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IMPROVING FRUIT QUALITY OF ORGANICALLY PRODUCED FRUITS OF EGYPTIAN LIME BY APPLYING SOME BIOFERTILIZERS AND GIBBERELLIC ACID

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Abstract: Citrus fruits have the majority importance of fruit crops in Egypt. Limes come next to both of oranges and mandarin, whereas lime trees are grown mainly in new reclaimed sandy soils for favorite production and exportation potential. This study was carried out during two seasons 2003 and 2004 on organically fertilized Egyptian lime trees grown in reclaimed sandy soil of the experimental station in El-Ghoraieb region, Faculty of Agriculture, Assiut University. The objective of this investigation was to improve fruit quality of organically fertilized Egyptian lime trees by using both of GA_3 (0, 5 or 10 ppm, sprayed 2 weeks after fruit set) and Nitropine (strains of nitrogen fixing bacteria) or Phosphorine (phosphate dissolving bacteria) at rate of 100 g/tree, each applied at fruit set. The obtained results of this study showed that spraying GA_3 alone and both of Nitropine or Phosphorine induced significant improvement of physical and chemical characteristics of mature fruit. Phosphorine was more effective in improving fruit quality than Nitropine. Moreover, the combination between GA_3 (10 ppm) and Phosphorine gave the best results in this connection, specially for fruits that were harvested at the 3rd harvest date. Therefore, it could be recommended to spraying GA_3 (10 ppm) combined with 100 g of Phosphorine/tree to organically fertilized trees and fruits should be picked at the 3rd harvest date (at the 2nd or 3rd week of August) for improving fruit quality of Egyptian lime under the same conditions of this study.

Key words : Egyptian lime, GA_3, Nitropine, Phosphorine, fruit quality.

Introduction

Citrus trees are the most important fruit crop in Egypt, moreover, Egyptian lime is considered the favorite fruit crop grown in new reclaimed sandy soil for its production and exportation potential. Biofertilization of citrus under Upper Egypt conditions has received little attention, although, biofertilizers play an important role in enhancement citrus productivity, specially of trees grown in new reclaimed sandy soils. Moreover, biofertilizers are safe for human, animal and environment. Furthermore, producing organic healthy lime fruits could not be achieved without using biofertilizers. Biofertilizers could be improved crop productivity through increasing biological N-fixation, availability and

uptake of nutrients, as well as stimulation of natural hormones (Kannayan, 2000; Abd El-Moniem and Radwan, 2003; Abd El-Moniem *et al*., 2003 and Hegab *et al*., 2005).

Spraying antioxidant and biofertilizer either alone or in combinations on some banana cvs. caused a remarkable promotion on growth pramaters, bunch weight, physical and chemical properties of fruits (El-Shammaa, 2001; Ahmed *et al*., 2003 and Mostafa and Abou-Raya, 2004).

Furthermore, applying some biofertilizers to Zaghloul date palm cv grown in Marriout region, Alexandria Governorate improved yield weight/palm as well as physical and chemical characteristics of fruits (Osman, 2003).

Several researches were carried out on gibberellins treatments on citrus crops. For instance, GA_3 increased yield and fruit juice % of Hamlin orange and had slightly effect on fruit weight in Clementine mandarin (Guardiola, 2000), improved physical and chemical characteristics of Balady lime fruits (*Citrus aurantifolia*) (El-Agamy *et al*., 2004). Studying effects of spraying GA_3 on treated citrus fruits with biofertilizers alone or in combination with GA_3 has been received a little attention, particularly under Upper Egypt conditions.

Therefore, the objective of this study was to examining the effects of spraying GA_3 on quality of organically produced Egyptian lime fruits treated with some biofertilizers alone or in combination with biofertilizers and GA_3 pramaters estimated during three different harvesting dates of Egyptian lime fruits grown in reclaimed sandy soil under Assiut governorate climatic conditions.

Materials and Methods

This investigation was carried out during two consecutive seasons 2002/2003 and 2003/2004 on Egyptian lime trees grown in reclaimed sandy soil (some physical and chemical properties of the experimental soil site are shown in Table (1,a), at El-Ghoraieb Experimental Station, Faculty of Agriculture, Assiut University. Fifty-four trees 14 years old, approximately at the same vigour were selected for this research. Each tree received 50 kg adult organic farmyard manure at the 1st week of November (analysis of organic farm manure is shown in Table, 1,b. Half of the selected trees were assigned for the 1st season and the other half for the 2nd season for achievement the objectives of this study. To assess the effects of applying some biofertilizers (Nitropene or Phosphorine at rate of 100 g/tree each, they were added after fruit set (20 April and 6 May in 2003 and 2004 seasons, respectively. As well as spraying GA_3 (0, 5 or 10 ppm, 2 weeks after fruit set during three different harvest dates (1st harvest was at 15th or 20th July), (2nd harvested was

at the 30th July or 6th August), and (3rd harvest was at 14th or 20th August in 2003 and 2004 season, respectively) on mature fruits quality characteristics.

Fruit quality parameters were estimated as follow:

1. Physical characteristics of fruits:

1.1. Fruit weight:

Fruit weight (g) was estimated as average of ten mature fruits per replicate.

1.2. Fruit volume:

This parameter was determined by measuring volume of the water displaced immersing the same ten fruit in graduated cylinder, thereafter, fruit volume (cc) was estimated as average of the ten fruit volume.

1.3. Juice weight percentage:

Juice weight % per fruit was calculated by dividing juice weight (g) by fruit weight (g) using the following equation:

$$Juice\ weight\ \% = \frac{Juice\ weight}{Fruit\ weight} x100$$

Table (1-a): Some soil physical and chemical properties of the experimental site*.

Soil property	Soil depth (cm)**	
	0-30	30-60
Sand (%)	91.38	91.60
Silt (%)	5.27	4.98
Clay (%)	3.35	3.91
Texture	Sandy	Sandy
Field capacity	15.95	16.17
$CaCO_3$ (%)	16.87	15.75
Organic matter (%)	0.083	0.067
pH (1:1 suspension)	8.44	8.38
EC (dS/m^{-1})	1.64	1.59
CEC (c mol^{+1}/kg)	7.07	6.56
Total N (%)	0.03	0.02
$NaHCO_3$-extractable P (ppm)	4.77	4.16
NH_4OAC-extractable K (ppm)	152.06	178.8
NH_4OAC-extractable Ca (ppm)	729.6	748.8
NH_4OAC-extractable Mg (ppm)	304.1	315.6
DTPA-extractable Fe (ppm)	3.43	3.89
DTPA-extractable Mn (ppm)	1.91	1.76
DTPA-extractable Zn (ppm)	0.27	0.21
DTPA-extractable Cu (ppm)	1.05	0.97

* Soil and water Department, Faculty of Agriculture, Assiut University.
** Each value represents the average of 3 samples.

Table (1-b): Analysis of organic farm manure

Character	Farm manure	
	1st season	2nd season
Moisture %	27.0	34.0
pH value	6.8	7.0
Total N %	0.56	0.62
Total P %	0.36	0.46
Total K %	1.54	1.60
O.M. %	8.19	9.16
C/N ratio	22.8:1	30.2:1

1.4. Juice volume/fruit:

This parameter was measured by using a graduated cylinder as average of the extracted juice of 10 fruits.

2. Chemical characteristics of fruit:

2.1. Total soluble solids percentage:

The percentage of total soluble solids (TSS%) in fruit juice was determined by using a hand refractometer.

2.2. Titratable acidity percentage:

Titratable acidity percentage (TA) (as grams of citric acid/100 ml juice) was determined by titrating 10 ml fruit juice against 0.3 N NaOH with phenolphythalin as an indicator according to A.O.A.C. (1995).

2.3. Total soluble solids/acid ratio:

The ratio between TSS% and titratable acidity % (TSS/TA ratio) was calculated by dividing TSS% by TA % in fruit juice.

Statistical analysis:

This study consists of three separate experiments; the 1st experiment was carried out on trees only fertilized with organic farm manure (untreated trees with biofertilizers as control trees), the 2nd experiments revealed trees treated with Nitropine (strains of nitrogen fixing bacteria *Azotobacter* sp.) beside applying the organic farm manure and the 3rd experiment was carried out on trees treated with Phosphorine (phospahte dissolving bacteria *Bacillus negaterium*), in addition to applying the organic farm manure. Moreover, each experiment of this study was conducted in split-plot arrangement of randomized complete block design (CRB) with three replicates, one tree each, whereas, GA_3 concentrations (0, 5 or 10 ppm) were assigned as four whole plots (A), while different harvest dates were considered as split-plot (B).

All recorded data were tabulated and statistically analysed according to Mead *et al*. (1993) using new L.S.D. at the level of 0.05 for comparing various treatment means.

Results and Discussion

The results obtained during the course of this study explaine physical and chemical characteristics of Egyptian lime fruits in response to Nitropine, Phosphorine and GA_3, as well as harvesting date in 2003 and 2004 seasons.

1.Physical Characteristics of Fruits:

1.1. Weight and volume of fruit:

As shown in Tables (2 and 3) data clearly prove that all treatments significantly increased weight and volume of mature fruits not only between Nitropine, Phosphorine and GA_3, but also among different harvest dates; all compared to untreated control fruits in 2003 and 2004 seasons.

Concerning the harvest date effect on weight or volume of fruit, the later harvest date (the 3rd date at 4th or 20th of August) produced the heaviest weight and bigger volume than the anticipated harvest date (the 1st harvest date at 15th or 20th of July) in the 2003 and 2004 seasons, respectively.

Regarding the effects of biofertilizers, it was noticed that Phosphorine resulted in heavier and bigger fruits than Nitropine, and both of them induced higher fruit weight than untreated control fruits.

Moreover, the combinations between GA_3 and biofertilizers or harvest date gave the heaviest weight and larger volume of mature fruits, specially the high concentration of GA_3 (10 ppm) sprayed on treated trees with Phosphorine and picked fruits at the 3rd harvest date in comparison with untreated control fruits in 2003 and 2004 seasons.

The positive effects of Nitropine or Phosphorine applications on weight or volume of fruits could be due to realizing more fixing N and more available soluble phosphorus to treated trees than untreated one (Abd El-Moniem *et al*., 2003). These results confirm early reports by Ebrahiem and Mohamed (2000) on Balady mandarin, Abd El-Rahman (2002) on Navel orange trees, Abd El-Moniem and Radwan (2003) on Williams banana and Mohamed and Ragab (2003) on Balady mandarin. Furthermore, the enhancement effects of GA_3 spraying on treated trees with biofertilizer on untreated trees could be attributed to increasing fruit juice volume through increasing cell enlargement during growth stages.

Additionally, mature fruit weights were higher weight and volume at the latest harvest date (the 3rd date) than at the anticipated harvest date (the 1st date), this, could be due to decreasing the competition upon foods, not only between fruits themselves, but also between new shoots and fruits. Finally, it could be concluded that the obtained results of this study are in harmony with early findings reported by Guardiola (2000) on Clementine mandarin and El-Agamy *et al*. (2004) on Balady lime who deduced that both fruit weight and size was improved by

GA_3 treatments. As well as Abd El-Moniem and Radwan (2003) found that applying biofertilizers (phosphate dissolving bacteria) to Williams banana increased bunch weight, number of heads/bunch and number of fingers/hand. The same trend was observed by El-Sese (2005) on Balady mandarin.

1.2.Juice weight and Volume/ Fruit:

Data presented in Tables (4 & 5) showed significant increases in both juice weight % and juice volume (cc) per mature fruit in response to application of either biofertilizers (nitropine or phosphorine) alone or combinations between biofertilizers and GA_3 spraying, as well as among different harvest dates in 2003 and 2004 seasons.

Moreover, the effects of biofertilizers or GA_3 spraying, as well as harvest dates on juice weight % or juice volume (cc)/fruits showed the same trend of the treatments effects on fruit weight or volume as discussed in aforementioned parameters of this study.

According to this results, spraying 20 ppm GA_3 on treated trees with phosphorine gave the best results when mature fruits were picked at the latest harvest date (the 3rd date).

These findings of this study are on line with what have been reported by Ebrahiem and Mohamed (2000) on Balady mandarin, Mohamed and Ragab (2003) on Balady mandarin and El-Agamy *et al*. (2004) on Balady lime who deduced that juice % was significantly higher in fruits of trees receiving GA_3 at 5 or 10 ppm in comparison with the untreated control trees.

2. Chemical Characteristics of Fruits:

2.1. Total soluble solids percentage in fruit juice:

Data from Table (6) indicated that total soluble solids % (TSS%) in juice of mature lime fruits significantly increased at later harvest date than the anticipated harvest date among all treatments in 2003 and 2004 seasons.

Regarding to biofertilizers application, it was obviously that phosphorine induced higher TSS% in juice than nitropine, in contrarily, GA_3 spraying resulted in significant decrease in TSS% in lime juice, with the exception of the high concentration of GA_3 (10 ppm) slightly increased TSS% in lime juice, all compared with untreated control fruits in 2003 and 2004 seasons.

The negative effect of GA_3 spraying on TSS% in lime juice could be due to delaying effect of GA_3 on fruit maturation rather than untreated fruits with GA_3.

These obtained results of this study confirm the early findings of Ahmed *et al*. (1988) on Egyptian Balady lime, Ebrahiem and Mohamed (2000), who deduced that farmyard manure improved TSS% in fruit juice of Balady mandarin, Ahmed *et al*.

(2003) on Flame seedless grapevines, El-Sese (2005) on Balady mandarin, Hegab *et al*. (2005) on Balady mandarin and El-Agamy *et al*. (2004) who demonstrated that TSS% was slightly increased as a result of GA_3 application. On the other hand, the obtained results of this study in this connection are in disagreement with those found by Mohamed and Ragab (2003) on Balady mandarin, who reported that organic manure applied at ratios above 50% of inorganic N source to Balady mandarin trees tended to decrease most of the chemical properties of fruit juice.

2.2. Titratable acidity % in fruit juice:

Data presented in Table (7) indicated that all treatments with biofertilizers or GA_3, as well as harvest date of mature fruits significantly increased titratable acidity % (as grams of citric acid/100 g. juice) in 2003 and 2004 seasons.

Concerning the effect of biofertilizers on titratable acidity % in lime fruit juice, it was clear that both of nitropine and phosphorine induced significant increase in comparison to untreated control trees in 2003 and 2004 seasons.

Regarding to GA_3 effects on titratable acidity % in lime juice, it was clear that spraying GA_3 alone or in combination with biofertilizers significantly increased titratable acidity % in lime juice, except spraying GA_3 on treated trees with phosphorine during the 1st studied season wherein induced a slight decrease in titratable acidity % in lime juice compared to untreated control fruits.

On the other hand, according to the obtained results of this table, it could be noticed that of the anticipated harvest date (1st harvest), fruit juice had lower titratable acidity, followed by the latest harvest date (the 3rd harvest), then the middle harvest date (the 2nd harvest) whereas the highest value of titratable acidity % was obtained from the untreated control fruits during the two studied seasons. Furthermore, the latest harvest date (the 3rd harvest) gave the highest value of titratable acidity % in mature of lime juice in comparison with untreated control fruits in 2003 and 2004 seasons.

These obtained findings of this study are in parallel with those pointed out by Mohamed and Ragab (2003) on Balady mandarin and Abd El-Hady (2003) on grapevines, Osman (2003) on Zaghloul date palm, Mostafa (2004) on Grand Nain banana, and El-Sese (2005) on Balady mandarin, whereas they reported that organic manure as well as biofertilizers induced remarketable increase in total acidity % in fruit juice.

On the other hand, the obtained results of this study are on contrary to the results revealed by Ebrhiem and Mohamed (2003) on Balady mandarin trees, Ahmed *et al*. (2003) on Flame

seedless grapevines, Abd El-Moniem *et al*. (2003) on Fagrikalan and Elphonse mango trees, Abd El-Rahman (2003) on Novel orange and Hegab *et al*. (2005) who reported that organic manure or biofertilizers induced a reduction in total acidity % in fruit juice. Concerning GA_3 effects on total acidity % in lime juice, El-Agamy *et al*. (2004) have been observed a slight effect on acidity % in Balady lime fruit juice.

2.3. Ratio between total soluble solids and titratable acidity in juice:

Data presented in Table (8) illustrated that all treatments with biofertilizers or GA_3 induced significant decrease in values of the ratio between the total soluble solids and the titratable acidity in lime fruit juice, with the exception of the combination between phosphorine and GA_3 at the 1st season of the study.

Concerning the effect of harvest date on TSS/acid ratio in lime fruit juice, it was obvious that anticipated harvest date of untreated mature fruits gave the highest value of TSS/acid ratio, followed by the later harvest date (the 3rd harvest), then the middle harvest (the 2nd harvest), where it had the lower value of TSS/acid ratio in 2003 and 2004 seasons.

Decreasing of TSS/acid ratio values in response to the treatments with biofertilizer or GA_3 could be due to inducing an increase of titratable acidity in lime fruit juice rather than the increase of TSS by the treatments carried out during this study.

These obtained results of this study are in harmony with those reported by Abd El-Moniem *et al*. (2003) on Fagrikalan and Elphonse mango trees, Abd El-Rahman (2003) on Navel orange trees, Hegab *et al*. (2005) on Balady orange who found that organic manure or biofertilizer alone or in combination with some growth regulators revealed on positive effects on TSS/acid ratio in fruit juice.

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تحسين جودة ثمار الليمون المصرى المالح المنتجة عضوياً بإضافة بعض المخصبات الحيوية وحمض الجبريليك

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للموالح أهمية رئيسية فى مصر وخاصة الليمون المصرى المالح لكونه من أنسب المحاصيل للزراعة العضوية فى الأراضى الرملية حديثة الاستصلاح لإنتاجيته المناسبة وإمكانية تصدير ثماره، لذا أجريت هذه الدراسة خلال موسمى 2003 ، 2004 على أشجار الليمون المصرى المالح المنزرعة فى أرض رملية مستصلحة بمحطة التجارب البحثية بالغريب بكلية الزراعة جامعة أسيوط ، كمحاولة لدراسة مدى إمكانية تحسين خصائص الجودة لثمار الليمون المصرى المالح المنتجة عضوياً بإضافة بعض المخصبات الحيوية (النتروبين والفوسفورين بمعدل 100 جم / شجرة بعد العقد مباشرة) مع رش حمض الجبريليك GA_3 (بتركيز 5 أو 10 جزء فى المليون بعد أسبوعين من عقد الثمار) ولقد تم جمع الثمار المكتملة النمو خلال ثلاثة مواعيد مختلفة للجمع هى الأول فى الأسبوع الثانى أو الثالث من يوليو والثانى بعده بأسبوعين من الأول والثالث بعد الأول بحوالى شهر . ولتحقيق هدف هذه الدراسة أجريت ثلاثة تجارب صممت كل منها بنظام القطع المنشقة كاملة العشوائية مع استخدام ثلاثة مكررات وشجرة لكل مكررة .

أهم النتائج فيما يلى :

- أدت جميع المعاملات بالمخصبات الحيوية أو حمض الجبريليك أو التفاعل بينهم إلى حدوث تأثيرات إيجابية معنوية فى كل من الصفات الطبيعية للثمار (وزن وحجم الثمرة ، وزن وحجم عصير الثمرة) والصفات الكيميائية لعصير الثمرة (النسبة المئوية للمواد الصلبة الذائبة الكلية %TSS ، الحموضة الكلية ، والعلاقة بينهما TSS/acid ratio) .
- تفوقت المعاملة بالفوسفورين على كل من النتروبين أو حمض الجبريليك فى تحسين خصائص جودة الثمار فى مواعيد الجمع الثلاثة .
- أدت المعاملة بالتركيز العالى من حمض الجبريليك (10 جزء فى المليون) بالتفاعل مع الفوسفورين إلى الحصول على أفضل خصائص جودة للثمار خاصة فى ميعاد الجمع المتأخر (الثالث خلال الأسبوع الثانى أو الثالث من شهر أغسطس) .
- بخصوص مواعيد الجمع الثلاثة وجد أن أفضل صفات جودة للثمار التى تم جمعها فى الميعاد المتأخر (الثالث) .

وعليه يمكن التوصية بإضافة 100 جم من المخصب الحيوى الفوسفورين مع رش الأشجار بتركيز 10 جزئ فى المليون من حمض الجبريليك بعد العقد بأسبوعين مع جمع الثمار متأخراً فى أغسطس وذلك للحصول على أفضل صفات جودة لثمار الليمون المصرى المالح تحت ظروف الدراسة .