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THE USE OF PHOSPHATE SOLUBILIZING BACTERIA, PHOSPHATE SOURCES AND ANTIOXIDANTS FOR IMPROVING FRUIT QUALITY OF EARLIGRANDE PEACH TREES

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ABSTRACT

This work was carried out during the two seasons of 2009 and 2010 to study the effect of phosphate solubilizing bacteria (*Enterobacter aerogenes* ECu3, *Bacillus subtilis* AC_2 and *Enterobacter aerogenes* ECu_3×*Bacillus subtilis* AC_2) and some phosphorus resources (rock phosphate, phosphoric acid, pentoxide phosphate and super phosphate) under foliar application of antioxidants (Ascorbic acid, vitamin E, and aqueous extract of *Hibiscus sabdariffa*) on fruit quality of EarliGrande peach trees at private Farm of El-Kharafeen region, North Sinai Governorate, Egypt. Results proved that all physical and chemical fruit parameters of treated trees with *Bacillus subtilis* AC_2 under spraying aqueous extract of *Hibiscus sabdariffa* were significantly increased. All phosphate fertilization sources markedly affected fruit parameters of EarliGrande peach trees. Generally, the highest values of fruit parameters were found with the interaction between phosphoric acid and spraying *H.sabdariffa* extract. As for yield, fruit yield was significantly increased as phosphate solubilizing bacteria were applied. *Bacillus subtilis* AC_2 spraying under aqueous extract of *Hibiscus sabdariffa* treatment recorded the highest yield compared with the remained treatments. At the same time treated trees with pentoxide phosphate and / or phosphoric acid under spraying *H. sabdariffa* and vitamin C extracts produced high values of fruit yield. Whereas, the interaction between *E. aerogenes x B. subtilis* with spraying Vitamin C produced lowest values of acidity and highest values of vitamin C content. *Bacillus subtilis* AC_2 under spraying aqueous extract of *Hibiscus sabdariffa* treatment recorded the highest values of T.S.S and moisture content in both seasons.

Key words: *Prunus persica L.*-phosphate sources- phosphate solubilizing bacteria- antioxidant materials.

INTRODUCTION

Peach tree (*Prunus persica* L.) is one of the most important deciduous fruits grown in Egypt successfully and are widespread in the newly reclaimed areas in Egypt. Most of these soil was considered calcareous. This type of soil has its own problems as an excess of $CaCO_3$ and high pH value that cause a precipitation of mineral contents in an unavailable form for plants. Among of methods followed for improving the quantity and quality of peach trees is the application of major fertilizers to satisfy the needs of plant from such elements, since good growth is mostly associated with good yield. Phosphorus is a very important nutrient for crop growth and high yield with good quality.

It plays a key role in the metabolic process of the conversion of sugar into starch and cellulose. Sandy soils may have some nutrient problems such as less fertility in general and less availability of some elements including phosphorus in case of high pH value.Yield and its components showed a positive response to phosphorus fertilizers. Papplications have increased flower formation, whereas, N was found to reduce it **(Chatzitheodorou *et al.,* 2004).**

Yield and fruit weight of peach fruits increased with increasing the rate of P application up to 700g.tree^{-1} **(Awasthi *et al.,* 1998).** Many soil fungi and bacteria can solubilize inorganic phosphates **(Asea *et al.* 1988; Singh and Sharma 1993)**.

Phosphate solubilizing microorganisms (PSMs) play an important role in supplementing phosphorus to the plants, allowing a sustainable use of phosphate fertilizers **(Gyaneshwar *et al.* 1998; Samah, 2002)**. Application of (PSMs) in the field has been reported to increase mycorrhiza development **(Toro *et al.* 1997 Ivanova *et al.,* 2006)**.The goal of this study was examining the possibility of replacing chemical fertilizers partially by using phosphate solubilizing bacteria and antioxidant applications to improve fruit quality and total yield of EarliGrande peach trees.

MATERIALS AND METHODS

This study was carried out during 2009 and 2010 seasons on seventy-two uniform in vigour of about thirteen years old EarliGrande peach trees budded onto bitter almond rootstock grown in a private peach orchard located at El-Kharafen in Rafah District, North Sinai Governorate, Egypt. The trees were planted at 5 × 5 m apart and growing in sandy soil depending on rainfall irrigation. Analysis of the soil samples are shown in Table (1) according to **(Piper, 1947)**.

The experiment included five phosphate source treatment combinations which were: Control, rock phosphate alone (29% ±5%) (400g.tree^{-1}), phosphoric acid (85%) (150 cm. m^{-3} water), pentoxide phosphate (150 cm. zm^{-3} water) and calcium super phosphate (16–18%) (400g.tree^{-1}).The strains were isolated and identified by **Abd El-Azeem (2006).**

Different Phosphorous sources were sprayed in three stages: the first application was applied after the first contract before flowering, the second was applied during fruit-set, and the third one was applied two weeks before harvest.

While, super phosphate and Rock phosphate were added on the beginning of January (In the winter). In addition, three Phosphate solubilizing bacteria treatments were applied *i.e., Enterobacter aerogenes* ECu_3^+ Rock phosphate, *Bacillus subtilis* AC_2^+Rock phosphate and (*Enterobacter aerogenes* ECu_3 x *Bacillus subtilis* AC_2).All solubilizing bacteria were added (200 ml) with 400 g of rock phosphate per tree.

Three antioxidant applications were sprayed (Ascorbic acid "V.C" about 100-200 ppm, Tocopherol "V.E" about 0.1-0.3 ppm and aqueous extract of *Hibiscus sabdariffa* at concentration about 125 g. litre^{-1}. All antioxidant applications were sprayed three times after fruit set stage and until pre-harvest.

Measurements of fruit quality:

Ten fruits from each treatment were collected and transferred to the laboratory, and used for the following measurements: Fruit size, fruit length, diameter, fruit weight, fruit size and firmness. Fruit juice was obtained individually by pressing the fruit flesh through doubled gauze. Total soluble solid (TSS) concentration was measured using a hand-held refractometer.

The total acidity was calculated by titrating 10 ml of juice with NaOH 0.1

Table (1): Soil physical and chemical analyses at depth (0-45cm) from the soil surface for the peach orchard at Rafah District in North Sinai Governorate, Egypt.

Soil Sample depth (cm)	Physical analysis				E.C ($dS.m^{-1}$)	pH	Chemical analysis							
							Cations ($meq.l^{-1}$)				Anions ($meq.l^{-1}$)			
	Sand (%)	Silt (%)	Clay (%)	Soil texture			Ca^{++}	Mg^{++}	Na^{+}	K^{+}	CO_3^{--}	HCO_3^{-}	Cl^{-}	SO_4^{--}
0-45	96.7	1.41	1.89	Sandy	0.44	8.34	2.01	1.38	0.86	0.34	-	2.60	1.61	0.38

$mol.l^{-1}$ to pH 8.2 and expressed as malic acid equivalent **(AOAC., 1990).**

Statistical analysis:

The results in the first stage were exposed to proper statistical analysis of variance for a randomized complete block design (two factors) using MSTATC computer program **(Russell, 1986)** with three replicates and each replicate was represented by two trees. Duncan's multiple range tests was used for comparison between means. Different alphabetical letters in the same column are significantly differed at (0.05) level of significance **(Duncan, 1955)**.

RESULTS AND DISCUSSION

1. Fruit physical parameters:

1.1. Fruit length:

Table (2) clears that treated trees with *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract gave the highest increase in fruit length (5.77 and 5.88 cm) in both seasons. While, the least ones were observed in the interaction between *Enterobacter aerogenes ECu₃* and vitamin E, The other interaction effects came in between.

Also, the interaction effect between phosphate sources and antioxidant applications ycted significantly on fruit length which ranged from (4.64 to 6.19cm) in the first season and from (4.83 to 6.25cm) in the second one.

The highest values of fruit length were found with the interaction between phosphoric acid and spraying with *H. sabdariffa* extract (6.19 & 6.25%). While, the least ones were recorded with untreated trees (control) (4.64 & 4.83cm) in two seasons. The other interaction came in between.

1.2. Fruit width:

As for the interaction, the effect between phosphate solubilizing bacteria and antioxidant treatments, data in Table (2) shows that treated trees with *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract gave the highest increment in fruit width (5.70 and 5.72 cm). While, the least ones were found with *Enterobacter aerogenes* ECu_3 and Vitamin E treatment, in both seasons. Concerning, the interaction effect between phosphate sources and antioxidant applications, treated trees with phosphoric acid and sprayed with *H. sabdariffa* extract proved to be the most effective treatment of fruit wide in the first season. While, treated trees with pentoxide phosphate and sprayed with *H.sabdariffa* extract gave the highest values in second season. On the other hand, the least ones were given in control treatment, during both seasons. The other treatments came in between.

Table (2):Effect of phosphate resources and/or phosphate solubilizing bacteria under antioxidant applications on fruit length, fruit width and shape index of 'EarliGrande' peach trees during 2009 and 2010 seasons.

Treatment	Antioxidant application	Fruit length (cm)		Fruit width (cm)		Shape index (L/W)	
		2009	2010	2009	2010	2009	2010
1. Effect of phosphate solubilizing bacteria and antioxidants application							
E. aerogenes	***Hibiscus sabdariffa ****	5.64^{abc}	5.77^{ab}	5.28^{bcd}	5.55^{b}	1.07^{a}	1.04^{a}
	Vitamin C.	5.44^{bcd}	5.55^{cd}	5.25^{cd}	5.38^{bc}	1.04^{a}	1.03^{a}
	Vitamin E.	5.06^{d}	5.17^{e}	5.04^{e}	4.82^{d}	1.00^{a}	1.07^{a}
B. subtilis	***Hibiscus sabdariffa***	5.77^{a}	5.88^{a}	5.70^{a}	5.72^{a}	1.0^{a}	1.03^{a}
	Vitamin C.	5.66^{ab}	5.70^{abc}	5.37^{bc}	5.35^{bc}	1.05^{a}	1.07^{a}
	Vitamin E.	5.37^{cd}	5.45^{d}	5.11^{de}	5.20^{c}	1.05^{a}	1.05^{a}
E. aerogenes x B. subtilis	***Hibiscus sabdariffa***	5.67^{ab}	5.71^{abc}	5.40^{b}	5.53^{b}	1.05^{a}	1.03^{a}
	Vitamin C.	5.45^{bcd}	5.63^{bcd}	5.60^{ab}	5.54^{b}	0.97^{a}	1.02^{a}
	Vitamin E.	5.51^{bc}	5.68^{bc}	5.19^{d}	5.37^{bc}	1.06^{a}	1.06^{a}
2. Effect of phosphate recourses and antioxidants application							
Control	***Hibiscus sabdariffa***	4.913^{fg}	5.267^{fg}	4.550^{fg}	5.563^{abc}	1.080^{a-d}	0.9467^{b}
	Vitamin C.	4.840^{fg}	4.977^{gh}	4.763^{ef}	4.690^{d}	1.027^{d}	1.060^{a}
	Vitamin E.	4.647^{g}	4.837^{h}	4.310g	4.943^{cd}	1.077^{a-d}	0.9800^{ab}
Rock Phosphate	***Hibiscus sabdariffa***	5.590^{cd}	5.900^{bcd}	5.013^{b-e}	5.580^{abc}	1.113^{a-d}	1.057^{ab}
	Vitamin C.	5.290^{c-f}	5.667^{de}	4.960^{cde}	5.357^{a-d}	1.067^{a-d}	1.057^{ab}
	Vitamin E.	5.103^{d-g}	5.460^{ef}	4.870^{cf}	5.397^{abc}	1.050^{a-d}	1.013^{ab}
Super phosphate	***Hibiscus sabdariffa***	5.463^{cde}	5.967^{a-d}	4.850^{def}	5.750^{ab}	1.127^{abc}	1.037^{ab}
	Vitamin C.	4.997^{efg}	5.810^{cd}	4.677^{efg}	5.617^{abc}	1.070^{a-d}	1.033^{ab}
	Vitamin E.	4.993^{efg}	5.307^{f}	4.813^{def}	5.113^{bcd}	1.040^{bcd}	1.040^{ab}
Pentoxide phosphate	***Hibiscus sabdariffa***	6.107^{ab}	6.197^{ab}	5.420^{a}	5.980^{a}	1.127^{abc}	1.037^{ab}
	Vitamin C.	5.470^{cde}	5.937^{a-d}	5.260^{abc}	5.757^{ab}	1.037^{cd}	1.033^{ab}
	Vitamin E.	5.660^{bc}	5.777^{cd}	5.193^{a-d}	5.430^{abc}	1.090^{a-d}	1.063^{a}
Phosphoric Acid	***Hibiscus sabdariffa***	6.190^{a}	6.25^{a}	5.527^{a}	5.977^{a}	1.120^{abc}	1.047^{ab}
	Vitamin C.	6.150^{a}	6.177^{ab}	5.437^{a}	5.820^{a}	1.130^{ab}	1.063^{a}
	Vitamin E.	6.077^{ab}	6.013^{abc}	5.360^{ab}	5.593^{bc}	1.133^{a}	1.077^{a}

Means followed by the same letter (s) within each column are not significantly different at the 0.05 level, according to Duncan’s multiple range test.

*** Aqueous extract of Roselle plant *Hibiscus sabdariffa.***

1.3. Shape index*:*

Data in Table (2) revealed that all treatment gave the similar effect in shape index in both seasons. Concerning, the interaction effect between phosphate sources and antioxidant applications, the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract, gave the highest values of shape index (1.133 and 1.077). While, the least ones were given in control treatment in both seasons.

1.4. Fruit volume:

Table (3) show that treated trees with *Bacillus subtilis* AC_2 and sprayed with *H.sabdariffa* extract gave the highest increase in fruit volume (74.47 and 79.07 cm^3) in both seasons. While, the least ones were found with *Enterobacter aerogenes* ECu_3 strain and Vitamin E. Regarding, the interaction effect between phosphate sources and antioxidant applications, data in Table (3) shows that the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract, caused high significant increase of fruit volume (73.97 and 111.5 cm^3). While, the least ones were observed in control treatment, during both seasons.

1.5. Fruit weight*:*

The interaction effect between rock phosphate, phosphate solubilizing bacteria and antioxidant treatments, is shown in Table (3). Data revealed that treated trees with rock phosphate, *Bacillus subtilis* AC_2 and sprayed with *H.sabdariffa* extract gave the highest increase in fruit weight in both seasons. While, the least ones were obtained with the interactions between rock phosphate with *Enterobacter aerogenes* ECu_3 and Vitamin E in fruit weight in both seasons. The remained interactions treatment came in between. As for the interaction effect between phosphate sources and antioxidant applications, Table (3) shows that the highest value of fruit weight were observed in the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract in both seasons. While, in both seasons the least ones were given in control treatment.

The remained treatments came in between. **Sahain *et al.* (2007)** mentioned that EM (a commercial Biostimulant) containing more than 60 selected strains more effective on fruit quality i.e., fruit weight, their dimensions, total sugars (%) and TSS (%) at harvest compared to the control on apple fruits. on the other hand **Yang Sheng (2010)** who demonstrated that after compound bio-fertilizer enrichment, fruit number per tree, weight per fruit and average yield per tree were all improved obviously in peach trees. Similar observations were reported by **Li *et al.*, (2007).** Who reported that spraying with KH_2PO_4 improved fruit quality parameters on 4 peach cultivars.

The increase in fruit weight, length, width, and size might be attributed to the increase in cell division and cell elongation caused by auxins and some antioxidants. In addition, ascorbic and citric acids as natural organic antioxidant compounds have an auxinic action and synergistic effect on improving fruit size and weight **(Kassem *et al.,* 2010)**. These results are in line with the findings of **Claudia *et al.* (2012)** who reported that Roselle (*Hibiscus sabdariffa* L.) extracts contain anthocyanin and ascorbic acid and some active antioxidants.

1.6. Total yield*:*

With regard to the interaction effect between rock phosphate, phosphate solubilizing bacteria and antioxidant treatments, Table (3) show that Rock phosphate with adding *Bacillus subtilis* AC_2 and sprayed with *H.sabdariffa* extract proved to be the most effective treatment in total yield (50.03 and 55.12 Kg. $tree^{-1}$) in both seasons.

Table (3): Effect of phosphate sources and/or phosphate solubilizing bacteria under antioxidant applications on fruit volume, fruit weight, total yield and fruit firmness of 'EarliGrande' peach trees during 2009 and 2010 seasons.

Treatment	Antioxidant application	Fruit volume (cm^3)		Fruit weight (g)		Total yield ($Kg.tree^{-1}$)		Fruit firmness	
		2009	2010	2009	2010	2009	2010	2009	2010
	1. Effect of phosphate solubilizing bacteria and antioxidants application								
E. aerogenes	***Hibiscus sabdariffa*** *	68.17^{ab}	75.42^{b}	67.30^{b}	70.11^{e}	47.68^{b}	51.42^{b}	3.41^{bcd}	3.59^{bc}
	Vitamin C.	63.75^{bc}	65.58^{e}	62.71^{d}	71.99^{cd}	42.67^{cd}	48.88^{cd}	3.51^{abc}	3.70^{b}
	Vitamin E.	54.08^{d}	63.13^{g}	55.25^{f}	68.27^{f}	37.90^{f}	39.64^{f}	3.26^{d}	3.26^{d}
B. subtilis	***Hibiscus sabdariffa***	74.47a	79.07^{a}	73.11^{a}	79.20^{a}	50.03^{a}	55.12^{a}	3.53^{ab}	3.98^{ab}
	Vitamin C.	66.54^{abc}	75.38^{bc}	66.62^{bc}	76.58^{b}	43.20^{c}	51.13^{b}	3.73^{a}	4.15^{a}
	Vitamin E.	59.80^{bcd}	68.62^{d}	59.42^{e}	76.18^{bc}	41.29^{de}	47.25^{d}	3.28^{d}	3.57^{bc}
E. aerogenes x B. subtilis	***Hibiscus sabdariffa***	72.17^{a}	77.23^{ab}	71.26^{ab}	76.47^{b}	49.92^{a}	54.45^{ab}	3.46^{abcd}	3.38^{cd}
	Vitamin C.	66.29^{abc}	75.08^{c}	64.92^{c}	74.37^{c}	42.44^{cde}	49.54^{c}	3.38 cd	3.55^{bcd}
	Vitamin E.	54.67^{cd}	64.65^{f}	59.74^{e}	71.3^{d}	40.4^{e}	45.67^{e}	2.92 e	3.01 e
	2. Effect of phosphate sources and antioxidants application								
Control	***Hibiscus sabdariffa***	54.70^{fgh}	51.80^{f}	51.77^{gh}	64.30^{e}	36.82^{e}	39.80^{ef}	2.873^{efg}	3.310^{h}
	Vitamin C.	51.17^{gh}	49.57^{f}	49.68^{gh}	63.48^{e}	36.83^{e}	45.30^{def}	2.900^{efg}	3.433^{gh}
	Vitamin E.	47.75^{h}	61.33^{ef}	44.87^{h}	55.76^{e}	35.13^{e}	38.90f	2.643^{g}	3.457^{gh}
Rock Phosphate	***Hibiscus sabdariffa***	60.67^{c-f}	71.23^{cf}	66.63^{bcd}	74.55^{cde}	52.47^{a-d}	57.86^{abc}	3.233^{c-f}	4.030^{def}
	Vitamin C.	62.17^{cde}	69.27^{c-f}	63.10^{cde}	68.35^{de}	42.97^{de}	46.58^{c-f}	3.580^{abc}	4.140^{cde}
	Vitamin E.	51.00^{gh}	65.40^{def}	52.93^{fgh}	74.1^{cde}	42.72^{de}	44.75^{def}	2.797^{fg}	3.77^{efg}
Super phosphate	***Hibiscus sabdariffa***	57.77^{d-g}	83.63^{b-e}	59.42^{c-g}	74.67^{cde}	44.04^{cde}	54.37^{a-d}	3.203^{c-f}	3.957def
	Vitamin C.	56.50^{efg}	82.17^{b-e}	55.95^{efg}	88.13^{abc}	52.85^{a-d}	51.29^{a-e}	3.593^{abc}	4.183^{bcd}
	Vitamin E.	52.17^{gh}	70.30^{c-f}	56.89^{d-g}	68.95^{de}	45.90^{b-e}	48.86^{b-f}	3.130^{def}	3.713^{fg}
Pentoxide phosphate	***Hibiscus sabdariffa***	65.83^{bc}	101.1^{ab}	68.67^{bc}	96.76^{ab}	59.53^{a}	62.12^{a}	3.913^{a}	4.700^{a}
	Vitamin C.	64.22^{cd}	83.50^{b-e}	61.77^{c-f}	91.00^{abc}	56.28^{ab}	61.23^{a}	3.973^{a}	4.873^{a}
	Vitamin E.	53.00^{gh}	95.67^{abc}	54.25^{e-h}	84.40^{bcd}	55.65^{abc}	58.74^{ab}	3.450^{bcd}	4.500^{abc}
Phosphoric Acid	***Hibiscus sabdariffa***	73.97^{a}	111.5^{a}	80.06^{a}	103.4^{a}	61.92^{a}	62.45^{a}	3.620^{abc}	4.573^{ab}
	Vitamin C.	71.67^{ab}	106.3^{ab}	74.23^{ab}	99.10^{ab}	60.46^{a}	62.22^{a}	3.710^{ab}	4.693^{a}
	Vitamin E.	64.67^{cd}	90.67^{a-d}	68.48^{bc}	92.52^{abc}	58.77^{a}	59.80^{ab}	3.310^{b-e}	4.223^{bcd}

Means followed by the same letter (s) within each column are not significantly different at the 0.05 level, according to Duncan's multiple range test.

* Aqueous extract of Roselle plant *Hibiscus sabdariffa*

1.7. Fruit firmness:

As for the interaction effect between rock phosphate, phosphate solubilizing bacteria and antioxidant treatments, Table (3) reveals that fruit firmness gave high significant increase in treated trees with rock phosphate, *Bacillus subtilis* AC_2 and sprayed with vitamin C. While, the least ones were found with the interactions between rock phosphate with (*Enterobacter aerogenes ECu₃* X *Bacillus subtilis* AC_2) and Vitamin E in both seasons. Regarding the interaction effect between phosphate sources and antioxidant applications, Table (3) clears that the interaction effect between Pentoxide phosphate and sprayed with vitamin C, caused high significant increment in fruit firmness in both season.While, control treatment gave the least ones, the other interactions came in between.

1.8. Flesh thickness:

With regard to the interaction effect between Rock phosphate, phosphate solubilizing bacteria and antioxidant treatments, Table (4) reveals that in both seasons treated trees with rock phosphate with adding *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract gave the highest increase in flesh thickness (2.20 and 2.89 cm) in both seasons. While, trees which treated with rock phosphate under *Enterobacter aerogenes* ECu_3 and sprayed with vitamin E gave the least ones in flesh thickness in 2009 and 2010 seasons. The other treatments came in between.

Concerning, the interaction effect between phosphate sources and antioxidant applications, the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract, proved to be the most effective treatment in flesh thickness in both Seasons. While, control treatment gave the least ones, in this respect. The other interaction treatments came in between.

1.9. Flesh weight*:*

As for the interaction effect between rock phosphate, phosphate solubilizing bacteria and antioxidant treatments, data in Table (4) revealed that treated trees with rock phosphate, *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract gave the highest increase in flesh weight (62.78 and 67.97g) in 2009 & 2010 seasons, respectively. While, the least ones were found with the interactions between rock phosphate with *Enterobacter aerogenes ECu3* and Vitamin E. The other interaction treatments came in between. Table (4) revealed that, in both seasons the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract, gave the highest values of flesh weight (69.60 and 103.4 g). On the contrary, the least ones were found in the control treatment, during both seasons. The other treatments came in between.

1.10. Stone weight:

With regard to the interaction effect between rock phosphate, phosphate solubilizing bacteria and antioxidant treatments, Table (4) clears that in both seasons treated trees with rock phosphate with adding *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract caused high significant increment in stone weight (10.33 and 11.23 g).While, trees treated with rock phosphate under *Enterobacter aerogenes* ECu3 and sprayed with vitamin E gave the least ones in 2009 season.

On the other hand, trees treated with rock phosphate under *Enterobacter aerogenes* ECu_3 and sprayed with *H. sabdariffa* extract gave the least value in stone weight in 2010 season. The other treatments came in between. Regarding, the interaction effect between phosphate sources and antioxidant applications, Table (4) showed that the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract, gave the highest values of stone weight in both seasons (10.45 and 11.82g).While, the least ones were observed in control treatment, during both seasons.

1.11. Flesh stone ratio:

As for the interaction effect between rock phosphate, phosphate solubilizing bacteria and antioxidant treatments, data in Table (4) reveal that flesh: Stone ratio generally ranged from 5.22 to 6.08 in the first season and from 5.56 to 6.05 in the second one.

The least values (5.22 & 5.56) in the two seasons were recorded by the interactions between rock phosphate with *Enterobacter aerogenes* ECu3 and sprayed with Vitamin E. While, the highest ones (6.05& 6.08) came from the interactions between rock phosphates, *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract during both seasons (Table 4).

Concerning, the interaction effect between phosphate sources and antioxidant applications, Table (4) revealed that the interaction effect between phosphoric acid

Table (4): Effect of phosphate sources and /or phosphate solubilizing bacteria under antioxidant applications on flesh thickness, flesh weight, stone weight and flesh: stone ratio of 'EarliGrande' peach trees during 2009 and 2010 seasons.

Treatment	Antioxidant application	Flesh thickness (cm)		Flesh weight (g)		Stone weight (g)		Flesh : stone ratio	
		2009	2010	2009	2010	2009	2010	2009	2010
1. Effect of phosphate solubilizing bacteria and antioxidants application									
E. aerogenes	***Hibiscus sabdariffa*** *	2.02^{b}	2.75^{bc}	57.00^{c}	60.11^{e}	9.14^{d}	10.01^{d}	5.54^{c}	6.01^{a}
	Vitamin C.	1.99^{bc}	2.72^{bc}	52.78^{e}	61.15^{cde}	9.93^{bc}	10.84^{bc}	5.32^{d}	5.65^{cd}
	Vitamin E.	1.84^{d}	2.55^{d}	46.37^{g}	57.83^{f}	8.88^{e}	10.44^{cd}	5.22^{e}	5.56^{d}
B. subtilis	***Hibiscus sabdariffa*** *	2.20^{a}	2.89^{a}	62.78^{a}	67.97^{a}	10.33^{a}	11.23^{a}	6.08^{a}	6.05^{a}
	Vitamin C.	2.07^{ab}	2.80^{abc}	56.38^{cd}	65.48^{b}	10.24^{abc}	11.10^{b}	5.52^{c}	5.90^{ab}
	Vitamin E.	1.94^{c}	2.66^{c}	50.28^{f}	65.00^{bcd}	10.29^{a}	11.18^{a}	5.55^{c}	5.8^{bc}
E. aerogenes x B. subtilis	***Hibiscus sabdariffa*** *	2.18^{a}	2.86^{ab}	60.99^{b}	65.32^{bc}	10.27^{ab}	11.15^{ab}	5.97^{ab}	5.86^{abc}
	Vitamin C.	2.07^{ab}	2.80^{abc}	55.43^{d}	63.52^{cd}	9.50^{c}	10.85^{bc}	5.84^{b}	5.87^{abc}
	Vitamin E.	1.95^{c}	2.67^{c}	50.26^{f}	60.68^{de}	9.48^{c}	10.63^{bcd}	5.41^{cd}	5.71^{bcd}
2. Effect of phosphate sourses and antioxidants application									
Control	***Hibiscus sabdariffa*** *	1.977^{ghi}	2.197^{h}	44.07^{gh}	64.30^{e}	7.700^{fg}	8.750^{de}	5.723^{a-d}	6.383^{de}
	Vitamin C.	1.943^{hi}	2.257^{gh}	41.99^{gh}	63.48^{e}	7.690^{fg}	8.407^{de}	5.460^{bcd}	6.677^{b-e}
	Vitamin E.	1.900^{i}	2.133^{h}	37.32^{h}	55.76^{e}	7.543^{g}	7.993^{e}	4.967^{d}	5.997 e
Rock Phosphate	***Hibiscus sabdariffa*** *	2.280^{ab}	2.490^{def}	57.53^{bcd}	74.55^{cde}	9.103^{cde}	9.797^{b-e}	6.317^{abc}	6.613 cde
	Vitamin C.	2.243^{abc}	2.380^{fg}	54.62^{cde}	68.35^{de}	8.483^{d-g}	9.007^{cde}	6.440^{ab}	6.590 cde
	Vitamin E.	2.140^{cde}	2.437^{ef}	44.38^{fgh}	74.17^{cde}	8.553^{d-g}	9.980^{a-d}	5.193^{cd}	6.433 cde
Super phosphate	***Hibiscus sabdariffa*** *	2.207^{bcd}	2.650^{bcd}	50.23^{c-g}	74.67^{cde}	9.193^{bcd}	10.10^{a-d}	5.463 bcd	6.397^{de}
	Vitamin C.	2.063^{efg}	2.590^{cde}	47.20^{efg}	88.13^{abc}	8.747^{def}	10.82 abc	5.403 bcd	7.147^{a-d}
	Vitamin E.	2.010^{fgh}	2.557^{c-f}	48.28^{d-g}	68.95^{de}	8.613^{d-g}	8.867cde	5.607^{a-d}	6.777^{a-e}
Pentoxide phosphate	***Hibiscus sabdariffa*** *	2.233^{bc}	2.723^{abc}	59.14^{bc}	96.76^{ab}	9.527^{a-d}	11.21^{ab}	6.210 abc	7.640^{ab}
	Vitamin C.	2.193^{bcd}	2.657^{bcd}	53.68^{c-f}	91.00^{abc}	8.083^{efg}	11.25^{ab}	6.643^{a}	7.103^{a-d}
	Vitamin E.	2.107^{def}	2.503^{def}	46.18^{e-h}	84.40^{bcd}	8.067^{efg}	10.06^{a-d}	5.747^{a-d}	7.397^{abc}
Phosphoric Acid	***Hibiscus sabdariffa*** *	2.347^{a}	2.893^{a}	69.60^{a}	103.4^{a}	10.45^{a}	11.82^{a}	6.673^{a}	7.753^{a}
	Vitamin C.	2.303^{ab}	2.787^{ab}	64.33^{ab}	99.10^{ab}	9.897^{abc}	11.47^{ab}	6.503 ab	7.643^{ab}
	Vitamin E.	2.273^{ab}	2.783^{ab}	58.25^{bc}	92.52^{abc}	10.23^{ab}	11.03^{ab}	5.703^{a-d}	7.397 abc

Means followed by the same letter(s) within each column are not significantly different at the 0.05 level, according to Duncan's multiple range test.

*** Aqueous extract of Roselle plant *Hibiscus sabdarffa*** other interaction treatments came in between.

and sprayed with *H. sabdariffa* extract, proved to be the most effective on flesh: Stone ratio in both seasons. While, the control treatment gave the least ones.

The other treatments came in between. The improvement occurring in vegetative growth and leaf nutrient contents certainly reflected their effect on improving yield as well as fruit weight, fruit volume and flesh weight.

Another interpretation of the positive effect of phosphorous sources, solubilizing bacterium and antioxidant application are the role of free N, P and K in producing adequate amounts of growth regulators, improving availability of nutrients which promoted the vegetative growth to go forward and then affect yield.

In addition, the beneficial effect of phosphate sources and p solubilizing bacteria on enhancing soil fertility and the uptake of different nutrients as well as adding antioxidants surely reflected on stimulating growth characters and nutritional status of the peach trees in favor of producing more fruits and yield **(El-Khawaga, 2011).**

2. Fruit chemical parameters:

2.1. TSS:

Data in Table (5) reveal that the highest increase in TSS in both seasons were observed in trees treated with rock phosphate with adding *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract (9.39 and 10.23). While, the least ones were found with trees treated with rock phosphate under *Enterobacter aerogenes* ECu_3 and sprayed with vitamin E.The other treatments came in between. Similar results were obtained by **Kassem *et al.* (2010)** who demonstrated that, ascorbic plus citric acids increased fruit weight, TSS, total sugars, reducing sugars, carotene and V.C contents and decreased fruit acidity and tannin contents as compared with untreated treatments.

Table (5) reveal that the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract, gave the highest values of T.S.S in both seasons (10.45 and 11.82). While, the least ones were found in control treatment, during both seasons. The other interaction came in between.

2.2. Acidity:

Data in Table (5) reveal that treated trees with rock phosphate, *Enterobacter aerogenes* ECu_3 and sprayed with vitamin E caused high significant in Acidity in both seasons. While, the least ones were found with the interactions between rock phosphates with *Enterobacter aerogenes* ECu_3× *Bacillus subtilis* AC_2 and sprayed with vitamin C during both seasons.

The other treatments came in between. Similar results were obtained by **Kassem *et al.*, (2010)** who demonstrated that, ascorbic plus citric acids increased fruit weight, TSS, total sugars, reducing sugars, carotene and Vitamin C contents and decreased fruit acidity and tannin **(Ta)** contents as compared with the control.

2.3. Vitamin C:

Table (5) reveal that treated trees with rock phosphate, *Enterobacter aerogenes* ECu3×*Bacillus subtilis* AC_2 and sprayed with vitamin C gave the highest increase in Vit.C (9.54 and 9.32) in 2009&2010 seasons respectively. While, the least ones were found with the interactions between rock phosphates with *Enterobacter aerogenes* ECu_3 and sprayed with vitamin E in Vit.C in both seasons.

In the mean time Table (5) reveal that the interaction effect between Pentoxide phosphate and sprayed with vitamin C, gave the highest values of Vit. C in both seasons. The least ones were given in control treatment, during both seasons.

Table (5): Effect of phosphate sources and / or phosphate solubilizing bacteria under antioxidant applications on total soluble solids, acidity, vitamin C content and fruit moisture content of 'Earli Grande' peach trees during 2009 and 2010 seasons.

Treatment	Antioxidant application	TSS (%)		Acidity (%)		Vit.C mgeq. Ascorbic Acid		Moisture content (%)	
		2009	2010	2009	2010	2009	2010	2009	2010
	1.Effect of phosphate solubilizing bacteria and antioxidants application								
E. aerogenes	***Hibiscus sabdariffa* ***	9.02^{ab}	9.98^{abc}	0.12^{ab}	0.1^{b}	7.33^{cde}	8.40^{c}	64.47^{f}	67.96^{cde}
	Vitamin C.	8.69^{cd}	9.50^{cd}	0.10^{abc}	0.14^{ab}	7.15^{de}	8.48^{c}	66.73^{e}	68.32^{cd}
	Vitamin E.	7.92^{e}	8.73^{e}	0.16^{a}	0.18^{a}	6.05^{f}	7.01^{e}	63.22^{g}	66.57^{e}
B. subtilis	***Hibiscus sabdariffa* ***	9.39^{a}	10.23^{a}	0.11^{abc}	0.12^{b}	8.29^{bcd}	8.92^{abc}	69.18^{a}	71.71^{a}
	Vitamin C.	8.96^{abc}	10.01^{ab}	0.09^{bc}	0.08^{cd}	8.55^{b}	9.06^{ab}	67.75 c	69.86^{bc}
	Vitamin E.	8.76^{bcd}	9.87^{bc}	0.13^{ab}	0.15^{ab}	6.32 e	7.81^{de}	63.36^{fg}	68.95^{c}
E. aerogenes x B. subtilis	***Hibiscus sabdariffa* ***	9.33^{a}	9.98^{abc}	0.08^{bc}	0.07^{d}	8.47^{bc}	8.84^{bc}	68.53^{b}	71.03^{ab}
	Vitamin C.	8.81^{bc}	9.77^{bcd}	0.07^{c}	0.07^{d}	9.54^{a}	9.32^{a}	67.19^{d}	70.38^{b}
	Vitamin E.	8.52^{d}	8.91^{d}	0.10^{abc}	0.09 c	7.68^{cd}	7.88^{d}	63.52^{f}	67.62^{de}
	2.Effect of phosphate sourses and antioxidants application								
Control	***Hibiscus sabdariffa* ***	8.337^{efg}	8.243^{fg}	0.547^{b}	0.653^{b}	5.03^{h}	5.590^{i}	61.43^{bc}	66.46^{fg}
	Vitamin C.	7.463^{ghi}	8.297^{fg}	0.373^{g}	0.547^{g}	5.333^{gh}	6.760^{ghi}	59.64^{c}	64.02^{hi}
	Vitamin E.	6.597^{i}	7.947^{g}	0.587^{a}	0.667^{a}	4.767^{h}	5.333^{j}	52.87^{d}	63.04^{i}
Rock Phosphate	***Hibiscus sabdariffa* ***	8.790^{def}	9.667^{de}	0.333^{h}	0.600^{e}	9.233^{a-d}	7.167^{fgh}	65.8^{ab}	67.37^{ef}
	Vitamin C.	7.820^{fgh}	9.937^{cd}	0.253^{k}	0.533^{h}	9.383^{a-d}	8.490^{def}	63.90^{abc}	65.38^{fgh}
	Vitamin E.	6.953^{hi}	9.047^{ef}	0.467 c	0.613^{d}	7.167^{ef}	6.333^{hij}	63.00^{abc}	64.87^{ghi}
Super phosphate	***Hibiscus sabdariffa* ***	9.377^{a-d}	10.13^{cd}	0.413^{d}	0.507^{j}	8.267^{c-f}	7.950^{efg}	64.70^{abc}	70.05^{cd}
	Vitamin C.	8.967^{cde}	10.12^{cd}	0.333^{h}	0.320^{m}	7.733^{def}	9.770^{a-d}	63.55^{abc}	69.62^{cd}
	Vitamin E.	8.720^{def}	10.04^{cd}	0.387^{f}	0.440^{k}	6.860^{fg}	6.933^{ghi}	59.54^{c}	66.68^{fg}
Pentoxide phosphate	***Hibiscus sabdariffa* ***	9.920^{abc}	10.03^{cd}	0.280^{j}	0.293^{n}	10.23^{ab}	10.92^{ab}	67.29^{a}	71.01^{abc}
	Vitamin C.	9.080^{b-e}	9.493^{de}	0.253^{k}	0.213^{o}	10.93^{a}	11.19^{a}	65.06^{abc}	70.76^{bcd}
	Vitamin E.	8.400^{d-g}	8.907^{ef}	0.320^{i}	0.400^{l}	9.543^{abc}	9.663^{bcd}	64.64^{abc}	68.84^{de}
Phosphoric Acid	***Hibiscus sabdariffa* ***	10.10^{a}	11.69^{a}	0.333^{h}	0.587^{f}	8.833^{b-e}	10.29^{abc}	68.5^{a}	72.96^{a}
	Vitamin C.	9.990^{ab}	11.28^{ab}	0.253^{k}	0.520^{i}	10.03^{abc}	10.57^{ab}	67.19^{a}	72.23^{ab}
	Vitamin E.	9.083^{b-e}	10.80^{bc}	0.400^{e}	0.627^{c}	7.733^{def}	9.000^{cde}	66.54^{ab}	69.95^{cd}

Means followed by the same letter (s) within each column are not significantly different at the 0.05 level, according to Duncan's multiple range test.

* Aqueous extract of Roselle plant *Hibiscus sabdariffa.*

Similar observations were reported by **(Li *et al.* 2007).**

2.4. Moisture content:

Table (5) reveal that treated trees with rock phosphate, *Bacillus subtilis* AC_2 and sprayed with *H. sabdariffa* extract gave the highest significant increase in Vit. C (68.53 and 71.03%) in both seasons. While, the least ones were found with the interactions between rock phosphates with *Enterobacter aerogenes* ECu_3 and sprayed with Vitamin E in Vit. C in both seasons.

The remained interaction treatments came in between. Similarly Table (5) show that the interaction effect between phosphoric acid and sprayed with *H. sabdariffa* extract, gave the highest values of moisture content in both seasons.

While, the least ones were found in control treatment and the other treatments came in between. Finally, it can be concluded that the increase in leaf nutrient contents, specially phosphorus leads to increase photo-synthesis and thereby increase the proportion of carbohydrates in the leaves and reflected in fruit chemical properties *i.e.,* total soluble solids (TSS) and decrease total acidity **(Stino *et al.* 2009).**

Table (5) reveal that the interaction effect between control treatment with vitamin E, gave the highest significant values of acidity in both seasons. While, the control treatment gave the least ones in 2009 and 2010 seasons and the other treatments came in between.

REFERENCES

AOAC. (1990). Official Methods of Analysis. 15th ed., Association of Official Agriculture Chemists Washington D.C., USA.

Asea, P. E. A., Kucey, R. M. N. and Stewart, J. W. B. (1988). Inorganic phosphate solubilization by two Penicillium species in solution culture and soil. Soil Biol. Biochem., **20**: 459-464.

Awasthi, R. P.; Bhutani, V. P.; Mankotia, M. S; Kith, N. S; and Dev, G. (1998). Potash improves to yield and quality of July Elberta peach. Better Crops Intern.12, 30

Chatzitheodorou, I. T; Sotiropoulos, T. E; and Mouhtaridou, G. I; (2004). Effect of nitrogen, phosphorous, potassium fertilization and manure on fruit yield and fruit quality of the peach cultivars "Spring Time" and "Red Haven". Agronomy research. 2 (2), 135- 143.

Claudia S. G.; Fidel T. V.; Ana E. O.; and José Á. G. (2012). Antioxidant properties and color of *Hibiscus sabdariffa* extracts. Cien. Inv. Agr. 39(1):79-90.

Duncan, B. D. (1955). Multiple Range and Multiple F-tests. Biometrics, 11: 1- 42.

El- Khawaga, A.S. (2012). Partial Replacement of Mineral N Fertilizers by Using Humic Acid And Spirulina. Platensis Algae Biofertilizer In Florida Prince Peach Orchards. Middle East Journal of Applied Sciences 1 (1): 5-10, 2011.

Gyaneshwar, P., Naresh, K. G. and Parekh, L. J. (1998). Cloning of mineral phosphate solubilizing genes from Synechocystis PCC 6803 P., Curr. Sci., **74:** 1097-1099.

Ivanova, R., Bojinova, D. and Nedialkova, K. (2006). Rock Phosphate solubilization by soil bacteria, J. Uni. of Chemical Techn. and Metallurgy, **41** (3)**:** 297-302.

Kassem H. A., Amal M. El-Kobbia, Hend A. Marzouk and Mohamed M. El- Sebaiey (2010). Effect of foliar sprays on fruit retention, quality and yield of Costata persimmon trees. Emir. J. Food Agric. 2010. 22 (4): 259-274.

Li J.; Wang Z.; Pang Z. Y.; Fang Q.; and Li Cheng B. (2007). Effects of spraying KH2PO4 on leaf chlorophyll content, leaf weight and fruit quality of peach cultivars. Journal of Fruit Science. 2007. 24: 4, 533-536. 7 ref.

Piper, C. S. (1947). Soil and plant analysis. The University of Adelaide (Australia) pp: 59-74.

Russell, D. F. (1986): MSTATC Director, Crop and Soil Sciences Department, Michigan State University, Computer Program Package Version 2.10.

Sahain M. F.M., Elham Z. Abd El Motty, Mohamed H.El- Shiekh and Laila. F. Hagagg (2007). Effect of Some Biostimulant on Growth and fruiting of Anna Apple Trees In Newly Reclaimed Areas. Research Journal of Agriculture and Biological Sciences, 2007.3(5): 422-429.

Samah, Y.A.E., (2002). Effect of biofertilizer on yield and berry qualities of grapevines. M. Sc. Thesis. Fac. Agric., Mansoura Univ., Egypt.

Singh, C. and B.B. Sharma, (1993). Leaf nutrient composition of sweet orange as affected by combined use of bio and chemical fertilizers. South Indian Hort., 41: 131-134.

Stino, R.G., A.T. Mohsen and M.A. Maksoud, (2009). Bio-Organic fertilization and its impact on apricot young trees in newly reclaimed soil. American - Eurasian J. Agric. And Environ. Sci., 6(1): 62-69.

Toro, M., Azcon, R. and Barea, J.M. (1997). Improvement of arbuscula mycorrhiza development by inoculation of soil with phosphatesolubilizing rhizobacteria to improve rock phosphate bioavailability ((sup32) P) and Nutrient Cycling. Appl. Environ. Microbiol., 63: 4408-4412.

Yang M. sheng (2010). The effects of Nongdage bio-fertilizer on peach growth output and quality. Chinese Agricultural Sc. Bulletin., 26(1): 130-133.

الملخص العربي

استخدام البكتريا المذيبة للفوسفات ومضادات الأكسدة لتحسين جودة ثمار الخوخ الايرلي جراند

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أجريت الدراسة خلال موسمي ٢٠٠٩، ٢٠١٠م بأحد المزارع الخاصة بمنطقة الخرافين بمركز رفح محافظة شمال سيناء مصر، لدراسة تأثير البكتريا المذيبة للفوسفات (*Enterobacter aerogenes* ECu_3 منفرد، *Bacillus subtilis* AC_2 منفرد، *Enterobacter aerogenes* ECu_3×*Bacillus subtilis* AC_2)، ومصادر مختلفة للفوسفات (الفوسفات الصخري، وحمض الفوسفوريك، وخامس أكسيد الفوسفات والسوبر فوسفات)، مع الرش الورقي ببعض مضادات الأكسدة (حمض الاسكوربيك، فيتامين E ، والمستخلص المائي من الكركديه) على الخصائص الفيزيائية والكيميائية والمحصول الكلي لثمار الخوخ الأيرلي جراند عمر سبعة عشر عاماً والمنزرعة في تربة رملية، وقد أظهرت النتائج أن جميع الخصائص الفيزيائية والكيميائية للثمار والمحصول الكلي للثمار زادت بشكل ملحوظ مع المعاملة بالبكتريا *Bacillus subtilis* AC_2 والمستخلص المائي للكركديه، كما أثرت جميع مصادر الفوسفات في الخصائص الفيزيائية والكيميائية للثمار، وقد سجلت أعلى القيم مع المعاملة بحامض الفوسفوريك والمستخلص المائي للكركدية. وبخصوص محصول الثمار سجلت الأشجار المعاملة بكلاً من خامس أكسيد الفوسفات وحمض الفوسفوريك مع الرش بالمستخلص المائي بالكركدية وحامض الأسكوربيك أعلى القيم مقارنة بباقي المعاملات خلال موسمي الدراسة.

الكلمات الإسترشادية: البكتريا المذيبة للفوسفات، ومضادات الأكسدة، الجودة، الخوخ الايرلي جراند.

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