationships between total corm dry weight and mother corm weight ($r = 0.89$), leaf dry weight ($r = 0.72$) and cormlet dry weight ($r = 0.65$). Nitrogen correlated positively with iron (0.75), phosphorus (0.64) and potassium (0.58).

Table 10. Effect of some bacteria and mycorrhiza on the iron of saffron (*Crocus sativus* L.) leaves

Bacteria	Mycorrhiza		Average
	0	1	
0	95	131	113
<i>A. brasilense</i>	138	137	137.5
<i>A. chroococcum</i>	102.33	121	111.67
<i>B. megaterium</i>	147	139.5	143.25
<i>P. aeruginosa</i>	119	149	134
Mixed bacteria	161	161	161
LSD _{0.5}	2.092		
Average	127.06	139.75	
LSD _{0.5}	0.854		1.479

LSD – Least significant difference

Discussion

Inoculation with *A. brasilense* increased saffron corm weight by 58 %, confirming its strong growth-promoting effect through enhanced nutrient uptake, nitrogen fixation and phytohormone production (6, 10, 31). Similarly, *P. aeruginosa* increased corm weight by 29 %, highlighting the contribution of PGPR to nutrient availability and pathogen suppression (8, 32, 33). In contrast, *B. megaterium* and *A. chroococcum* reduced corm weight, possibly due to nutrient competition or incompatibility with saffron physiology in calcareous soils. Mixed bacterial inoculation produced only a 12 % increase, suggesting interspecific competition reduced individual strain efficiency (11). Inoculation with *G. mosseae* alone decreased corm weight by 6 %, likely due to reduced AMF efficiency under high-pH soils (25, 35). However, co-inoculation with *A. brasilense* produced the greatest effect, increasing corm weight by 65 %. This synergy reflects enhanced nutrient uptake, particularly of phosphorus, nitrogen and micronutrients (12, 19, 36, 37). By contrast, combining fungi with *A. chroococcum* or *B. megaterium* reduced growth, emphasizing the importance of selecting compatible microbial consortia. Mother corm dry weight was unaffected by most bacterial treatments, except for *B. megaterium*, which reduced it. Mixed bacterial inoculation, however, significantly increased corm weight, likely due to complementary nutrient-solubilizing activities. Inoculation with isolates such as *B. subtilis*, *B. anthracis*, *B. cereus*, *Paenibacillus*, *P. fluorescens*, *P. putida*, *E. coli*,

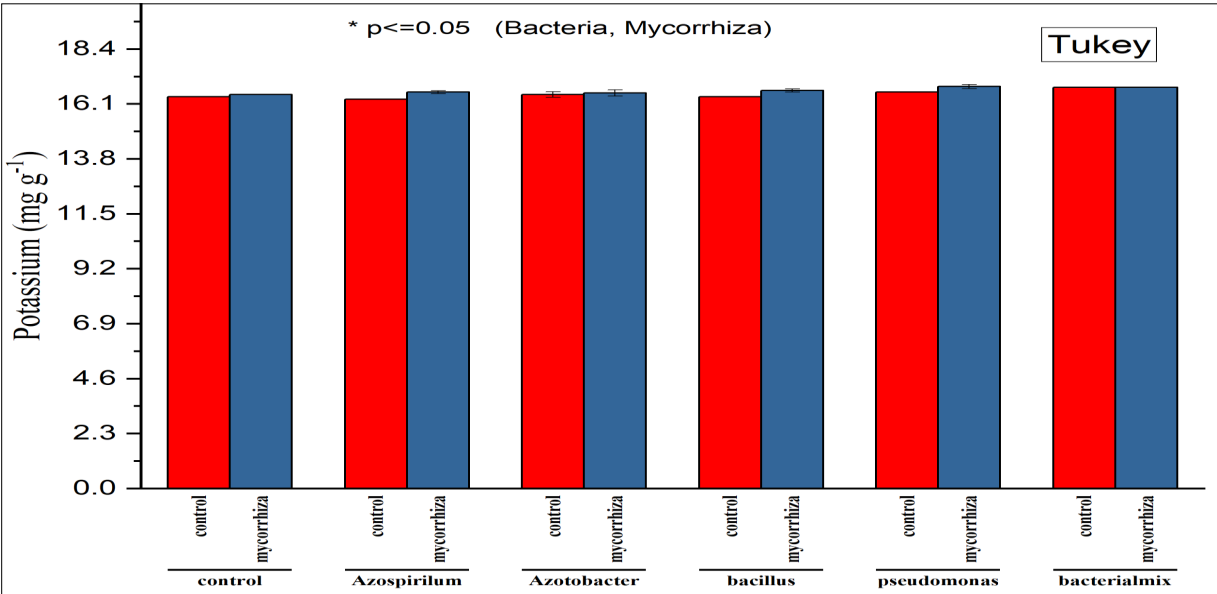


Fig. 10. Effect of some bacteria and mycorrhiza on the potassium of saffron (*Crocus sativus* L.) leaves. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

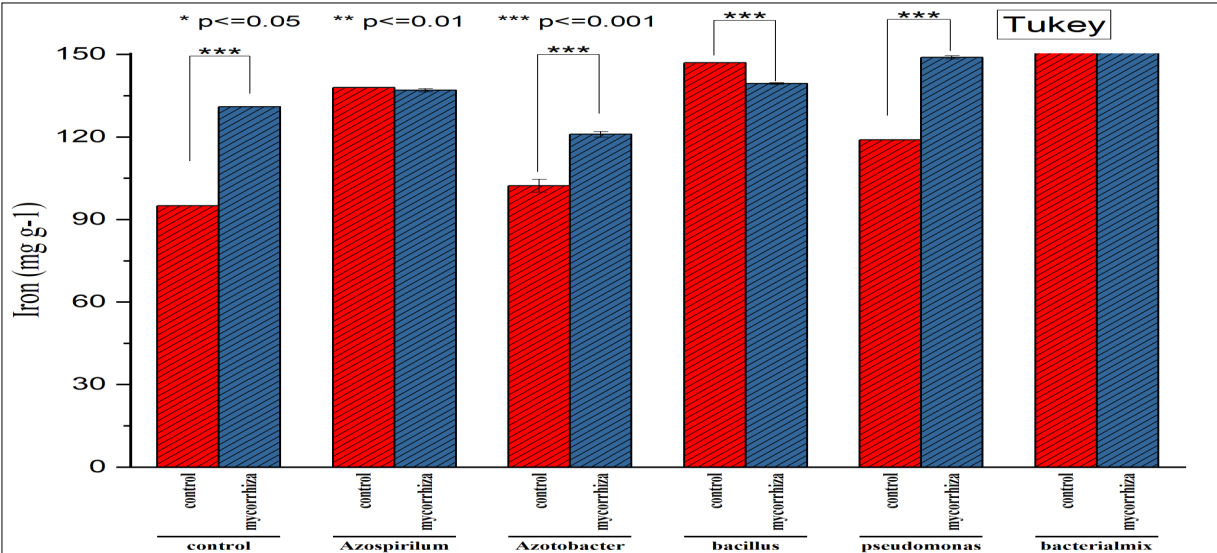


Fig. 11. Effect of some bacteria and mycorrhiza on the iron of saffron (*Crocus sativus* L.) leaves. (*, significant on 0.05, ** on 0.01, *** on 0.001) *Azospirillum brasilense*, *Azotobacter chroococcum*, *Bacillus megaterium* and *Pseudomonas aeruginosa*.

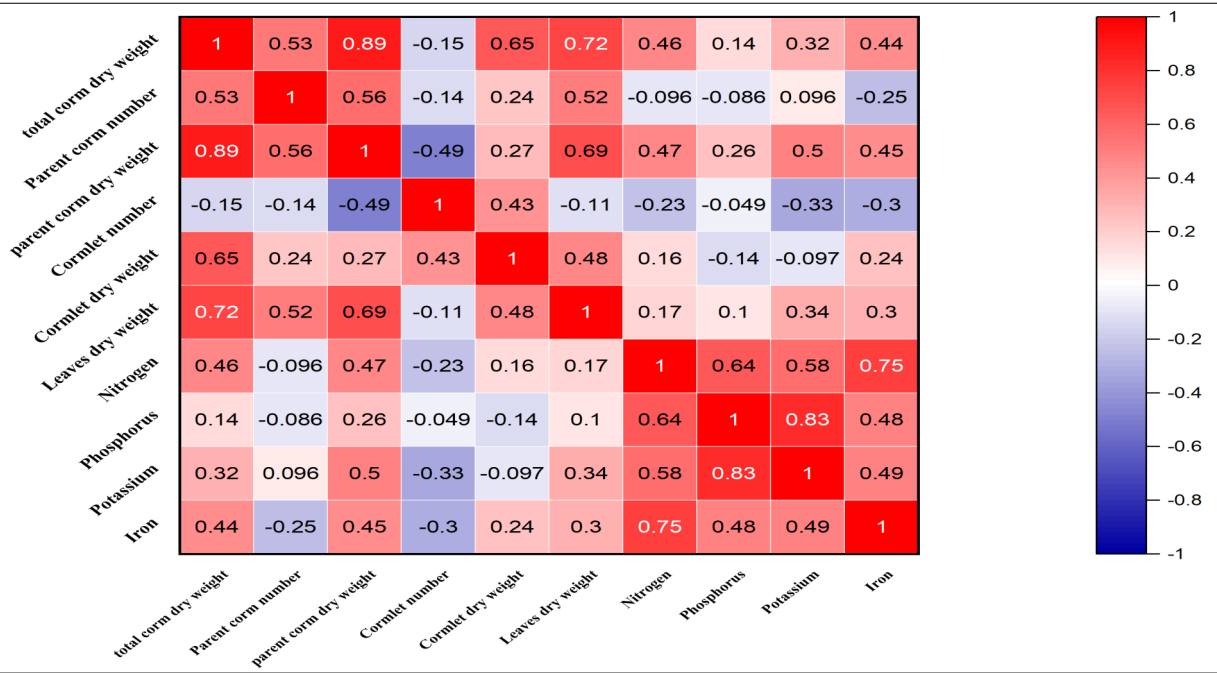


Fig. 12. Pearson correlation coefficients plot for studied traits of saffron (*Crocus sativus* L.)

Pectobacterium and *Pantoea* had no effect on leaf dry weight, consistent with previous reports (38, 39).

P. aeruginosa doubled the number of daughter corms, demonstrating its capacity to stimulate saffron propagation (16, 31, 33). Conversely, mixed bacterial inoculants reduced cormlet numbers by 66 %, likely due to resource competition (36). Mycorrhizal fungi maintained stable cormlet numbers, indicating that AMF mainly improve nutrient acquisition and biomass rather than reproductive output (22, 35). Notably, *A. brasilense* (9 %) and combined bacteria-fungi inoculation (up to 21 %) increased mother corm dry weight, reflecting enhanced carbohydrate storage, vital for subsequent growth cycles (34). Microbial treatments also improved leaf nutrient concentrations. *G. mosseae* significantly enhanced phosphorus, consistent with its role in P mobilisation, while bacterial inoculants were more effective for nitrogen and iron via fixation and siderophore production (31, 32, 37). Potassium remained largely unchanged, reflecting saffron's intrinsic regulation of K uptake, as observed in other crops (38, 39, 40, 41). Overall, biofertilizers proved effective for enhancing saffron growth and quality in calcareous soils of Iraq. Improved biomass, yield and secondary metabolites (crocin, picrocrocin, safranal) position microbial inoculation as a sustainable alternative to chemical fertilizers. Future research should focus on optimizing microbial consortia and testing long-term performance under field conditions.

Conclusion

The results indicate that saffron cultivation was successful and economically viable under Iraqi conditions, particularly in calcareous soils with moderate lime content. Studies suggest a significant improvement in saffron quality, color, aroma and growth traits when cultivated in lime-rich soils. The application of biofertilizers to saffron enhances both qualitative and quantitative traits, improving market value. These traits include plant growth, height, leaf number, fresh and dry weight, flower number, fresh flower weight, stigma yield (fresh and dry), corm number, corm weight and the content of active compounds (crocin, picrocrocin and safranal) in *Crocus sativus* L. Positive correlations between N, P and Fe and corm weight highlight nutrient assimilation as the primary mechanism. Synergy vs antagonism: Co-inoculation of *A. brasilense* and *G. mosseae* was highly beneficial, while mixed bacterial inoculations caused antagonistic effects. Potassium homeostasis: Weak associations with growth traits reflect saffron strict physiological regulation of K uptake. Investigating the interactive effects of bacterial and fungal inoculation on saffron yield and secondary metabolite production.

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Authors' contributions

AAA, MHMA, SAA and AFA made equal contributions to its writing, data verification and interpretation. Each author reviewed and approved the final version of the manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

Ethical issues: None

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