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Effectiveness of Rock phosphate and liquid Bio-fertilizer on fruit quality and quantity of pomegranate trees Wonderful cv.

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ABSTRACT

Rock phosphate is one of the natural rocks present in the earth's crust, which is characterized by its high content of phosphorous. Rock phosphate has been used as a source of fertilization with phosphorous as a substitute for chemical fertilizers that may have significant damage to the soil, groundwater and the environment in the short and long term. Pomegranate is a tree of great economic importance. It also has great health and nutritional benefits, which increases the importance of conducting studies on it to produce high quality fruits. The aim of the research is to study the use of rock phosphate with phosphorous-facilitating bacteria as a substitute for traditional phosphate fertilizers in order to reach the best treatment that gives the highest amount of yield and physical and chemical characteristics of pomegranate fruits. The present investigation was carried out in two consecutive seasons (2021 and 2022) on seven-year-old pomegranate trees "Wonderful" cv., planted at 3×5m apart around (420 trees / Fed) grown under saran shading net and drip irrigation system was used. Trees irrigated with drip irrigation system, at a private orchard. The experiment included 3 rates of rock phosphate 1, 2, and 3 kg/ tree + phosphorus and sulfur dissolving bacteria at two times. The effect of different treatments on fruit yield, physical and chemical characteristics of pomegranate trees were evaluated. In this study, the most treatments gave more or less physical and chemical results similar to the control except, trees received 3kg rock phosphate + phosphorus and sulfur dissolving bacteria at two times, which increased yield of seed (kg)/tree, yield of juice (L)/tree and improved quality of pomegranate fruits "Wonderful" cv. comparing with all treatments including the control.

Keywords: Pomegranate, Wonderful cv., rock Phosphate, bio-fertilizer, fruit quality, yield and mineral content.

1. Introduction

The cultivation of pomegranate trees (*Punica granatum* L.) has spread for thousands of years and following to the (Family: punicaceae), and the meaning of the name pomegranate in the Latin language is "seeded apple", and it is considered a small tree or shrub with deciduous leaves (Lansky and Newman 2007). the pomegranate fruit is a berry fruit and it has been cultivated in the subtropical, tropical and Mediterranean regions since ancient times, and among the most important varieties of pomegranate are Almnfouti, Sukkari, Wonderful and Hamid. Pomegranate juice is considered one of the best types of healthy juices, and it is varied, including sweet, sour, and the dark red color of the seeds as a result of the high content of anthocyanins. The fruits may be marketed fresh, intact, with lobed seeds, or in the form of juice (Roy and Waskar, 1997).

Phosphorus is considered one of the most important nutrients necessary for plants and has many physiological roles, including that it is involved in the formation of phospholipids, the formation of DNA and NADP, as well as ATP, and it is an activator of enzymes that produce amino acids. Phosphorus also participates in many vital processes within plants such as respiration, photosynthesis, fatty acid synthesis, seed germination, stimulation of seed and root formation, flowering and bud formation, and acceleration of fruit ripening (Espinnosa *et al.,* 1993).

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Rock phosphate is one of the most important natural rocks, which is a basic source for the manufacture of phosphate fertilizers. It is a sedimentary rock that contains many minerals, including apatite. Rock phosphate is a safe fertilizer and a natural source of phosphorus (Reddy *et al.,* 2002).

It was observed that rock phosphate is well suited for acidic soils, as the low pH of the soil increases the speed of decomposition of rock phosphate, but when rock phosphate is added to alkaline and calcareous soils, it does not easily decompose due to the high pH of the soil (Caravaca *et al.*, 2004). Therefore, bacteria that decompose rock phosphate must be added to alkaline and calcareous soils to increase the efficiency and speed of decomposition. It has been observed that the use of organic fertilizers and bacteria with rock phosphate in alkaline and calcareous soils improved the chemical, physical and biological properties of the soil (Adhami *et al.,* 2014 and Lim *et al.,* 2015). Also, this method has a positive effect on crop productivity because organic fertilizers contain many macro and microelements (Kalaivanan and Hattab, 2016).

Rock phosphate contains, in addition to phosphorous, many of the nutrients that are necessary for high plant growth and productivity, Haque and Lupmayi (1999) on *Trifolium tembense*, Sastry *et al.*, (1997) on Chrysanthemum cinerariaefolium, noticed that, rock phosphate is a natural, economical and efficient source of phosphorous fertilization. Grham and Timmer (1985) reported that, the use of superphosphate in the soil leads to the loss of the phosphorus element without the benefit of the plants, compared to rock phosphate, which supplies the plants with the phosphorus element on a regular basis without losing phosphorus with the waste water. Rock phosphate can also be used in a crushed form, which gives a better result (El-Sayed, 2006). On the same side, El- Sayed *et al.* (2009) on Tarragon (*Artemisia dracunculus* L.) reported that, the use of Egyptian rock phosphate stimulated the growth characteristics of plants such as fresh weight, dry weight and plant height.

Naturally growing soil bacteria that capable of stimulating plant growth named as Plant-Growth-Promoting-Rhizobacteria (PGPR) such as Bacillus spp. (Kloepper and Schorth, 1981). PGPR is able to positively influence the growth and productivity of plants. Phosphate-dissolving bacteria (PDB) are important for supplying the plant with phosphorus by dissolving the existing phosphorus in a precipitated and complex form and converting it into a form that is easily absorbed by plant roots, and depends on its effect on the production of acids, whether organic or inorganic, or carbon dioxide. Therefore, phosphorus-dissolving bacteria can be added to the soil at appropriate rates to improve soil biological activity (Zayed, 1998). Moreover, Chakrabotry *et al.* (2006) reported that, *Bacillus megaterium* from tea rhizosphore is able to produce IAA and thus it helps plant growth promotion. Regarding the interaction between phosphorus fertilizer and PDB. Ahmed *et al.* (2002) and Badran (2003) noticed that, P fertilizer and PDB treatment led to higher nutrient concentrations in seed and an increment in seed yield of soybean and Lentil plants. Also, Mekhemar *et al.* (2007) found that, improvement of peanut yield could be achieved by soil application of 16 kg P_2O_5/fed as rock phosphate in addition to the inoculation with Brady rhizobium + PDB) under sandy soil conditions.

Therefore, the targets of this study to determine the effect of Egyptian rock phosphate as a source of phosphorus (natural fertilizer) and phosphate dissolving bacteria on the growth, leaf mineral content and yield of pomegranate trees Wonderful cv.

2. Materials and Methods

The present investigation was carried out in two consecutive seasons (2021 and 2022) on seven-year-old pomegranate trees "Wonderful" cv., planted at 3×5m apart around (420 trees / Fed) grown under saran shading net and drip irrigation system was used. Trees irrigated with drip irrigation system, at a private orchard "Hegazi" farm located at 57 km. from Cairo on the road to Alex., Egypt. The orchard soil texture was sandy loam, the soil and water were analyzed according to Wilde *et al.* (1979) as presented in Table (1) and Table (2). To investigate this experiment, twenty-eight trees were selected as mostly uniform in vigorous growth, healthy, fruitful, no visual nutrient deficiency symptoms and were subjected to the same agriculture practices adopted in the farm program.

Rock phosphate (R.P) was added by rate (1, 2 or 3 kg per tree / year), Bio-fertilizers (B) was added by rate 2 Kg compost (2:3 % N) + 0.5 kg agricultural sulfur per tree for all treatments. All applications added by digging and burying in the soil at the last week of December.

Biofertilizers (Microbial cultures) consisted of liquid cultures of two bacteria; *Bacillus megaterium* var. *phosphaticum* and *Acidithia bacillus thiooxidanse*, kindly provided by the microbiology department, agricultural and biological institute, National Research Centre. Each

organism was grown separately in batch culture to the late exponential phase of each microorganism. Cultures were mixed by ratio 2 (*Bacillus megaterium* var. *phosphaticum*): 1 (*Acidithia bacillus thiooxidanse*) on site then each tree received 20 ml of the mix, and this dose was repeated for two or three times (January, March and May) during the season.

The experiment including the following treatments:
T1: Untreated (control)
T2: 1kg rock phosphate + bacteria at two times
T3: 1kg rock phosphate + bacteria at three times
T4: 2kg rock phosphate + bacteria at two times
T5: 2kg rock phosphate + bacteria at three times
T6: 3kg rock phosphate + bacteria at two times
T7: 3kg rock phosphate + bacteria at three times

Table 1: Physical and chemical analysis of soil

Soil characteristics	Surface sample	30 cm depth	60 cm depth
	Particle size distribution %		
Sand (%)	96.17	94.73	93.03
Silt %	1.51	3.11	3.58
Clay %	2.32	2.16	3.39
Soil texture		Sandy	
	Chemical characteristics		
pH	7.81	7.77	7.48
EC(dsm^{-1})	2.79	2.55	2663
	Soluble anions (meq / 100g soil)		
$CO_3^{=}$	-	-	-
HCO_3^{-}	1.00	0.70	0.90
Cl^{-}	22.3	21.80	22.00
$SO_4^{=}$	4.62	3.06	3.42
	Soluble cations (meq / 100g soil)		
Ca^{++}	4.50	3.00	3.70
Mg^{++}	1.00	1.10	1.20
Na^{+}	22.12	20.13	20.41
K^{+}	0.30	1.33	1.01

Table 2: Chemical characteristics of water

Parameters	Values
pH	8.40
EC(dSm^{-1})	1.19
Soluble cations (meq\l)	
Ca^{++}	1.50
Mg^{++}	1.12
Na^{+}	8.45
K^{+}	0.89
Soluble anions (meq\l)	
$CO_3^{=}$	-
HCO_3^{-}	1.40
Cl^{-}	6.46
$SO_4^{=}$	4.10

2.1. Measurements

2.1.1. Yield

At maturity on the first week of October in each season the average number of fruits / trees was counted. Twenty fruits from each tree (replicate) were harvested to get the average fruit weight of each treatment. Such average was multiplied by the average number of fruits / trees to get the average yield/tree (kg).

2.1.2. Fruit quality

For each season, sample of six fruit / tree were randomly taken for the evaluation of physical and chemical fruit properties: Fruit weight (g), Fruit seed weight (g), Fruit peel weight (g), seed/Peel ratio, Fruit grain %, Fruit peel %, Fruit T.S.S (Brix), MI (T.S.S/ Acidity of Fruit), Fruit juice content (ml/100gm), Fruit Ascorbic acide (g/100g), Fruit Anthocyanin (mg/g), yield of grain (kg) / tree, yield of juice (L) / tree.

2.1.3. Leaf mineral content

Dry leaves which collected to determine chlorophyll content and leaf dry matter % were grounded and digested using sulphoric acid and oxygen peroxide to determine N, P, K, Ca, Mg, Fe, Zn and Mn. Nitrogen was determined by the Micro- Kjeldahlmethod, phosphorus was determined by the spectrophotometer, potassium was determined by a flame photometer (Jackson, 1973). Iron, zinc and manganese were estimated by using an atomic absorption according to the method of Cottenie *et al.* (1982).

2.2. Statistical analysis

Analysis of variance was used as mentioned by Snedecor and Cochran, (1980) to analyze the data statistically. Means were differentiated by using Duncan's multiple range tests at 5 % (Duncan, 1955).

Results

Data in Table (3) and (4) revealed that yields (as number and weight of fruits) per tree was affected significantly by Rock Phosphate (R.F) and bacteria application times (B.A.T) pronouncing positive in the second season comparing with the control Highest number of fruits and weight of fruits was obtained from trees received fertilized with 3kg (R.F) and two (B.A.T) at two application times. Average yields number and weight (Kg) per tree increment obtained by (T6) recorded 166 and 41.4 (Kg) comparing with the control that reached 92 and 28 (Kg) per tree respectively, the average yield recorded 72% over than the control.

Table 3: Effect of Rock phosphate and liquid Bio-fertilizer yield, fruit weight and number of pomegranate trees Wonderful cv.in 2021 and 2022 seasons.

Treatments	No. fruit /tree		Fruit weight (g)		Yield (kg)/tree	
	2021	2022	2021	2022	2021	2022
Untreated (control)	67 b	58 c	456.67 d	463.33 c	30.75 b	25.34 d
1kg rock phosphate + bacteria at two times	43 e	63 bc	512.67 c	392.99 e	27.00 c	24.24 d
1kg rock phosphate + bacteria at three times	42 e	65 b	643.83 a	432.68 d	26.85 c	29.21 c
2kg rock phosphate + bacteria at two times	45 d	64 bc	565.67 b	566.67 a	25.64 c	37.09 b
2kg rock phosphate + bacteria at three times	53 c	66 b	474.5 c	426.67 d	25.14 c	30.90 c
3kg rock phosphate + bacteria at two times	82 a	84 a	519.5 c	458.33 c	42.60 a	40.23 a
3kg rock phosphate + bacteria at three times	42 e	66 b	505.83 d	506.67 b	20.55 d	35.16 b

Mean in each column with similar letter(s) are not significantly different at 5 % level.

Table 4: Effect of Rock phosphate and liquid Bio-fertilizer yield of grain and juice of pomegranate trees Wonderful cv.in 2021 and 2022 seasons.

Treatments	Yield of grain (kg) / tree			Yield of juice (L) / tree		
	2021	2022	Average	2021	2022	Average
Untreated (control)	15.98 b	14.5 d	15.24 b	11.84 b	10.37 b	11.11 b
1kg rock phosphate + bacteria at two times	10.36 e	11.52 f	10.94 f	6.78 d	7.29 d	7.04 e
1kg rock phosphate + bacteria at three times	11.73 cd	16.92 ab	14.33 c	7.07 d	10.19 b	8.63 cd
2kg rock phosphate + bacteria at two times	11.46 d	16.31 bc	13.89 c	7.58 c	10.33 b	8.96 c
2kg rock phosphate + bacteria at three times	12.28c	13.6 e	12.94 d	7.94 c	8.53 c	8.24 d
3kg rock phosphate + bacteria at two times	18.74 a	17.43 a	18.09 a	12.35 a	11.19 a	11.77 a
3kg rock phosphate + bacteria at three times	10.44 e	15.89 cd	13.17 d	7.07 d	10.53 b	8.80 c

Mean in each column with similar letter(s) are not significantly different at 5 % level.

Obtained results in table (5) cleared that all treatments increased fruit peel % than the control in both studies seasons fruit peel % ranged between 39.84& 46.04 and 47.79 & 56.01 in the first and second season respectively.

Fruit seed % results varied from season to another in the first season fruit seed percentages ranged between44.96% and 60.16% Highest Percentage was in treatment (5) Fruit samples However, in the second season fruit seed weight % in untreated trees (Control) was the highest (52.21%) comparing with all other that treatment which recorded seed weight fruit % ranged between 43.36 and 49.16.

Results in Table (5) indicated that all treatments deceased seed/peel ratio values, which ranged between 0.97 and 0.74 Comparing with the Control which recorded 1.09 as seed/peel ratio similar trend was Observed in the second season except T3 (1 kg R.P + B) at three times (resulted seed/peel ratio higher than all treatments including the control. This may be referred to higher fruit seed weight and lower peel weight in the second season.

Table 5: Effect of Rock phosphate and liquid Bio-fertilizer on fruit seed and peel of pomegranate trees Wonderful cv. in 2021 and 2022 seasons.

Treatments	Fruit seed (g)		Fruit peel (g)		Seed /Peel ratio	
	2021	2022	2021	2022	2021	2022
Untreated (control)	238.48 bcd	249.99 ab	218.19 f	213.34 d	1.09 a	1.17 b
1kg rock phosphate + bacteria at two times	240.86 bcd	182.84 d	271.80 d	210.15 d	0.89 c	0.87 cd
1kg rock phosphate + bacteria at three times	279.29 a	260.30 a	364.55 a	172.38 e	0.77 d	1.51 a
2kg rock phosphate + bacteria at two times	254.61 b	254.77 a	311.0 b	311.9 a	0.82 d	0.82 d
2kg rock phosphate + bacteria at three times	231.64 cd	206.11 c	242.86 e	220.55 d	0.95 b	0.93 c
3kg rock phosphate + bacteria at two times	228.54 d	207.46 c	290.96 c	250.88 c	0.79 d	0.83 d
3kg rock phosphate + bacteria at three times	248.63 bc	240.81 b	257.21 de	265.86 b	0.97 b	0.91 c

Mean in each column with similar letter(s) are not significantly different at 5 % level.

Fruit chemical Characteristics, T.S.S, acidity as shown in table (6) were Influenced by different treatment comparing with the control in this respect T.S.S % as well T.S.S/Acidity values. Resulted from control showed high T.S.S and T.S.S/Acidity ratio than other treatments this may most treatments Except, T7 which gave T.S.S and T.S.S/Acidity. Similar to those of the control specially in the second season on the control most treatments Most treatments showed higher Acidity % than the control in this was clearly noticed in the first season from the obtained results it seems that low rates of soil application of rock phosphate and bacteria number of time application delayed fruit maturity.

Table 6: Effect of Rock phosphate and liquid Bio-fertilizer fruit T.S.S and acidity of pomegranate trees Wonderful cv.in 2021 and 2022 seasons.

Treatments	T.S.S (Brix)		Acidity %		MI (T.S.S/ Acidity)	
	2021	2022	2021	2022	2021	2022
Untreated (control)	16.5 a	17.0 a	1.14 d	1.50 d	14.44 a	11.33 a
1kg rock phosphate + bacteria at two times	13.4 c	14.9 b	1.68 a	1.70 a	8.01 e	8.76 e
1kg rock phosphate + bacteria at three times	11.5 d	15 b	1.73 a	1.66 ab	6.67 f	9.04 e
2kg rock phosphate + bacteria at two times	14.1 c	15.2 b	1.45 c	1.51 d	9.73 cd	10.07 c
2kg rock phosphate + bacteria at three times	15.1 b	16.3 a	1.48 c	1.55 cd	10.20 c	10.52 bc
3kg rock phosphate + bacteria at two times	14.8 b	15.5 b	1.59 b	1.62 bc	9.28 d	9.57 d
3kg rock phosphate + bacteria at three times	16.2 a	16.9 a	1.44 c	1.56 cd	11.23 b	10.83 ab

Mean in each column with similar letter(s) are not significantly different at 5 % level.

Results in Table (7) indicated that Juice content decreased significantly by rock phosphate and bio fertilizer application times juice contain studied treatments ranged between 60. 23 and 60.24 ml/ 100 gm. comparing with those of the control that gave 74.07 and 71.52 in the first and second seasons respectively.

Ascorbic acid g/100 gm tended to increase with increasing rock phosphate application dose as well as number of bio fertilizer application times trend, was obviously noticed when comparing between T2& T3, T4&T5 and T6& T7 where ascorbic acid g/100gn recorded the following values 11.20, 15.85& 14.69, 17.65 followed in an increasing order by 15.28, 19.66 & 16.86, 18.65 as well as 18.77, 21.85, 20.44, 22.02 in the first and second season respectively. This means the dose of rock phosphate and bio fertilizer application times, the higher ascorbic acid. Results indicated that Rock phosphate at the high rate 3 kg + bacteria application at 2 or 3 times significantly increased Ascorbic acid (T6 &T7) comparing with other treatments.

Anthocyanin (mg/gm) as shown in Table (7) was affected significantly by different treatment. However, their values ranged between 1.03 & 1.33 and 1.78 & 2.54 in the first and second seasons respectively. No particular trend control noticed except under medina rock phosphate application dose (2kg /tree) as well as the high dose (3 kg/tree) Increasing bio fertilizer application times tended to decrease Anthocyanin This may be related to other fruit chemical properties i.e increased T. S. S and T.SS/acid ratio as well as decreased acidity.

Table 7: Effect of Rock phosphate and liquid Bio-fertilizer fruit juice content, ascorbic acide and anthocyanin of pomegranate trees Wonderful cv.in 2021 and 2022 seasons.

Treatments	Juice content (ml/100gm)		Ascorbic acid (g/100g)		Anthocyanin (mg/g)	
	2021	2022	2021	2022	2021	2022
Untreated (control)	74.07 a	71.52 a	15.40 d	17.98 cd	1.49 d	2.08 cd
1kg rock phosphate + bacteria at two times	65.45 c	63.25 bcd	11.20 f	15.85 e	1.03 f	1.78 e
1kg rock phosphate + bacteria at three times	60.23 d	60.24 d	14.69 e	17.65 d	1.85 ab	1.96 de
2kg rock phosphate + bacteria at two times	66.18 bc	63.35 bcd	15.28 de	19.06 b	1.67 c	2.54 a
2kg rock phosphate + bacteria at three times	64.65 c	62.69 cd	16.86 c	18.65 bc	1.33 e	2.21 b
3kg rock phosphate + bacteria at two times	65.89 c	64.23 bc	18.77 b	21.85 a	1.76 bc	2.19 bc
3kg rock phosphate + bacteria at three times	67.70 b	66.25 b	20.44 a	22.02 a	1.49 d	2.00 cd

Mean in each column with similar letter(s) are not significantly different at 5 % level.

Data in Table (8) cleared that rock phosphate and bio fertilizer treatment increase N, P, K content in the leaves than the control N, P, K content in the control ranged between (1.74,1.62) +(0.22,0.16) and (0.63, 0.62) in the first and second seasons respectively. while However, N. P and K contents % leaves in different treatments ranged between (2.1, 2.9 & 1.9,2.8) for N, (0.29,0.35, 0.32,0.6) for P and (0.66, 0.95) & (0.71, 0.98) for K in 2021 & 2022 seasons respectively. Highest N and P k, Leaf content

was obtained T7 which received the high dose of rock phosphate (3kg/tree) and application times of bio fertilizer (3 times).

From the above results a particular trend on N and P content in leaves was observed that, under any rate of rock phosphate rate increasing number application times of bio fertilizer increase in N and P content in leaves.

In other words, within any application rate of RP either low rate (1 kg) , medium rate (2 kg) or high rate (3kg) per tree , increasing bio fertilizer application from two to three times, Significantly increased N and P comparing the lower one.

Table 8: Effect of Rock phosphate and liquid Biofertilizer nitrogen, phosphor and ptasium leaf content of pomegranate trees Wonderful cv.in 2021 and 2022 seasons.

Treatments	N%		P%		K%	
	2021	2022	2021	2022	2021	2022
Untreated (control)	1.74 g	1.46 e	0.22 f	0.16 g	0.63 d	0.62 f
1kg rock phosphate + bacteria at two times	2.4 cd	2.4 bc	0.29 e	0.32 f	0.69 c	0.76 d
1kg rock phosphate + bacteria at three times	2.5 bc	2.5 b	0.35 d	0.39 e	0.68 c	0.81 c
2kg rock phosphate + bacteria at two times	2.1 e	1.9 e	0.38 c	0.43 d	0.76 b	0.85 b
2kg rock phosphate + bacteria at three times	2.3 d	2.1 d	0.42 b	0.48 c	0.95 a	0.98 a
3kg rock phosphate + bacteria at two times	2.6 b	2.3 c	0.49 b	0.55 b	0.82 b	0.80 c
3kg rock phosphate + bacteria at three times	2.9 a	2.8 a	0.55 a	0.6 a	0.66 cd	0.71e

Mean in each column with similar letter(s) are not significantly different at 5 % level.

Leaf Magnesium and calcium as shown in Table (9) followed the trend of other macro elements N, P, and K Table (8) when comparing with the control. Generally, all treatments increased Leave Magnesium and calcium contents than the control in both studies seasons. in this report Mg % in leaves ranged between (0.21,0.25) & (0.2,0.24) and for Ca % in the leaves ranged between (1.40,1.69) & (1.2,1.75) in 2021 and 2022 respectively. highest Magnesium content in leaves was recorded from T7, meanwhile highest Ca % in the leaves was present, in T6 treatment. This was proved in the two seasons of the experiment.

Table 9: Effect of Rock phosphate and liquid Biofertilizer magnesium and calcium leaf content of pomegranate trees Wonderful cv.in 2021 and 2022 seasons.

Treatments	Mg%		Ca%	
	2021	2022	2021	2022
Untreated (control)	0.21 d	0.20 d	1.40 c	1.20 e
1kg rock phosphate + bacteria at two times	0.24 ab	0.22 bc	1.46 c	1.35 d
1kg rock phosphate + bacteria at three times	0.25 a	0.24 a	1.55 b	1.55 c
2kg rock phosphate + bacteria at two times	0.23 bc	0.21cd	1.60 ab	1.61 bc
2kg rock phosphate + bacteria at three times	0.22 cd	0.23 ab	1.62 ab	1.65 b
3kg rock phosphate + bacteria at two times	0.23 bc	0.23 ab	1.65 a	1.75 a
3kg rock phosphate + bacteria at three times	0.25 a	0.24 a	1.61 ab	1.40 d

Mean in each column with similar letter(s) are not significantly different at 5 % level.

Table (10) cleared that Fe, Mn, Zn, and Cu content in the leaves significantly increased due to rock phosphate and bio fertilizer application treatment comparing with the control in both season Fe % in the leaves ranged between (81,186.3) & (85.2, 135.0) & Mn % ranged between (29, 49) / (35, 49) as for Zn % in the leaves it ranged between (13.0,19.6). Cu % ranged between (3.0, 7.5, 4.5, and 5.0) the above percentages of Fe, Mn, Zn and Cu in the Leaves was recorded in the first and second seasons, respectively.

Table 10: Effect of Rock phosphate and liquid Biofertilizer iron, manganese, zinc and copper leaf content of pomegranate trees Wonderful cv.in 2021 and 2022 seasons.

Treatments	Fe ppm		Mn ppm		Zn ppm		Cu ppm	
	2021	2022	2021	2022	2021	2022	2021	2022
Untreated (control)	81.1 f	85.2 e	29 g	35 e	16.3 e	13.0 d	3.0 f	4.5 f
1kg rock phosphate + bacteria at two times	118.0 e	121.5 b	32 f	39 d	21.6 a	19.6 a	4.8 c	5.4 d
1kg rock phosphate + bacteria at three times	121.5 e	138.0 a	39 d	46 b	19.9 b	18.5 bc	6.0 b	6.0 b
2kg rock phosphate + bacteria at two times	133.0 d	102.6 d	36 e	41 cd	20.3 b	19.3 ab	7.5 a	6.5 a
2kg rock phosphate + bacteria at three times	186.3 a	121.5 b	49 a	43 c	18.5 c	18.5 bc	6.2 b	5.7 c
3kg rock phosphate + bacteria at two times	145.3 c	102.3 d	46 b	49 a	17.6 cd	18.9 ab	4.5 d	5.5 cd
3kg rock phosphate + bacteria at three times	175.6 b	110.7 c	42 c	46 b	17.3 d	17.9 c	4.0 e	5.0 e

Mean in each column with similar letter(s) are not significantly different at 5 % level.

The work included 3 rates of rock phosphate 1, 2, and 3 kg/ tree + phosphorus dissolving bacteria times (January and March) dictated that most treatments gave more or less physical and chemical results similar to the control except, whereas, mineral status in treatments included high rates of Rock phosphate either combined with any number of bio fertilizer used However, macro and micro nutrients in leaves raised from the deficient or low levels.

Discussion

The low amount of phosphorous element in the soil leads to a reduction of productivity and decrease tree growth in general. In addition, in organic farming, the use of single and triple superphosphate is not allowed. Therefore, the use of rock phosphate as a natural source of the phosphate element is the main resort for phosphate fertilization in organic farming, but there is a slow in the decomposition of rock phosphate, especially with the high pH of the soil, so phosphorus and sulfur dissolving bacteria are added to the soil, which increases the speed of decomposition of rock phosphate in the soil and thus increases the growth and productivity of crops. The results are in line with those reported by Andoh-Mensah *et al.,* (2011) who cleared that the use of rock phosphate on mature coconut trees at a rate of 4.5 kg/tree led to a significant increase in the phosphorous content of leaves, as well as increased nut yield of tree and nut weight gain compared to the control, in the acidic soil of Ghana. Similar data were obtained by Salwa *et al*., (2013) who found that the use of rock phosphate at concentrations (115:190 kg / acre) with phosphorus dissolving bacteria led to an increase in vegetative growth, the quantity of the crop, and the improving of the quality of the seeds, and an increase oil content of the seeds of the coriander plant, also application by rock phosphate and bacteria increased microbial activity in the soil, which was reflected in growth and productivity. Similar results were obtained by Kamal (2008) who observed that, additions of phosphorein at 1 kg/ feddan with 60 or 90 kg P2O5 of rock phosphate and gypsum at 4 ton/ feddan improved the growth of pepper plants and increased leaves content of nitrogen, phosphorus and potassium, as well as increased the number of fruits and the average weight of the fruits, as well as improved the quality of the fruits compared to other additions.

Similar results were obtained by Lukiwati (2002) explained that natural rock phosphate is characterized as a source of low-cost phosphorus compared to other sources of phosphate fertilizers, as well as adding rock phosphate to the soil led to an increase in the amount of yield and dry matter of maize compared to untreated plants with P fertilizer. Similar results related rock phosphate (P source) in yield and fruit quality were obtained by (Danso *et al.*, 2010) on oil palm and Hani *et al.,* (2015) on coriander. While, Hellal *et al.,* (2011) on dill, (El-Iraqy 2014) on olives, and Hassan *et al.,* (2010) on Khella presented similar results related the impact of bio-fertilization on fruit yield per feddan as well as Sakr *et al.,* (2014) on *Hibiscus subdariffa* and Rezapour *et al.,* (2011) on *Brassica napus* regard the combination impact between bio-fertilization + rock phosphate (P source) gave the highest most values of measurements.

Conclusions

It could be concluded that trees received 3kg rock phosphate + two phosphorus dissolving bacteria two times (January and March) is the recommended treatment which increased yield of seed (kg) / tree and yield of juice (L). Moreover, the findings of our research could be useful for pomegranate fruit growers for export, fruit juice product where Juice of wonderful pomegranate was rich in vitamin C and Acidity and Anthocyanin, increases the notional value and also suitable for market for export and production as natural product sold or as juice mixture of other fruit juice high notional volume with compatible favorite taste.

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