

The possible use of scarce soluble materials as a source of phosphorus in *Vicia faba* L. grown in calcareous soils

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Received September 13, 2020; accepted August 06, 2021.
Delo je prispelo 13. septembra 2020, sprejeto 6. avgusta 2021

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Abstract: Phosphorus (P) is affected by many factors that minimize its solubility especially in calcareous soils. The aim of this work was to conduct laboratory and greenhouse experiments to study the effect of using P solubilizing substances, *i.e.*, compost, humic acid (HA), citric acid and ethylene di-amine tetra acetic acid (EDTA), and rhizobacteria, *Bacillus megaterium* var. *phosphaticum* on solubilizing P from different sources, ordinary superphosphate (OSP), rock phosphate (RP) and basic slag (BS). The effect of these treatments on the P- availability in El-Nubaria calcareous soil and P- uptake by faba bean (*Vicia faba* 'Giza 843') were studied. Obtained results showed that the solubility of P sources differs in their ability to release soluble P in the following order: OSP > RP > BS. The following descending order was appeared of available P in soil with addition of solubilizing agents: citric acid > EDTA > HA > compost for these sources of P, for both experiments. Regarding the interaction between solubilizing agents, the treatments of HA combined with EDTA or citric acid were superior in giving high concentrations in soil, and vigor plant growth. In addition, the solubility of P increased by about 5-6 times for all sources in the presence of P- dissolving bacteria. It seemed that the presence of appreciable amounts of Mg, S, Fe, Mn, B and other elements in BS played a role in enhancing plant growth and increasing yield, especially in the presence of added bacteria. BS could be used in calcareous soils and for soils characterized by low nutrient supply as sandy.

Key words: phosphorus sources; basic slag; organic substances; chelating substances; P availability; P dissolving bacteria; calcareous soils; *Vicia faba*

Možnost rabe slabo topnih snovi kot vir fosforja pri gojenju boba (*Vicia faba* L.) na apnenčastih tleh

Izvleček: Na topnost fosforja (P) vplivajo številni dejavniki, še posebej v apnenčastih tleh. Namen te raziskave je bil izvesti poskus v laboratoriju in rastlinjaku za preučevanje učinkov fosfor sproščajočih snovi kot so kompost, huminska kislina (HA), citronska kislina, etilen diamin tetra ocetna kislina (EDTA) in rizobakterij (*Bacillus megaterium* var. *phosphaticum*) na topnost fosforja iz različnih virov kot so navaden superfosfat (OSP), fosfat v kamnini (RP) in tomaževa žlindra (BS). Preučevani so bili učinki teh obravnavanj na dostopnost fosforja v apnenčastih tleh v El-Nubaria, Egipt in prizvem fosforja v bob (*Vicia faba* 'Giza 843'). Rezultati so pokazali, da se topnost fosforja iz različnih virov razlikuje glede na njegovo sposobnost sproščanja v naslednjem vrstnem redu: OSP > RP > BS. Po dodatku agensov za topnost se je v tleh pojavil naslednji padajoči redosled razpoložljivega P: citronska kislina > EDTA > HA > kompost, za vse vire fosforja v obeh poskusih. Glede na interakcije med agensi za topljenje se je obravnavanje HA v kombinaciji z EDTA ali citronsko kislino izkazalo kot najboljše, z največjo koncentracijo topnega P v tleh in najboljšo rastjo rastlin. Dodatno se je vsebnost P povečala za okrog 5-6 krat pri vseh virih P v prisotnosti fosfor sproščajočih bakterij. Zdi se, da je prisotnost precejšnih količin Mg, S, Fe, Mn, B in drugih elementov v tomaževi žlindri vplivala na pospešeno rast rastlin in povečanje pridelka, še posebej ob dodatku bakterij. Tomaževa žlindra bi se torej lahko uporabljala na apnenčastih tleh in v peščenih tleh, ki jih označuje majhna vsebnost hranil.

Ključne besede: viri fosforja; tomaževa žlindra; organske snovi; helatirajoče snovi, razpoložljivost P; P raztapljajoče bakterije; apnenčasta tla; *Vicia faba*

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1 INTRODUCTION

In Egypt, phosphorus (P) is the second major fertilizer comes after nitrogen and it is added to the soil mainly as an ordinary superphosphate (OSP). Phosphorus is an insoluble element in alkaline soil especially in soils containing high calcium carbonate, e.g., calcareous soils, which causes rapid precipitation to insoluble phosphate forms (Elgala & Amberger, 2017). The definition of calcareous soils, as reported by Hopkins & Ellsworth (2005), that are having significant quantities of calcium or magnesium carbonate (2-12 % depending on their particle size). These salts dissolve in neutral to acid soil pH (7-6.5), but not readily dissolve in alkaline soil (at about pH \geq 8) and, instead, serves as a sink for surface adsorbed calcium phosphate precipitation. In other words, calcareous soils with high pH resulting from high content of salts or Na⁺ and OH⁻ ions, made P is a limiting factor, causing nutritional stress conditions.

Many factors affect the solubility of P in soil and its availability to growing plants, particularly under P-stressed conditions: with using rock phosphate or other untraditional components as a source of P; such as acidifying the root medium (Houassine, 2020) and adding organic acids, amino acids and other chelating substances (Grover, 2003; Taskin et al., 2019; Elhag et al., 2019). Accordingly, getting benefit of factors that help in increasing solubility and availability of P from insoluble sources may encourage the use of rock phosphate (RP) or recycling untraditional sources *i.e.*, basic slag (BS), even under alkaline conditions, with preserving the environment from contamination. Basic slag or steel slag, as common, contains calcium oxide (CaO, 40-50 %) and silica (SiO₂, 10-28 %). Also, it includes alumina (Al₂O₃, 1-3.5 %) and magnesium oxide (MgO, 2.5-10 %), as well as iron oxide (FeO, 14-22 %) and manganese oxide (MnO, 1.5-6 %), total Fe (17-27 %), and appreciable amounts of P, K, S, and micronutrients (Tsakiridis et al., 2008; Yildirim & Prezzi, 2011; Bing et al., 2019). BS could be used in agricultural fertilizers, and environmental protection (Bing et al., 2019). Negim et al. (2010) found that the BS additions increased soil pH and conductivity, while immobilized Cu, Zn, Cr and Cd in the studied contaminated acid soil, which reflected on *Phaseolus vulgaris* L. growth. Also, Ning et al. (2016) reported that BS was an effective amendment for soil acidity adjustment, plant Si nutrition and stabilization of Cd in acidic soils.

Humic acid (HA) is a common fertilizer containing most elements that improve soil fertility and increase nutrients availability, thus enhances plant growth, and yield as well as decreases the harmful effect

of stresses (Doran et al., 2003). The effect of HA on the availability of P and micronutrients in calcareous soils have been given especial attention because of observed increases in uptake rates of these nutrients following application of HA (Satisha & Devarajan, 2005; Elhag et al., 2019). Also, compost is seen to be beneficial in improving soil fertility and crop productivity (Adugna, 2016), remediating polluted environment, recycling agricultural wastes (Taiwo, 2011), reducing the phytotoxicity of heavy metals (Huang et al., 2016), increasing water use efficiency (Adugna, 2016), and microbial activity (Huang et al., 2016; Lee et al., 2019). In addition, organic chelating agents such as EDTA and citric acid, significantly enhance element solubility and uptake by plants (Afshan et al., 2015), and are commonly used as they are more effective in chelating elements and increasing their concentrations in the upper plant organs (Kanwal et al., 2014).

Bio-fertilizers are playing a vital role in sustainable agricultural management to reduce environmental contamination (Bulut, 2013). *Bacillus megaterium* var. *phosphaticum*, which is considered a rhizobacteria, can exert a positive effect on plant growth through solubilizing inorganic phosphate and mineralizing organic phosphate, helping P to be readily available to plants with time (Abd-Elrahman, 2016; Saxena et al., 2020). Due to the P solubilization capacity, *B. megaterium* var. *phosphaticum* could be used along with RP or any other natural source to raise their efficiency in the soil. These cells can produce amino acids, vitamins, indole acetic acid (IAA), gibberellic acids, antibiotics, siderophore, as well as organic and inorganic acids that mobilize P and other nutrients and encourage the plant growth (Cakmakci et al., 1999; Amalraj et al., 2012). In addition, for the mineralization of organic P compounds, it could be due to the release of phosphatase enzymes (Illmer et al., 1995; Płaza et al., 2021).

So, the aim of this work was to conduct laboratory and greenhouse experiments to study the effect of using P solubilizing substances and rhizobacteria to solubilize P from different sources. The effect of these treatments on the P- availability in calcareous soils and P- uptake by faba bean plants (*Vicia faba* 'Giza 843') were also studied.

2 MATERIALS AND METHODS

The current study involves two trial types:

2.1 INCUBATION EXPERIMENT

To assess P content in a pure media (any salts,

CaCO₃ and P were removed) treated by several scarce soluble materials, 200 g of acid (HCl 10⁻⁴ M) washed quartz sand were placed in a plastic bowl, kept at the laboratory conditions (24 ± 2.5 °C). Thirty combinations generated from application of fifteen treatments either without adding bacteria or in the presence of dissolving bacteria (*Bacillus megaterium* var. *phosphaticum*) as follows were tested with three replications:

- 1- Ordinary Superphosphate (OSP)
- 2- OSP + Compost 1 %
- 3- OSP + Humic acid 1 %
- 4- OSP + Citric acid 1 %
- 5- OSP + EDTA 1 %
- 6- Rock Phosphate (RP)
- 7- RP + Compost 1 %
- 8- RP + Humic acid 1 %
- 9- RP + Citric acid 1 %
- 10- RP + EDTA 1 %
- 11- Basic Slag (BS)
- 12- BS + Compost 1 %
- 13- BS + Humic acid 1 %
- 14- BS + Citric acid 1 %
- 15- BS + EDTA 1 %

Extractable P concentration in each treatment and total element concentrations in basic slag were measured before adding to the soil. The P sources, *i.e.*, ordinary superphosphate (OSP), rock phosphate (RP) and basic slag (BS), were added at a rate of 4.0 g kg⁻¹ sand (equal to 9.6 t ha⁻¹). To meet the proper requirements as recommended by the Egyptian Ministry of Agriculture for faba bean cultivation in newly reclaimed soils (55.8 kg P₂O₅ ha⁻¹ in the form of ordinary superphosphate). Each of rock phosphate granules fertilizer (obtained from Abou Zaabal Company) and basic slag (obtained from Iron and Steel Company in Helwan) were ground to pass through a 2.0 mm sieve. According to the treatment, 20 ml bowl⁻¹ of *Bacillus megaterium* var. *phosphaticum* bacterial suspension, 1 × 10⁹ cells ml⁻¹, (supplied by the Department of Microbiology, Faculty of Agriculture, Ain Shams University) were added. Tap water was added to keep the moisture of the medium at the field capacity till the end of the incubation period.

Sand samples (20 g bowl⁻¹) were taken 3 times; after 2, 4 and 8 weeks. The collected samples were air dried, crushed, sieved to pass through a 2.0 mm sieve, and prepared to determine available P spectrophotometrically using Olsen extract (0.5 M NaHCO₃ at pH 8.5) according to the method described by Watanabe & Olsen (1965).

2.2 POT EXPERIMENT

A pot experiment was carried out in autumn season of 2019 at the greenhouse of Soil Science Department, Faculty of Agriculture, Ain Shams University, Qalubia governorate, Egypt. The experiment was kept in air temperature (21.9 ± 3.8 °C). Representative soil samples were collected from the surface layers (0-20 cm) of a calcareous soil, *Typic Torripsammets* (according to Soil Survey Staff, 2010), sandy loam soil from EL-Nubaria district (30°39'55" N and 30°41'49" E), Beheira governorate, Egypt. The polythene lined pots (18 cm in diameter and 15 cm in height) were packed uniformly with 3.0 kg of the investigated soil which was already air dried and ground to pass through a 2.0 mm sieve. Some initial physical and chemical properties of the studied soil were tested before plant cultivation according to the standard methods outlined by Page et al. (1982) and Klute (1986). The abovementioned 15 treatments plus 4 solubilizing agents' treatments (compost, humic acid, citric acid and EDTA) and their 6 combinations (compost+ humic acid, citric acid+ EDTA, compost+ citric acid, compost+ EDTA, humic acid+ citric acid and humic acid+ EDTA) were added to the pots and mixed well with the soil during packing, with the same doses. Two control (check) treatments were also applied (one for soil free of P sources and without adding bacteria, and the other for soil free of P sources in the presence of dissolving bacteria). Tap water was used to keep the moisture of the soil before and after plant cultivation at the field capacity till the end of the experimental work.

After one week from adding the treatments, pots were cultivated with faba bean seeds (*Vicia faba* 'Giza 843', 5 seeds pot⁻¹) on 16th of October 2019. At the same time, according to the treatment, 20 ml pot⁻¹ of *Bacillus megaterium* var. *phosphaticum* bacterial suspension (1 × 10⁹ cells ml⁻¹) were added. After seeds germination, plants were thinned to one plant pot⁻¹. Nitrogen fertilizer in the form of (NH₄)₂SO₄ and potassium in the form of K₂SO₄ were applied, at a rate of 1.0 g kg⁻¹ soil (equal to 2.4 t ha⁻¹) for each, in two batches the first one at the vegetative growth stage, 60 days after sowing (DAS), and the other one at the flowering stage (90 DAS).

2.3 MEASUREMENTS

2.3.1 Soil P content

Soil in the investigated pots was sampled 4 times: (i) after seeds germination (14 DAS), (ii) at the vegetative growth stage (60 DAS), (iii) at the flowering stage (90 DAS), and (iv) at plant harvest (145 DAS). The col-

lected samples were air dried, crushed, sieved through a 2.0 mm sieve, and prepared to determine available P spectrophotometrically using Olsen extract, as described by Watanabe & Olsen (1965).

2.3.2 Crop traits

Plants were harvested on the second week of March 2020, to assess plant height, plant fresh and dry mass, as well as number of pods plant⁻¹, fresh mass of pods and seeds plant⁻¹. Also, samples of plant leaves were oven dried at 70 °C for 48 h and digested by H₂SO₄/H₂O₂ mixture according to the method described by Chapman & Pratt (1961). Total nitrogen in leaves was determined using Kjeldahl method according to the procedure described by Chapman & Pratt (1961), total phosphorus was determined using Spectrophotometer according to Watanabe & Olsen (1965) and total potassium in plant leaves was determined using Flame photometer as described by Chapman & Pratt (1961).

2.4 EXPERIMENTAL DESIGN AND STATISTICAL ANALYSIS

The two experiments (incubation and pot experiments) were designed in a completely randomized de-

sign and each treatment was replicated three times. The obtained data were then statistically analyzed using SAS software package (SAS, 2000). Values expressed as mean and were compared for each other using Duncan's multiple range test (at $p \leq 0.05$ considered significant) \pm standard error of the mean (SEM, $n = 3$).

3 RESULTS AND DISCUSSION

3.1 INITIAL CHARACTERISTICS OF SOIL AND TREATMENTS

Extractable P concentration in each treatment before adding to the investigated soil are shown in Table 1a. The treatment of OSP is rich with P, followed by humic acid, RP, compost and BS respectively. Regarding the mixtures between treatments (Table 1a), the treatment of citric acid+ OSP gave high concentration of soluble P, followed by EDTA+ OSP, citric acid+ RP, EDTA+ RP, EDTA+ BS and citric acid+ BS respectively. Total element concentrations in basic slag were measured before adding to the soil (Table 1b). It seems good that finding appreciable amounts of P, Mg, S, Fe, Mn, B and other elements in BS. Some initial physical (soil texture, field capacity, wilting point, and saturation percent) and chemical (CaCO₃ fractions content, organic matter content, soil cation exchange capacity, pH, electrical conductivity of salts, soluble ions concentration, total and available concentration of macronutrients NPK) properties of the studied soil before plant cultivation are presented in Table 2. The studied soil is calcareous sandy loam with no saline hazards and low macronutrients concentration.

3.2 INCUBATION EXPERIMENT

Data in Table 3 shows the availability of P concentrations in acid washed sand with time; after applying different P- sources and solubilizing agents, with or without adding P dissolving bacteria. As P fertilizers (OSP, RP and BS) which vary in their P contents were added to washed sand at equal rates (4 g kg⁻¹), the extractable amounts in washed sand were significantly different between all sources after 2 weeks. With time, the soluble amounts of P increased to about 4 times at 4 weeks then dropped to about the values of the first pe-

Table 1a: Extractable- P in some of the studied treatments

Treatment	P, $\mu\text{g g}^{-1}$
Available form in:	
OSP	60.4
RP	21.6
BS	11.2
Compost	19.5
Humic acid	32.0
Mixtures (1:1, v/v)	
Citric acid 1 % + OSP (1:5)	133
Citric acid 1 % + RP (1:5)	47.0
Citric acid 1 % + BS (1:5)	30.7
EDTA 1 % + OSP (1:5)	125
EDTA 1 % + RP (1:5)	42.5
EDTA 1 % + BS (1:5)	32.0

Table 1b: Total elements concentration in the basic slag sample

Element	P	K	Ca	Mg	S	Fe	Mn	Si	Al	Cr	B	Mo
Concentration, %	0.55	0.08	32.1	5.40	0.06	19.5	2.32	7.02	1.32	0.06	0.02	<0.01

Table 2: Some initial physical and chemical characteristics of the surface layer of the experimental soil (0-20 cm) before plant cultivation

Particle size distribution, %		Soluble cations, mmol _c l ⁻¹	
Sand	65.8	Ca ²⁺	10.2
Silt	20.3	Mg ²⁺	6.34
Clay	13.9	Na ⁺	1.11
Textural class	Sandy loam	K ⁺	0.52
FC, %	12.3	Soluble anions, mmol _c l ⁻¹	
WP, %	4.20	CO ₃ ²⁻	n.d.*
SP, %	31.5	HCO ₃ ⁻	4.27
CaCO ₃ fractions, %		Cl ⁻	2.49
Coarse sand	16.8	SO ₄ ²⁻	6.28
Fine sand	8.30	Total macronutrients, %	
Silt	6.10	N	0.01
Clay	5.30	P	0.01
CaCO ₃ , g kg ⁻¹	365	K	0.02
OM, g kg ⁻¹	1.10	Available macronutrients, µg g ⁻¹	
CEC, cmol _c kg ⁻¹	11.7	N	12.3
pH (1:2.5 soil:water suspension)	8.06	P	1.00
EC _e , dS m ⁻¹	0.99	K	92.6

*n.d. means not detected, field capacity (FC), wilting point (WP), saturation percent (SP), organic matter content in soil (OM), cation exchange capacity (CEC), and electrical conductivity of salts in soil extract (EC_e)

riod for the various P- sources. It also appears that the P solubility increased to about 5-6 times for all sources in the presence of the added bacteria compared to the treatments without adding P dissolving bacteria.

With respect to the effect of the natural compounds (compost and humic acid), there was a slight increase in P solubility of the three sources when compost was added. Also, the same results were found when humic acid was added, but for OSP the P solubility increased more than the double. These results agreed with those obtained by Elhag et al. (2019) concerning the effect of humic acid on increasing P availability in washed sand. On the other hand, the effect of these natural organic sources was different when the dissolving bacteria was added, as the values of P solubility increased with compost or humic acid in these sources with more P solubility for OSP than for RP or BS. This could be related to increasing the activity of the dissolving bacteria in the presence of organic sources. The bacteria decompose these compounds to simple materials beside the materials excreted by the bacteria. All these new compounds act in dissolving the P- sources. The effect was almost double in the OSP treatment than with RP or BS treatments. These results agreed with those obtained by Abd-Elrahman (2016) about the effect of P dissolv-

ing bacteria on the solubility of P from OSP and RP fertilizers.

Results of the ability of humic acid (HA) added or formed from the decomposition of compost or any simple or complex organic compounds resulted from the action of the bacteria added could be explained on the bases that these compounds may have positive or negative charges. The positive charges bind PO₄³⁻ groups, so help in releasing the phosphate from the insoluble sources. On the other hand, the negative charge can bind Ca²⁺ ion or any cation, so, also, helps in releasing the phosphate group from the insoluble sources (Campitelli et al., 2003).

Regarding to the action of citric acid and EDTA, results show that in the absence of dissolving bacteria these compounds were superior to the compost and humic acid as they play their role directly without the action of the bacteria. This was clear with the insoluble sources RP and BS. The following descending order generally appeared in the chemically extractable amount of P with addition of solubilizing agents: citric acid > EDTA > HA > compost for these sources of P. Mihoub et al. (2018) reported that after a period of 960 h from incubation of highly calcareous soil samples (50 % CaCO₃) fertilized with triple superphosphate

and mono-ammonium phosphate and treated with citric acid and oxalic acid solutions, treatments showed a significant decrease in extractable P with time, however, applying these solutions exerted a very favorable effect on P solubility in soil. Also, in our study, with the addition of dissolving bacteria to the studied treatments activated the bacteria in dissolving the insoluble sources beside binding phosphate groups. Results also indicate that in the absence of dissolving bacteria, the soluble phosphates decrease by increasing the time of incubation at 8 weeks. But in the presence of dissolving bacteria the values in most treatments remain stable at 8 weeks. This clearly indicates that the compound formed in the presence of bacteria were more stable than that in the absence of bacteria. Abd-Elrahman (2016) reported that P dissolving bacteria increases P availability from OSP, and RP fertilizers added to a calcareous soil, with time.

3.3 POT EXPERIMENT

3.3.1 Available P in soil

Table 4 shows the effect of different P sources and solubilizing agents on available P in calcareous soil in

the presence or absence of P dissolving bacteria, during the physiological stages of faba bean growth. Results indicate that the values ranged from 1.2 to 10.4 $\mu\text{g g}^{-1}$ in the absence of bacteria, and from 3.4 to 29.8 $\mu\text{g g}^{-1}$ in the presence of bacteria. The solubility of P sources differs in their ability to release soluble P in the following order: OSP > RP > BS. In fact, this is related to their difference in their content of total- and extractable- P amounts. With respect to the ability of solubilizing agents in releasing P in the soil, it appears that they differ in the following descending order: citric acid > EDTA > HA > compost, similar to that found in the incubation experiment. As these agents were applied at the rate of 1 % (w/w), so it is expected that the active material of citric acid will be more than in compost and humic acid, as humus is composed of simple and complex compost as lignin (Taiwo, 2011). The superiority of citric acid compared to EDTA, could be explained on the basis that citric acid is smaller molecule compared to EDTA, so the active molecule will be more in 1 % of the material added compared to EDTA (Kanwal et al., 2014; Afshan et al., 2015). Besides the ability of EDTA to react with Ca to release P in the soil, it can chelate elements as Mg, Fe, Mn and Pb with higher stability (Hamed & Gamaal, 2014; Kanwal et al., 2014). This may be the reason

Table 3: Effect of the studied treatments on chemically available P ($\mu\text{g g}^{-1}$) in washed sand with time, in the presence or absence of P- dissolving bacteria

Treatment	Available P in washed sand ($\mu\text{g g}^{-1}$)					
	Without adding bacteria			In the presence of dissolving bacteria		
	after 2 weeks	4 weeks	8 weeks	after 2 weeks	4 weeks	8 weeks
OSP	1.00 \pm 0.03hi	4.20 \pm 0.12hi	0.80 \pm 0.04i	6.40 \pm 0.11f	24.4 \pm 1.92d	25.0 \pm 2.14d
OSP + Compost 1 %	1.80 \pm 0.04g	5.80 \pm 0.15f	1.60 \pm 0.06g	7.60 \pm 0.13d	24.6 \pm 2.02cd	25.6 \pm 2.19c
OSP + Humic Acid 1 %	4.00 \pm 0.14c	10.6 \pm 0.26c	1.80 \pm 0.06f	8.20 \pm 0.16c	24.8 \pm 2.03bc	25.0 \pm 2.10d
OSP + Citric Acid 1 %	9.40 \pm 0.21a	12.8 \pm 0.28a	13.2 \pm 0.17a	13.8 \pm 0.23a	54.6 \pm 2.54a	57.6 \pm 2.64a
OSP + EDTA 1 %	6.80 \pm 0.19b	11.0 \pm 0.26c	4.60 \pm 0.09b	11.4 \pm 0.17b	25.0 \pm 2.11b	27.4 \pm 2.23b
RP	0.80 \pm 0.03i	3.80 \pm 0.10ij	0.60 \pm 0.05j	6.20 \pm 0.10f	10.2 \pm 0.21j	10.2 \pm 0.19j
RP + Compost 1 %	1.00 \pm 0.03hi	5.20 \pm 0.14g	1.40 \pm 0.06h	6.40 \pm 0.12f	11.4 \pm 0.25h	10.8 \pm 0.22hi
RP + Humic Acid 1 %	1.60 \pm 0.04g	5.60 \pm 0.15f	1.40 \pm 0.07h	6.80 \pm 0.12e	12.2 \pm 0.26f	12.6 \pm 0.29f
RP + Citric Acid 1 %	3.60 \pm 0.11d	12.0 \pm 0.27b	3.80 \pm 0.08c	7.00 \pm 0.13e	12.6 \pm 0.27e	11.6 \pm 0.23g
RP + EDTA 1 %	3.00 \pm 0.12e	6.80 \pm 0.17e	3.60 \pm 0.07d	7.60 \pm 0.15d	11.8 \pm 0.26g	13.0 \pm 0.26e
BS	0.40 \pm 0.02j	3.40 \pm 0.11j	0.40 \pm 0.03k	4.40 \pm 0.08k	8.60 \pm 0.14k	9.00 \pm 0.08k
BS + Compost 1 %	1.00 \pm 0.03hi	4.60 \pm 0.13h	0.60 \pm 0.04j	4.80 \pm 0.08j	10.3 \pm 0.22ij	10.6 \pm 0.21i
BS + Humic Acid 1 %	1.20 \pm 0.03h	5.20 \pm 0.15g	0.80 \pm 0.06i	5.20 \pm 0.09i	10.4 \pm 0.22i	11.4 \pm 0.23g
BS + Citric Acid 1 %	3.40 \pm 0.14d	9.60 \pm 0.25d	3.60 \pm 0.08d	5.40 \pm 0.09hi	10.4 \pm 0.24i	11.0 \pm 0.22h
BS + EDTA 1 %	2.60 \pm 0.09f	6.60 \pm 0.16e	3.40 \pm 0.07e	5.60 \pm 0.11h	10.2 \pm 0.23j	10.8 \pm 0.20hi

Ordinary Superphosphate (OSP), Rock Phosphate (RP), Basic Slag (BS). Values expressed as mean \pm SE, the significant value was set at $p \leq 0.05$. Different letters indicate significant difference between treatments.

why extractable P from the treated soil with EDTA was less than citric acid. Mihoub et al. (2016) found that organic acids, i.e., citric acid and oxalic acid, decreased P sorption capacity on the investigated calcareous soil whereas increased Gibbs free energy (ΔG) of P which reflected on increasing its solubility in soil, however, with citric acid more than oxalic acid.

It appears that, as a function of time with growing the faba bean plants, extractable P decreased with time for the three P sources added alone and when compost and humic acid were added. This is related to activity of plant roots to utilize and withdraw the soluble P to fulfill the plant requirements. Also, it probably due to refixation of soluble P in soil by released Ca or another cation. In addition, data of extractable P in the presence of bacteria were significantly higher compared when bacteria were not added, and still the sequence was as follow: OSP > RP > BS. The reason for remaining BS in the last order may be due to its low P content. Yildirim & Prezzi (2011) reported that BS is containing small amounts of total P in the form of P_2O_5 ranged from 0.01 to 3.3 % in all different types.

Regarding the combinations between organic substances and chelating agents, the treatment of HA+ citric acid was superior in giving high extractable amount of P in soil, followed by HA+ EDTA in most physiological stages of growing bean plants, with significant differences in the presence of added bacteria. Humic acid plays a vital role in increasing P availability in soil (Doran et al., 2003; Sahin et al., 2014), plus its considerable content of P (Table 1a). Citric acid, in addition to decrease soil pH, it makes complexes with Ca forming calcium citrate and releasing P in soluble form in the soil (Drouillon & Merckx, 2003). EDTA may solubilize the insoluble P forms in calcareous soils by chelating Ca^{2+} and Mg^{2+} cations, lowering soil pH and/ or the partial occupation of active anionic groups on the surface of $CaCO_3$ and clay minerals (Hamed & Gamal, 2014). It appears that the presence of bacteria played a protective role against P-fixation (Abd-Elrahman, 2016; Plaza et al., 2021).

3.3.2 Vegetative growth parameters

Data in Table 5 shows the effect of the studied P-treatments on faba bean plant height, and plant fresh and dry mass, in the presence or absence of P-dissolving bacteria. The data of plant dry mass indicate that the addition of P-sources increased the dry mass yield more than the control and the rate of increase was in the same sequence mentioned for P availability in the soil: OSP > RP > BS (Table 4). The role of P in enhancing

roots growth and their absorption efficiency in the soil was observed, which reflected on the plant growth and its yield. Razaq et al. (2017) found that applying P fertilizer increased root surface area, specific root length and root-shoot ratio. Fouda (2017) confirmed the effect of P fertilizer on increasing faba bean productivity.

With respect to the effect of solubility agents, the sequence was different: EDTA > citric acid > HA > compost for OSP and RP, but the values were almost the same for BS. This indicates that despite relatively low total P content in BS (Table 1b), the P is found in a form easily released by the solubilizing agents. The addition of solubilizing bacteria increased the yield of dry mass to the extent to record slight significant difference between the studied P-sources. This was also found when comparing the effect of solubilizing agents on yield in case of RP and BS were almost the same. Such soil with its high content of $CaCO_3$ is characterized by deficiency problems with some elements, particularly the micro-nutrients. The presence of appreciable amounts of Fe, Mn, Mg, S, B and other elements in BS had an effect of plant growth despite the low P content added (Table 1b).

It is interesting to note that the highest yield recorded for this experiment was when EDTA was added to OSP treatment, or mixed with HA, in the presence of P-dissolving bacteria. The above results indicate that solubilizing agents and dissolving bacteria not only act in solubilizing P from added materials and from soil, but also act in solubilizing other elements as Fe, Mn and Mg which are essential for plant growth. EDTA is known to chelate elements as Fe, Mn, Zn and Mg with higher stability as compared to citric acid or natural compounds as humic acid (Hamed & Gamal, 2014).

Similar trend was observed with the other vegetative growth parameters of faba bean plants as affected by the studied P treatments, with significant effect in the presence of P-dissolving bacteria as compared to not adding bacteria. Plant height ranged from 25 cm (in control treatment, without adding P sources and dissolving bacteria) to 70 cm (with applying the treatment of HA combined with EDTA, in the presence of dissolving bacteria). Also, the plant fresh mass ranged from 22.8 g plant⁻¹ in control treatment (without any additions) to 60.5 g plant⁻¹ with applying the treatment of HA combined with EDTA, in the presence of P-dissolving bacteria.

3.3.3 Numbers of pods plant⁻¹, fresh mass of pods plant⁻¹ and fresh mass of faba bean seeds plant⁻¹

Results of mass of seeds (g plant⁻¹) shown in Table

Table 4: Effect of the applied P sources on chemically available P ($\mu\text{g g}^{-1}$) in El-Nubaria calcareous soil, in the presence or absence of P- dissolving bacteria, during the physiological stages of growing faba bean plants

Treatment	Available P in soil ($\mu\text{g g}^{-1}$)							
	After seeds germination (14 days after sowing)		At the vegetative growth stage (60 days after sowing)		At the flowering stage (90 days after sowing)		At plant harvest (145 days after sowing)	
	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**
Control (-P)	0.80 ± 0.04q	2.40 ± 0.12o	1.00 ± 0.04r	3.60 ± 0.26v	0.80 ± 0.04r	3.40 ± 0.18s	0.80 ± 0.03p	2.40 ± 0.12q
OSP	2.64 ± 0.07k	6.40 ± 0.51h	4.20 ± 0.26jk	17.0 ± 1.16f	3.20 ± 0.13j	18.2 ± 1.26e	2.10 ± 0.08jk	9.20 ± 0.71l
OSP + Compost 1 %	3.60 ± 0.10g	7.80 ± 0.80f	5.20 ± 0.34g	18.6 ± 1.21d	3.80 ± 0.15i	19.0 ± 1.33d	2.60 ± 0.09h	10.0 ± 0.83k
OSP + Humic Acid 1 %	5.20 ± 0.23d	9.20 ± 0.86c	8.00 ± 0.62c	19.8 ± 1.28c	5.60 ± 0.34d	19.5 ± 1.35c	3.20 ± 0.17f	11.0 ± 0.92hi
OSP + Citric Acid 1 %	9.83 ± 0.49a	15.6 ± 1.02a	10.4 ± 0.81a	29.0 ± 2.06a	10.2 ± 0.83a	29.8 ± 2.31a	4.80 ± 0.21a	18.4 ± 1.27a
OSP + EDTA 1 %	7.80 ± 0.33b	12.4 ± 0.97b	8.80 ± 0.64b	21.2 ± 1.78b	8.20 ± 0.60b	22.0 ± 2.01b	4.20 ± 0.20c	13.6 ± 1.16c
RP	2.00 ± 0.06m	5.60 ± 0.29i	3.80 ± 0.15l	10.6 ± 0.84mn	2.40 ± 0.11n	10.7 ± 0.80m	1.40 ± 0.07n	10.0 ± 0.85k
RP + Compost 1 %	2.41 ± 0.09k	6.80 ± 0.48g	4.00 ± 0.23kl	11.4 ± 0.90l	2.44 ± 0.12n	11.2 ± 0.92kl	2.01 ± 0.09k	10.8 ± 0.89ij
RP + Humic Acid 1 %	3.20 ± 0.12h	8.00 ± 0.85ef	5.20 ± 0.30g	12.2 ± 0.95k	4.00 ± 0.23h	12.8 ± 0.96j	2.40 ± 0.09i	11.6 ± 0.91g
RP + Citric Acid 1 %	5.60 ± 0.26c	8.41 ± 0.84d	6.80 ± 0.42d	13.4 ± 0.98j	6.40 ± 0.41c	13.6 ± 0.098i	4.40 ± 0.23b	13.4 ± 1.15cd
RP + EDTA 1 %	4.20 ± 0.18f	8.20 ± 0.80de	6.40 ± 0.40e	12.5 ± 0.93k	4.80 ± 0.20f	12.8 ± 0.95j	4.03 ± 0.21c	12.8 ± 1.06e
BS	1.80 ± 0.09n	4.40 ± 0.23l	3.20 ± 0.17n	8.23 ± 0.61s	2.00 ± 0.10o	8.40 ± 0.61q	1.00 ± 0.06o	9.20 ± 0.72l
BS + Compost 1 %	2.20 ± 0.10l	5.20 ± 0.28j	4.00 ± 0.22kl	9.20 ± 0.74q	2.40 ± 0.12n	9.00 ± 0.73op	1.61 ± 0.07m	10.6 ± 0.85j
BS + Humic Acid 1 %	2.80 ± 0.13j	6.20 ± 0.47i	4.63 ± 0.25hi	9.60 ± 0.76p	3.00 ± 0.16k	10.0 ± 0.80n	1.80 ± 0.07l	12.2 ± 1.02f
BS + Citric Acid 1 %	4.60 ± 0.17e	6.80 ± 0.50g	5.80 ± 0.35f	10.8 ± 0.81m	5.20 ± 0.29e	11.6 ± 0.94k	3.80 ± 0.17d	13.2 ± 1.11de
BS + EDTA 1 %	3.60 ± 0.15g	6.40 ± 0.49h	4.86 ± 0.26h	10.4 ± 0.80n	4.20 ± 0.23g	10.8 ± 0.84lm	3.61 ± 0.16e	12.6 ± 1.06e
OSP+ RP+ BS (50 % for everyone)	2.40 ± 0.11k	7.02 ± 0.68g	4.40 ± 0.24ij	18.0 ± 1.27e	3.20 ± 0.16j	18.8 ± 1.20d	1.80 ± 0.08l	9.00 ± 0.78l
Compost 1 %	1.80 ± 0.07n	4.60 ± 0.28kl	2.40 ± 0.08p	8.81 ± 0.62r	2.40 ± 0.13n	8.60 ± 0.67pq	2.00 ± 0.09k	4.82 ± 0.21o
Humic Acid 1 %	2.20 ± 0.013l	5.60 ± 0.30i	3.20 ± 0.18n	10.0 ± 0.81o	3.00 ± 0.15k	10.2 ± 0.83n	2.20 ± 0.09j	5.70 ± 0.23n
Citric Acid 1 %	1.40 ± 0.08o	3.80 ± 0.19m	1.80 ± 0.06q	7.40 ± 0.54t	2.00 ± 0.11o	7.80 ± 0.55r	2.00 ± 0.08k	4.80 ± 0.22o
EDTA 1 %	1.20 ± 0.06p	3.40 ± 0.17n	1.60 ± 0.06q	6.80 ± 0.44u	1.60 ± 0.08q	7.40 ± 0.53r	1.42 ± 0.07n	4.00 ± 0.19p
Compost 1 %+ Humic Acid 1 % (50 % for both)	2.20 ± 0.10l	5.20 ± 0.26j	2.80 ± 0.09o	14.6 ± 1.10h	2.60 ± 0.13m	14.7 ± 1.15hi	2.20 ± 0.10j	11.1 ± 0.90h
Citric Acid 1 %+ EDTA 1 % (50 % for both)	1.40 ± 0.07o	3.60 ± 0.18mn	1.89 ± 0.07q	9.87 ± 0.71op	1.80 ± 0.08p	9.20 ± 0.72o	1.60 ± 0.08mn	6.60 ± 0.42m
Compost 1 %+ Citric Acid 1 % (50 % for both)	2.20 ± 0.12l	5.20 ± 0.29j	3.00 ± 0.23no	15.0 ± 1.18j	2.80 ± 0.15l	15.8 ± 1.24f	2.60 ± 0.12h	10.8 ± 0.89ij
Compost 1 %+ EDTA 1 % (50 % for both)	2.02 ± 0.09m	4.80 ± 0.25k	3.00 ± 0.25no	14.0 ± 1.05i	2.60 ± 0.14m	14.6 ± 1.13hi	2.60 ± 0.11h	10.0 ± 0.90k
Humic Acid 1 %+ Citric Acid 1 % (50 % for both)	3.00 ± 0.15i	6.60 ± 0.54h	4.02 ± 0.26kl	17.0 ± 1.17f	3.80 ± 0.17i	19.2 ± 1.38cd	3.00 ± 0.18g	15.6 ± 1.18b
Humic Acid 1 %+ EDTA 1 % (50 % for both)	2.40 ± 0.11k	5.40 ± 0.32i	3.40 ± 0.25m	14.2 ± 1.12i	3.00 ± 0.18k	15.2 ± 1.20g	2.65 ± 0.13h	11.2 ± 0.97h

*(-PDB) means without adding P dissolving bacteria, **(+PDB) means in the presence of P dissolving bacteria. Ordinary Superphosphate (OSP), Rock Phosphate (RP), Basic Slag (BS). Values expressed as mean ± SE, the significant value was set at $p \leq 0.05$. Different letters indicate significant difference between treatments.

Table 5: Effect of the applied P sources on vegetative growth parameters of faba bean plants cultivated on El-Nubaria calcareous soil, in the presence or absence of P-dissolving bacteria

Treatments	Plant height, cm		Plant fresh mass, g		Plant dry mass, g	
	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**
Control (-P)	25.0 ± 2.21q	35.5 ± 2.44s	22.8 ± 2.13s	28.4 ± 2.38u	4.94 ± 0.23s	7.48 ± 0.54s
OSP	32.5 ± 2.80k	39.0 ± 2.68o	26.2 ± 2.25m	34.9 ± 3.29o	7.12 ± 0.50l	10.9 ± 0.81no
OSP + Compost 1 %	34.0 ± 2.87i	44.0 ± 3.15j	28.1 ± 2.31j	37.8 ± 3.40l	8.34 ± 0.57k	12.1 ± 0.98k
OSP + Humic Acid 1 %	36.3 ± 3.01g	47.0 ± 3.21g	29.8 ± 2.45i	41.7 ± 4.04ij	9.18 ± 0.65gh	13.5 ± 1.07h
OSP + Citric Acid 1 %	50.0 ± 4.43b	58.4 ± 4.01d	33.3 ± 2.70e	49.1 ± 4.25d	9.90 ± 0.72d	16.1 ± 1.29c
OSP + EDTA 1 %	53.0 ± 4.50a	60.1 ± 4.29c	34.6 ± 2.76c	58.6 ± 4.46b	11.3 ± 0.86a	18.0 ± 1.33b
RP	28.0 ± 2.52o	36.5 ± 2.35r	23.4 ± 2.22q	32.2 ± 2.89qrs	6.69 ± 0.43p	9.83 ± 0.76r
RP + Compost 1 %	30.5 ± 3.22l	40.7 ± 2.80n	27.1 ± 2.29l	35.4 ± 2.93n	7.19 ± 0.52l	11.1 ± 0.91no
RP + Humic Acid 1 %	32.0 ± 2.34k	43.0 ± 3.39k	28.3 ± 2.36j	38.5 ± 3.45k	8.90 ± 0.64i	11.4 ± 0.90m
RP + Citric Acid 1 %	40.0 ± 3.11f	46.0 ± 3.50h	31.8 ± 2.83g	45.5 ± 4.11f	9.30 ± 0.73f	14.8 ± 1.12f
RP + EDTA 1 %	41.0 ± 3.23e	48.0 ± 3.55f	32.3 ± 2.87f	47.8 ± 4.19e	9.82 ± 0.79d	15.8 ± 1.18d
BS	26.3 ± 2.25p	35.9 ± 2.37rs	23.1 ± 2.26r	30.9 ± 2.47t	5.93 ± 0.32r	9.65 ± 0.76r
BS + Compost 1 %	29.1 ± 2.53n	38.0 ± 2.41pq	24.1 ± 2.32o	32.5 ± 2.90qr	6.89 ± 0.39no	10.4 ± 0.87pq
BS + Humic Acid 1 %	30.5 ± 2.87l	38.2 ± 2.38p	24.4 ± 2.34n	32.7 ± 2.89p	6.93 ± 0.45mno	10.5 ± 0.88p
BS + Citric Acid 1 %	36.5 ± 2.91g	43.5 ± 3.60jk	27.6 ± 2.78k	37.7 ± 3.37l	8.90 ± 0.62i	11.8 ± 0.94l
BS + EDTA 1 %	35.4 ± 2.76h	45.0 ± 3.62i	27.8 ± 2.76k	41.9 ± 4.12i	9.11 ± 0.71h	12.6 ± 0.98ij
OSP+ RP+ BS (50 % for everyone)	32.0 ± 2.79k	41.3 ± 3.22lm	27.2 ± 2.69l	37.1 ± 3.32m	8.63 ± 0.59j	12.5 ± 0.97j
Compost 1 %	28.0 ± 2.23o	37.8 ± 2.43q	23.4 ± 2.23q	32.0 ± 3.45s	6.33 ± 0.44q	10.3 ± 0.86q
Humic Acid 1 %	30.0 ± 2.69m	38.0 ± 2.47pq	23.9 ± 2.29p	32.6 ± 3.51pq	6.81 ± 0.47o	10.4 ± 0.87pq
Citric Acid 1 %	33.4 ± 2.80j	41.1 ± 3.26mn	24.0 ± 2.38op	33.1 ± 3.60p	6.97 ± 0.48mn	10.8 ± 0.92o
EDTA 1 %	34.0 ± 2.85i	41.7 ± 3.35l	24.0 ± 2.37op	35.4 ± 3.66n	6.98 ± 0.51m	11.9 ± 0.98kl
Compost 1 %+ Humic Acid 1 % (50 % for both)	35.4 ± 2.49h	47.0 ± 3.56g	30.5 ± 2.44h	41.3 ± 4.13j	9.23 ± 0.42fg	12.7 ± 1.02i
Citric Acid 1 %+ EDTA 1 % (50 % for both)	42.5 ± 3.44d	45.0 ± 3.41i	34.1 ± 2.51d	44.0 ± 4.20g	10.1 ± 0.78c	14.8 ± 1.13f
Compost 1 %+ Citric Acid 1 % (50 % for both)	40.0 ± 3.39f	54.3 ± 4.52e	32.3 ± 2.49f	43.1 ± 4.18h	9.10 ± 0.69h	12.9 ± 1.05i
Compost 1 %+ EDTA 1 % (50 % for both)	42.0 ± 3.38d	60.0 ± 4.58c	32.5 ± 2.47f	48.7 ± 4.70d	9.31 ± 0.73f	14.3 ± 1.08g
Humic Acid 1 %+ Citric Acid 1 % (50 % for both)	46.0 ± 2.56c	64.0 ± 4.71b	36.1 ± 2.72b	53.9 ± 4.93c	9.50 ± 0.75e	15.3 ± 1.17e
Humic Acid 1 %+ EDTA 1 % (50 % for both)	50.0 ± 2.70b	70.0 ± 4.93a	39.4 ± 3.14a	60.5 ± 5.03a	10.9 ± 0.82b	18.7 ± 1.38a

*(-PDB) means without adding P dissolving bacteria, **(+PDB) means in the presence of P dissolving bacteria. Ordinary Superphosphate (OSP), Rock Phosphate (RP), Basic Slag (BS). Values expressed as mean ± SE, the significant value was set at $p \leq 0.05$. Different letters indicate significant difference between treatments

6 indicates the values ranged between 1.21 g to 11.5 g with lower values for treatments without bacteria addition. The addition of P fertilizers significantly increased the seed yield by about 5 times with OSP and only about 3 times for RP and BS compared to the control without bacteria addition. When solubilizing bacteria was added the magnitude of increase was only about the double in case of OSP and slightly less than double in case of RP and BS. This means that the solubilizing bacteria play a major role than the P- sources. *Bacillus megaterium* var. *phosphaticum* produced acids that enhanced the availability of phosphates and increased the uptake of other nutrients, leading to increased yields (Cakmakci et al., 1999; Saxena et al., 2020; Plaza et al., 2021).

With respect to solubilizing agents, there are slight significant difference among solubilizing agents in the absence of bacteria. However, when solubilizing bacteria was added the effect follows the sequence: EDTA > citric acid > HA > compost in case of OSP and RP, but the effect was almost the same when BS was used. This means there are factors other than P in BS contributed to the production of seeds yield. It is possible that the presence of appreciable amounts of Mg, S, Fe, Mn, B and other elements played a role in plant growth, although P percentage was relatively low compared to RP and OSP (Yildirim & Prezzi, 2011).

Regarding the interaction among the studied treatments, the treatment of HA+ EDTA gave the highest seeds yield, recording 8.57 g plant⁻¹ without adding bacteria. While recorded 11.5 g plant⁻¹ in the presence of added bacteria. Similar trend was found with the other yield parameters, with high significant difference in the presence of P- dissolving bacteria. Number of pods plant⁻¹ varied from 1 in control treatment (without any additions) to 6.67 that recorded by many treatments, OSP in combination with citric acid or EDTA and the treatment of HA combined with EDTA, in the presence of P- dissolving bacteria. Regarding the fresh weight of pods, ranged from 2.59 g plant⁻¹ (in control treatment, without any additions) to 22.3 g plant⁻¹ with applying the treatment of HA combined with EDTA, in the presence of P- dissolving bacteria (Table 6).

3.3.4 N, P and K concentrations in faba bean leaves

Results of P concentration in leaves of faba bean plants (Table 7) indicated that by addition of P sources, P contents increased with remarkable increase with applying OSP treatment than applying RP or BS treatments. This was found in case of without adding or with adding solubilizing bacteria, but the values were

generally higher with the later. Elhag et al. (2019) reported that, increasing available P in soil by addition of P sources was reflected on increasing P concentration in bean roots and shoots, with high concentrations in shoots more than roots.

However, no remarkable difference in concentration among solubilizing agents, except when EDTA or citric acid was added with BS. This again clearly indicate the role of these relatively small compounds in solubilizing not only insoluble P, but also other elements as Mg, Fe, Mn in BS which may activate plant roots to absorb nutrients. The enhanced role of EDTA or citric acid in calcareous soils is not only due to acidification of the plant rhizosphere, but also to its Ca and Mg complexing capacity (Drouillon & Merckx, 2003; Hamed & Gamal, 2014). Mihoub et al. (2019) found that P uptake by wheat plants grown in alkaline calcareous soil was 0.493 mg P pot⁻¹ in the control treatment, however, it reached 0.701 and 0.785 mg P pot⁻¹ in the amended pots with pigeon manure juice and citric acid, respectively.

Regarding the interaction between solubilizing agents, the treatment of HA+ EDTA, followed by that plus citric acid gave the highest concentration of P in plant leaves, with significant difference as compared to the other treatments. Sahin et al. (2014) found that humic substances in interaction with P in the soil could decrease the P- fixation and increase the P- uptake by plants.

With respect to the effect of the studied treatments on N and K concentrations in plant leaves (Table 7), it was clear that the addition of P increased N and K content in all treatments. Also, there was a remarkable difference recorded with respect to the solubilizing agents and bacterial additions. Although HA and compost enhanced the plant uptake from N and K, due to their considerable content of the macro elements; the treatments of EDTA and citric acid were superior in increasing plant uptake of both. These may be due to enhancing root efficiency in absorbing nutrients from soil and added fertilizers, lowering soil pH, and their high Ca and Mg complexing capacity (Drouillon & Merckx, 2003; Hamed & Gamal, 2014).

The main problem of the investigated soil is its high content of CaCO₃ (36.5 %, Table 2), and this is influence on nutrients availability and uptake by plants. So, the soil moisture content should be kept up to field capacity till the end of the experimental work. Also, the role of EDTA and citric acid in enhancing root growth and absorption, besides their interaction with CaCO₃ in soil as well as their effects on lowering soil pH; reflected on enhancing plant growth and productivity, as compared to the effect of compost or HA (Campitelli et al., 2003; Drouillon & Merckx, 2003). The interac-

Table 6: Numbers of pods plant⁻¹, fresh mass of pods plant⁻¹ and fresh mass of faba bean seeds plant⁻¹ cultivated on El-Nubaria calcareous soil as affected by the different P sources, in the presence or absence of P- dissolving bacteria

Treatments	No. of pods plant ⁻¹		F. mass of pods (g plant ⁻¹)		F. mass of seeds (g plant ⁻¹)	
	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**
Control (-P)	1.00 ± 0.03k	2.00 ± 0.05l	2.59 ± 0.10r	5.93 ± 0.33t	1.21 ± 0.05q	2.58 ± 0.09p
OSP	3.00 ± 0.07g	5.00 ± 0.27e	7.42 ± 0.50m	11.6 ± 0.90n	4.19 ± 0.12k	6.43 ± 0.39i
OSP + Compost 1 %	3.67 ± 0.10e	6.00 ± 0.30c	8.23 ± 0.62j	13.7 ± 0.98j	4.56 ± 0.18h	7.61 ± 0.47g
OSP + Humic Acid 1 %	4.00 ± 0.10d	6.33 ± 0.30b	8.55 ± 0.65i	15.7 ± 1.02h	4.72 ± 0.20g	8.06 ± 0.49f
OSP + Citric Acid 1 %	4.33 ± 0.13c	6.67 ± 0.33a	9.72 ± 0.70h	17.8 ± 1.13f	5.23 ± 0.24e	9.78 ± 0.56d
OSP + EDTA 1 %	4.67 ± 0.17b	6.67 ± 0.35a	9.84 ± 0.74g	19.9 ± 1.29c	5.50 ± 0.27d	10.1 ± 0.70c
RP	2.33 ± 0.05i	4.00 ± 0.25h	6.87 ± 0.43o	10.1 ± 0.81q	3.53 ± 0.08o	5.11 ± 0.21o
RP + Compost 1 %	2.67 ± 0.07h	4.33 ± 0.30g	6.92 ± 0.47o	10.3 ± 0.82q	3.87 ± 0.09m	5.56 ± 0.23m
RP + Humic Acid 1 %	3.00 ± 0.07g	4.67 ± 0.30f	7.61 ± 0.52l	12.6 ± 0.86l	4.31 ± 0.14j	6.53 ± 0.30i
RP + Citric Acid 1 %	3.67 ± 0.10e	5.00 ± 0.33e	7.98 ± 0.53jk	13.6 ± 0.94j	4.43 ± 0.16i	7.32 ± 0.43h
RP + EDTA 1 %	3.67 ± 0.10e	5.33 ± 0.35d	9.76 ± 0.73gh	14.8 ± 1.05i	5.42 ± 0.21d	8.12 ± 0.49f
BS	2.00 ± 0.05j	3.00 ± 0.20k	6.10 ± 0.43q	9.38 ± 0.70s	3.51 ± 0.08o	5.17 ± 0.21n
BS + Compost 1 %	2.67 ± 0.07h	3.67 ± 0.25i	6.55 ± 0.45p	10.1 ± 0.82q	3.75 ± 0.10n	5.15 ± 0.20n
BS + Humic Acid 1 %	3.00 ± 0.10g	4.00 ± 0.30h	7.27 ± 0.57n	11.5 ± 0.93n	4.07 ± 0.13l	6.20 ± 0.33k
BS + Citric Acid 1 %	3.00 ± 0.07g	4.33 ± 0.35g	7.88 ± 0.56k	12.2 ± 0.96m	4.39 ± 0.15ij	6.41 ± 0.35ij
BS + EDTA 1 %	3.67 ± 0.13e	4.67 ± 0.30f	8.11 ± 0.60j	12.5 ± 0.97l	4.58 ± 0.16h	7.30 ± 0.41h
OSP+ RP+ BS (50 % for everyone)	4.33 ± 0.20c	6.00 ± 0.37c	9.76 ± 0.74gh	13.1 ± 1.04k	5.45 ± 0.25d	7.60 ± 0.49g
Compost 1 %	2.33 ± 0.07i	3.33 ± 0.17j	6.50 ± 0.45p	9.81 ± 0.72r	3.23 ± 0.10p	5.13 ± 0.24no
Humic Acid 1 %	2.67 ± 0.10h	4.00 ± 0.30h	6.67 ± 0.46p	10.3 ± 0.85q	3.86 ± 0.12m	5.47 ± 0.27m
Citric Acid 1 %	3.00 ± 0.07g	4.33 ± 0.30g	7.01 ± 0.49o	10.7 ± 0.87p	3.98 ± 0.15l	5.83 ± 0.30l
EDTA 1 %	3.33 ± 0.15f	4.33 ± 0.25g	7.29 ± 0.51mn	11.1 ± 0.95o	4.03 ± 0.15l	6.25 ± 0.38ijk
Compost 1 %+ Humic Acid 1 % (50 % for both)	4.33 ± 0.20c	5.00 ± 0.23e	10.6 ± 0.78d	16.0 ± 1.15g	5.27 ± 0.23e	8.53 ± 0.46e
Citric Acid 1 %+ EDTA 1 % (50 % for both)	5.00 ± 0.27a	6.00 ± 0.27c	13.7 ± 0.99b	19.5 ± 1.28d	7.15 ± 0.45b	10.3 ± 0.53c
Compost 1 %+ Citric Acid 1 % (50 % for both)	4.00 ± 0.20d	6.00 ± 0.30c	10.1 ± 0.80f	18.8 ± 1.16e	5.08 ± 0.28f	9.51 ± 0.49d
Compost 1 %+ EDTA 1 % (50 % for both)	4.00 ± 0.23d	6.33 ± 0.30b	10.4 ± 0.87e	19.7 ± 1.27cd	5.31 ± 0.29e	10.2 ± 0.54c
Humic Acid 1 %+ Citric Acid 1 % (50 % for both)	4.67 ± 0.25b	6.33 ± 0.35b	12.2 ± 0.91c	21.0 ± 1.33b	6.94 ± 0.32c	11.2 ± 0.63b
Humic Acid 1 %+ EDTA 1 % (50 % for both)	5.00 ± 0.27a	6.67 ± 0.37a	16.8 ± 1.12a	22.3 ± 1.35a	8.57 ± 0.50a	11.5 ± 0.64a

*(-PDB) means without adding P dissolving bacteria, **(+PDB) means in the presence of P dissolving bacteria. Ordinary Superphosphate (OSP), Rock Phosphate (RP), Basic Slag (BS). Values expressed as mean ± SE, the significant value was set at $p \leq 0.05$. Different letters indicate significant difference between treatments

Table 7: N, P and K concentrations in faba bean leaves cultivated on El-Nubaria calcareous soil as affected by the different P sources, in the presence or absence of P-dissolving bacteria

Treatments	N, %		P, %		K, %	
	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**	(-PDB)*	(+PDB)**
Control (-P)	2.10 ± 0.07s	2.34 ± 0.09t	0.16 ± 0.02m	0.22 ± 0.03p	1.69 ± 0.08r	1.95 ± 0.08v
OSP	2.39 ± 0.09o	2.81 ± 0.12o	0.27 ± 0.05h	0.34 ± 0.09j	2.04 ± 0.10kl	2.16 ± 0.12q
OSP + Compost 1 %	2.43 ± 0.09jkl	2.92 ± 0.14l	0.30 ± 0.07e	0.37 ± 0.09g	2.13 ± 0.10i	2.27 ± 0.23jkl
OSP + Humic Acid 1 %	2.49 ± 0.10h	2.97 ± 0.15hi	0.33 ± 0.07c	0.39 ± 0.08e	2.17 ± 0.12g	2.31 ± 0.22i
OSP + Citric Acid 1 %	2.65 ± 0.12c	3.11 ± 0.18f	0.37 ± 0.08b	0.42 ± 0.10c	2.21 ± 0.15e	2.39 ± 0.28g
OSP + EDTA 1 %	2.72 ± 0.13b	3.26 ± 0.21e	0.39 ± 0.08a	0.46 ± 0.14a	2.30 ± 0.19b	2.45 ± 0.30f
RP	2.31 ± 0.09q	2.74 ± 0.10r	0.24 ± 0.04j	0.30 ± 0.06m	1.92 ± 0.09p	2.11 ± 0.09s
RP + Compost 1 %	2.40 ± 0.10no	2.84 ± 0.10n	0.27 ± 0.05h	0.32 ± 0.06l	2.03 ± 0.09l	2.18 ± 0.10op
RP + Humic Acid 1 %	2.46 ± 0.10i	2.88 ± 0.12m	0.28 ± 0.05g	0.35 ± 0.06i	2.12 ± 0.10ij	2.25 ± 0.18l
RP + Citric Acid 1 %	2.50 ± 0.12gh	2.96 ± 0.13ij	0.30 ± 0.07e	0.37 ± 0.07g	2.17 ± 0.13g	2.28 ± 0.20j
RP + EDTA 1 %	2.59 ± 0.13d	3.09 ± 0.15g	0.31 ± 0.07d	0.40 ± 0.07d	2.25 ± 0.18c	2.32 ± 0.21i
BS	2.28 ± 0.08r	2.69 ± 0.10s	0.21 ± 0.03l	0.26 ± 0.04o	1.87 ± 0.08q	2.08 ± 0.10t
BS + Compost 1 %	2.36 ± 0.09p	2.75 ± 0.11qr	0.24 ± 0.04j	0.30 ± 0.07m	1.97 ± 0.09n	2.14 ± 0.11r
BS + Humic Acid 1 %	2.41 ± 0.10mn	2.76 ± 0.11q	0.26 ± 0.06i	0.33 ± 0.07k	2.01 ± 0.09m	2.19 ± 0.11o
BS + Citric Acid 1 %	2.45 ± 0.10ij	2.87 ± 0.12m	0.28 ± 0.07g	0.36 ± 0.08h	2.13 ± 0.11i	2.23 ± 0.15m
BS + EDTA 1 %	2.55 ± 0.12e	2.98 ± 0.14h	0.29 ± 0.07f	0.39 ± 0.09e	2.21 ± 0.16e	2.26 ± 0.15kl
OSP+ RP+ BS (50 % for everyone)	2.35 ± 0.08p	2.83 ± 0.12n	0.30 ± 0.08e	0.35 ± 0.06i	2.05 ± 0.10k	2.05 ± 0.11u
Compost 1 %	2.32 ± 0.08q	2.70 ± 0.11s	0.22 ± 0.04k	0.28 ± 0.03n	1.94 ± 0.09o	2.10 ± 0.12t
Humic Acid 1 %	2.39 ± 0.09o	2.74 ± 0.11r	0.24 ± 0.04j	0.29 ± 0.03n	1.98 ± 0.09n	2.16 ± 0.14q
Citric Acid 1 %	2.42 ± 0.11lm	2.81 ± 0.12o	0.27 ± 0.05h	0.32 ± 0.07l	2.11 ± 0.012j	2.17 ± 0.15pq
EDTA 1 %	2.51 ± 0.12fg	2.94 ± 0.15k	0.28 ± 0.07g	0.34 ± 0.08j	2.16 ± 0.13gh	2.21 ± 0.18n
Compost 1 %+ Humic Acid 1 % (50 % for both)	2.43 ± 0.12jkl	2.79 ± 0.12p	0.26 ± 0.06i	0.32 ± 0.07l	2.15 ± 0.15h	2.36 ± 0.23h
Citric Acid 1 %+ EDTA 1 % (50 % for both)	2.55 ± 0.13e	3.28 ± 0.19d	0.28 ± 0.08g	0.38 ± 0.08f	2.23 ± 0.19d	2.48 ± 0.26e
Compost 1 %+ Citric Acid 1 % (50 % for both)	2.52 ± 0.14f	3.27 ± 0.20de	0.27 ± 0.06h	0.39 ± 0.09e	2.19 ± 0.17f	2.59 ± 0.27d
Compost 1 %+ EDTA 1 % (50 % for both)	2.56 ± 0.13c	3.36 ± 0.22c	0.28 ± 0.08g	0.40 ± 0.10d	2.21 ± 0.19e	2.67 ± 0.30c
Humic Acid 1 %+ Citric Acid 1 % (50 % for both)	2.74 ± 0.18b	3.45 ± 0.25b	0.28 ± 0.09g	0.43 ± 0.11b	2.29 ± 0.21b	2.81 ± 0.30b
Humic Acid 1 %+ EDTA 1 % (50 % for both)	2.90 ± 0.19a	3.58 ± 0.26a	0.30 ± 0.08e	0.46 ± 0.12a	2.34 ± 0.25a	2.94 ± 0.33a

*(-PDB) means without adding P dissolving bacteria, **(+PDB) means in the presence of P dissolving bacteria. Ordinary Superphosphate (OSP), Rock Phosphate (RP), Basic Slag (BS). Values expressed as mean ± SE, the significant value was set at $p \leq 0.05$. Different letters indicate significant difference between treatments

tion among the studied treatments gave better results, especially between chelating agents and organic compounds. In addition, the P dissolving bacteria produce organic and inorganic acids that mobilize P and other nutrients and encourage plant growth, as well as releasing phosphatase enzymes to mineralize organic P (Illmer et al., 1995; Cakmakci et al., 1999; Amalraj et al., 2012; Płaza et al., 2021).

4 CONCLUSION

Many factors are affecting the solubility of P in calcareous soils. In our study we tried to use some sources of P as well as organic substances and chelating agents, the interaction between them being the best. BS gave promising results especially when combined with citric acid and EDTA not only in calcareous soil, but possibly in soils poor in nutrient elements as sandy soils, even under the alkaline conditions. Using BS gained many benefits such as recycling of wastes, protecting the environment from contamination, and being a source of P fertilizer. Also, application of citric acid and EDTA enhanced faba bean growth in the investigated soil, particularly with addition of organic substances. In addition, adding solubilizing bacteria played a major role than the P- sources in enhancing the availability of P and increasing the uptake of other nutrients, leading to increased yield.

5 RECOMMENDATIONS

The use of RP as a source of P in calcareous soil at the time of applying the organic and bio fertilizers and close to plant roots is so beneficial. Applying RP or BS during the preparation of compost will enrich the compost with P. Application of BS along with organic fertilizers and chelating agents can be recommended in low fertile soil as sandy soil.

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