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PGPR AS AN AVAILABILITY IMPROVER OF NATURAL IRON AND MANGANESE SOURCES

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Abstract

Natural minerals are essential plant nutrient resources that can be used as natural fertilizers for cultivated plants. Bacterial inoculation improves plant-available elements in these materials. This pot experiment investigated the effects of Fe or Mn-containing natural minerals incorporation into the soils, either alone or with PGPR, on biomass development and some microelement concentrations. Fe and Mn doses were selected as 0, 100, and 200 kg ha^{-1}. The results revealed that Fe treatments promoted the biomass development of the maize plant. Bacteria inoculation increased biomass; however, the effect was statistically insignificant. In terms of mean values, increasing iron doses increased the iron concentration in the plant steadily. Bacteria inoculation increased iron uptake by 29%. Similarly, manganese-containing substrate treatments increased the biomass yield of the plant. Plant root weight and plant height were not statistically affected by manganese treatments. Dual application of manganese-containing material and bacteria inoculation increased the manganese concentration by 28%. The results clearly showed that natural minerals can be used to increase the nutrient concentration of the plant, while bacteria application further improves the uptake by about one-third. It is recommended to apply natural minerals together with the respective bacteria.

Key words: *PGPR, natural minerals, micronutrients, plant nutrition, nutrient bioavailability.*

INTRODUCTION

The two most deficient elements in Turkish soils are iron, zinc, or both (Eyüpoğlu, 1998). Even if these elements are sufficiently present in the soil, they may not be adequately uptake by plants due to unfavorable environmental factors (Ay et al., 2022). In this case, improving unfavorable soil conditions may promote nutrient uptake and therefore plant development. For instance, Ramos et al. (2024) reported that liming practice to increase base saturation in soils that have acidic soil pH resulted in an increase in microelement uptake by plants, and this increase was greater by Fe which was followed by Mn. The uptake of many microelements, including Fe and Mn, is positively affected by basic fertilization with N, P, and K (Bulut & Erdal, 2023). Durukan et al. (2022) reported that conventional fertilization was the most effective practice for Fe concentration of maize plant, while the highest Mn concentration value was reached in vermicompost application. These results revealed that each fertilizer source has a particular act on nutrient uptake. Similarly, Ay et al. (2022) reported differences in plant Fe uptake concerning different chelated iron sources they tested. Animal manure and biochar that are obtained from animal manure incorporation also increase plant uptake of micronutrients, including Fe and Mn (Mounirou, 2022). The application of various bacteria can also affect the microelement nutrition status of plants. Ertekin and Çakmakçı (2020) reported that bacterial applications may be more effective than mineral fertilization. It has been determined that inoculation of rhizobacteria such as *Bacillus* sp. together with regular fertilization promotes plant nutrient uptake and biomass development (Neta et al., 2024). This study aimed to determine the effects of Fe or Mn-containing minerals alone and in combination with bacteria on plant growth and nutrient uptake.

MATERIALS AND METHODS

To determine the effects of natural substrates on nutrient uptake of the maize plant (Monsanto DKC5709), a pot experiment was conducted. An amount of 5 kg of soil was placed in each pot

and 300 mg kg^{-1} N, 150 mg kg^{-1} P_2O_5, and 150 mg K_2O kg^{-1} were applied. The soil used in the experiment has a silty loam texture, slightly alkaline ($pH_{1:2.5}$: 7.7), slightly saline ($EC_{1:2.5}$: 0.248 dS m^{-1}), considerably highly calcareous (27%), with low organic matter as 1.76% (Alpaslan et al., 1998). While potassium, magnesium, zinc, and copper contents were found to be sufficient, calcium, iron, and manganese contents were high (Alpaslan et al., 1998). Soil texture was determined by the hydrometer method (Bouycous, 1951) whereas pH and EC were determined at 1:2.5 soil water suspension by pH and EC meter (Salinity Laboratory Staff, 1954). $CaCO_3$ was determined using a Scheibler Calcimeter (Caglar, 1949) where the organic matter was determined by the Walkley Black (Kacar and Inal, 2010) method.
Iron and manganese sources were provided by Panoxide Corporation (Yalınayak Mah. 102100 Sokak No: 44-B-C Toroslar, Mersin, Türkiye; http://panoxide.com.tr/). Iron source contains 96% FeO and 3% water-soluble salts. Mangan sources on the other hand contain 54.8% Mn, 4.74% CaO, 3.24% SiO_2, 3.11 Al_2O_3, and 0.39% Fe. The Cu and Zn concentration of the material were considerably low which were 0.0056% for Cu, and 0.014% for Zn. An analytical scale was used for dry matter determination. Micro and macro plant nutrient concentration determination was done as described by Kacar and Inal (2010).
Bacterial isolates were selected among isolates that were previously tested by Jawad and Coşkan (2019). Isolates labelled Adana3, which provided the highest Fe concentration of plants in the previous experiments, and Sivas3, which resulted in the highest Mn concentration, were tested in combination with Fe and Mn-containing minerals in this study.

RESULTS AND DISCUSSIONS

Fe incorporation results

The shoot and root dry weights and plant height values are presented in Table 1. In terms of mean values, bacterial treatments did not improve the effect of iron mineral on the dry weight of the root. In some cases, the combined application of PGPR and FeO may not provide any additional benefits if the plants are already receiving optimal levels of nutrients and other growth-promoting factors from the soil.

Table 1. Shoot and root dry weight, and plant height values influenced by iron source

bacterial inoculation	doses	shoot dry weight (g)	root dry weight (g)	plant height (cm)
- bacteria	0 kg ha^{-1}	2.63 c	2.21	62 b
	100 kg ha^{-1}	5.53 a	2.57	83 a
	200 kg ha^{-1}	3.63 bc	2.78	70 ab
+ bacteria	0 kg ha^{-1}	3.45 bc	2.81	76 ab
	100 kg ha^{-1}	4.60 ab	3.01	71 ab
	200 kg ha^{-1}	4.84 ab	2.93	83 a
averages				
- bacteria		3.93 A	2.52	72
+ bacteria		4.30 A	2.92	77
doses	0 kg ha^{-1}	3.04 B	2.51	69
	100 kg ha^{-1}	5.07 A	2.79	77
	200 kg ha^{-1}	4.24 A	2.85	77
ANOVA				
$F_{bacterial\ inoculation}$		1.75 ^{ns}	1.68 ^{ns}	3.35 ^{ns}
F_{doses}		17.50 ***	0.47 ^{ns}	3.74 ^{ns}
$F_{bacterial\ inoculation\ x\ doses}$		5.46 *	0.17 ^{ns}	9.32 **

However, mean values revealed that the mineral iron application increased the weight of the shoot significantly (P<0.001). When bacteria and doses were evaluated together as interaction, it was determined that the highest value was 5.53 g plant^{-1} at a dose of 100 kg FeO ha^{-1} without bacteria application. Root dry weight values were not significantly affected by the treatments. In plant height values, the interaction was significant, and the highest values were obtained from 100 kg ha^{-1} FeO dose without bacteria and 200 kg ha^{-1} dose with bacteria inoculation. Iron availability is mainly associated with the complex nature of plant growth and development, which is influenced by multiple factors, including soil type, nutrient availability, water availability, temperature, and other environmental factors. Thus, multiple factors should be tested to evaluate the best combination of successful bacteria and mineral application dose.

Microelements such as Fe, Zn, Mn, and Cu were analyzed, and the values obtained are presented in Table 2. Interactions were not statistically significant for all elements (P>0.05). In terms of mean values, bacterial inoculations were found to be effective on Fe and Zn concentrations, and higher Fe and Zn contents were found in bacterial inoculations compared to non-bacterial treatments. Since the material contains iron, it is expected that the iron concentration in the plant will increase with its incorporation. The increase in zinc content was associated with the possible Zn existence in the material. While increasing application doses caused a regular increase in iron concentration, the highest dose had a negative effect on Mn concentration. The effects of increasing doses on zinc and copper concentrations were found insignificant (P>0.05).

Table 2. Fe, Zn, Mn, and Cu concentrations influenced by iron source

Bacterial inoculation	Doses	Fe (mg kg^{-1})	Zn (mg kg^{-1})	Mn (mg kg^{-1})	Cu (mg kg^{-1})
- bacteria	0 kg ha^{-1}	186	21.2	78.5	30.0
	100 kg ha^{-1}	240	24.1	117.3	17.3
	200 kg ha^{-1}	252	20.2	68.0	23.0
+ bacteria	0 kg ha^{-1}	236	26.4	116.0	20.7
	100 kg ha^{-1}	263	30.1	122.7	22.7
	200 kg ha^{-1}	376	36.2	45.3	32.3
averages					
- bacteria		226 B	21.8 B	87.9	23.4
+ bacteria		292 A	30.9 A	94.7	25.2
doses	0 kg ha^{-1}	211 B	23.8	97.3 A	25.3
	100 kg ha^{-1}	251 AB	27.1	120.0 A	20.0
	200 kg ha^{-1}	314 A	28.2	56.7 B	27.7
ANOVA					
$F_{bacterial\ inoculation}$		5.69 **	20.82 ***	0.42 ns	0.29 ns
F_{doses}		4.75 **	1.78 ns	12.7 ***	1.87 ns
$F_{bacterial\ inoculation\ x\ doses}$		1.20 ns	3.06 ns	2.79 ns	2.92 ns

Correlations between increasing doses and iron concentration of the plant (Figure 1) revealed that the correlation coefficient was insignificant under non-bacterial inoculation conditions (P=0.070). Once the bacteria inoculated, the relationship between iron doses and iron concentration in the plant became significant (P<0.05). This indicates that iron application and bacterial inoculation will have significant beneficial results in improving plant iron uptake.

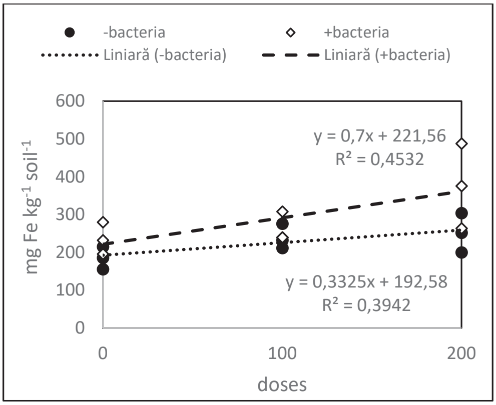

Figure 1. Correlation of dose and Fe concentrations

Mn incorporation results

The shoot and root dry weights, and plant height values influenced from Mn application are presented in Table 3. The results obtained from the manganese application were not as significant as iron application. Neither shoot and root dry weight nor plant height were influenced by Mn application based on the dual effects of bacteria and increased Mn doses. In terms of mean values, the only factor that significantly ($P<0.01$) influenced from Mn application was shoot dry weight which was increased by 54.8% at 200 kg ha^{-1} Mn containing substrate application.

Table 3. Shoot and root dry weight, and plant height values influenced from Mn source

Bacterial inoculation	Doses	Shoot dry weight (g)	Root dry weight (g)	Plant height (cm)
- bacteria	0 kg ha^{-1}	2.63	2.21	62
	100 kg ha^{-1}	4.32	2.58	73
	200 kg ha^{-1}	4.40	2.86	75
+ bacteria	0 kg ha^{-1}	3.45	2.81	76
	100 kg ha^{-1}	4.62	2.83	75
	200 kg ha^{-1}	5.02	3.13	71
averages				
- bacteria		3.78	2.55	70
+ bacteria		4.36	2.92	74
doses	0 kg ha^{-1}	3.04 B	2.51	69
	100 kg ha^{-1}	4.47 A	2.70	74
	200 kg ha^{-1}	4.71 A	2.99	73
ANOVA				
$F_{bacterial\ inoculation}$		3.6 ns	2.35 ns	1.12 ns
F_{doses}		11.53 **	1.34 ns	0.71 ns
$F_{bacterial\ inoculation\ x\ doses}$		0.25 ns	0.21 ns	2.14 ns

Table 4 represents the Fe, Zn, Mn, and Cu concentrations of the plants determined. Increasing doses of Mn incorporation increased iron concentrations in the plant. This may be related to the Fe ions in the natural mineral, or it may be related to the fact that Mn application increased biological activity and consequently increased iron bioavailability. Although bacterial inoculation increased Mn concentration in general, the difference observed was not significant ($P>0.05$). Interactions were also not significant due to this situation which is thought to be caused by high variation within observations. Zinc concentration values were not statistically significant both as main factors and as interactions. In other words, Mn-containing substrate application did not affect Zn. As expected, Mn application increased the Mn concentration in the plant. This increase was obtained from the treatment without bacterial inoculation and there was no significant relationship between the doses in bacterial inoculation. When the main factors were analyzed in terms of mean values, it was revealed that bacterial inoculation increased the Mn concentration. Cu concentration was affected by bacterial treatment (P<0.001) and in general bacterial inoculations decreased Cu concentration. This finding indicates that the bacteria inoculation may aid in reducing copper toxicity in copper-contaminated areas.

Table 4. Fe, Zn, Mn, and Cu concentrations influenced by manganese source

Bacterial inoculation	Doses	Fe (mg kg^{-1})	Zn (mg kg^{-1})	Mn (mg kg^{-1})	Cu (mg kg^{-1})
- bacteria	0 kg ha^{-1}	186	21.2	78.5 ab	30.0
	100 kg ha^{-1}	212	28.8	44.0 b	28.0
	200 kg ha^{-1}	284	35.6	94.0 a	29.3
+ bacteria	0 kg ha^{-1}	236	26.4	116.0 a	20.7
	100 kg ha^{-1}	252	36.6	92.5 a	16.0
	200 kg ha^{-1}	382	30.8	93.0 a	22.3
averages					
- bacteria		227	28.5	72.2 B	29.1 A
+ bacteria		290	31.3	100.5 A	19.7 B
doses	0 kg ha^{-1}	211 B	23.8	97.3 A	25.3
	100 kg ha^{-1}	232 AB	32.7	68.3 B	22.0
	200 kg ha^{-1}	333 A	33.2	93.5 A	25.8
ANOVA					
$F_{\text{bacterial inoculation}}$		3.30 ns	0.49 ns	14.38 *	9.30 ***
F_{doses}		4.75 *	2.42 ns	5.9 **	0.60 ns
$F_{\text{bacterial inoculation x doses}}$		0.27 ns	0.96 ns	4.03 *	0.22 ns

The relationship between increasing doses and the Mn concentration of the plant was insignificant (P>0.05) under both non-bacteria-treated and bacteria-treated conditions (Figure 2).

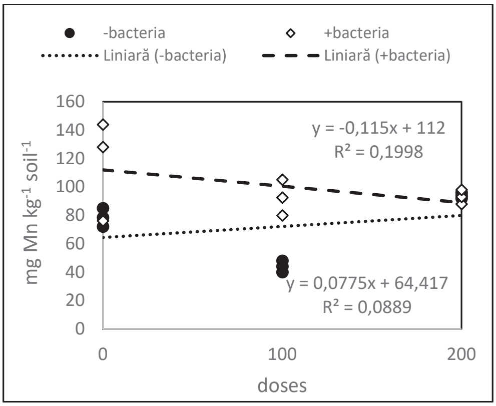

Figure 2. Correlation of dose and Mn concentrations

However, it was revealed that increasing doses of Mn-containing minerals with bacteria application decreased the Mn content of the plant depending on the increasing dose.

CONCLUSIONS

The results obtained from the experiment showed that, in general, plant nutrients containing natural substrates increased the concentration of the nutrient in the plant. While increasing doses of the substrates increased the concentration of the respective element in the plant, it is thought that in some cases higher doses should be selected to achieve effective results.

Although bacterial inoculation aims to increase plant nutrient uptake from natural minerals, this increase was found to be related to the type of the nutrient. For instance, Mn concentration decreased with bacterial inoculation while iron was positively influenced by bacterial inoculation. The results obtained revealed that the appropriate bacteria and the appropriate dose should be determined to reach the desired target.

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