

Trials for Reducing Pollution and Improving Productivity of Valencia Orange Trees

Faissal, F. Ahmed¹; Aly, H. Aly¹; Tawfik A., Ebrahiem² and Huda M.H. Ismael²

¹Hort. Dept. Fac. of Agric. Minia univ. Egypt

² Citriculture Dept. Hort. Res. Instit. ARC, Giza

Faissalfadel@yahoo.com

Abstract: During 2011/ 2012 and 2012/ 2013 seasons, Valencia orange trees fertilized with N as 100% inorganic N , as well as 37.5 to 75% inorganic N + 12.5 % organic + 50 to 400 ml EM/ tree and/ or foliar fertilized with amino acids (tryptophan, methionene and arginine) enriched with NPKMgFeZnFe and B. Leaf area, nutritional status of the trees, yield , physical and chemical characteristics of fruits as well as nitrate and nitrite in the juice in response to the present treatments were investigated. The impact of these treatments on the amount of CO₂, nitrogenase activity and total counts of bacteria in the soil was also studied. Results showed that using N as 50 to 75% inorganic N + 12.5% organic + 50 to 200 ml EM/ tree was superior than using N as 100% inorganic N or as 37.5% inorganic N + 12.5% organic N + 400 ml EM/ tree in improving the leaf area, total chlorophylls, leaf content of N, P, K and Mg, initial fruit setting%, fruit retention % and yield. The same trend was observed with foliar application of amino acids enriched with nutrients. Decreasing percentages of mineral N from 100 to 37.5% and increasing levels of EM from 0.0 to 400 ml / tree caused a gradual promotion on both physical and chemical characteristics of the fruits as well as amounts of CO₂, nitrogenase activity and total counts of bacteria in the soil. A progressive reduction on both nitrate and nitrite in the juice was observed with reducing percentages of inorganic N and increasing levels of EM. Application of amino acids enriched with nutrients materially improved fruit quality. The best results with regard to yield and fruit quality of Valencia orange trees were obtained with supplying the trees with N as 50% inorganic N + 12.5 % organic + 200 ml EM / tree in combined with spraying amino acids enriched with nutrients at 0.1 % three times.

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1. Introduction

In Valencia orange orchards; many attempts were accomplished for using N at balanced rate through using organic, organic and bioforms of N, biofertilization with EM and application of amino acids enriched with different nutrients. Organic and biofertilization especially with EM are responsible for enhancing soil fertility and uptake of nutrients (Mengel *et al.*, 2001 and Kannaiyan, 2002). Amino acids have many regulatory effects on the biosynthesis of proteins and natural hormones (Davies, 1982). Nutrients are very essential in enhancing all organic foods natural hormones, plant pigments, activation of enzymes and cell division (Mengel *et al.*, 2001 and Ahmed *et al.*, 2009).

Application of organic and biofertilizers (Abdelaal *et al.*, 2012; Mohamed and Mohamed, 2013; Al- Khawaga and Maklad, 2013 and Faraag, 2013); EM (Ahmed Samah, 2011 and Ibrahiem, 2012) amino acids (Fathalla, 2013; Ahmed *et al.*, 2013 ; El- Badawy and Abd El-aal, 2013; Haggag-Leila *et al.*, 2013; Hassan, 2014; Ahmed *et al.*, 2014 a & 2014 b and Sayed – Ola, 2014) and nutrients (El-Sayed- Esraa, 2010, Gamal, 2013 and Ibrahiem and

Al- Wasfy, 2014) was very effective in improving productivity of fruit crops.

The target of this study was elucidating the effect of partial replacement of inorganic N by using EM as well as foliar application of amino acids enriched with nutrients on yield and fruit quality of Valencia orange trees.

2. Material and Methods

This study was carried out during two consecutive experimental 2011/ 2012 & 2012/ 2013 seasons on uniform in vigour thirty 49- years old Valencia orange trees onto sour orange rootstock. The selected trees are grown in the orchard of Agricultural Research Station located at Malloway district, Minia Governorate (about 300 km southern Cairo). The trees planted at a spacing of 6 x 6 meters. The soil of the orchard is well drained clay (Table 1) in texture with a water table not less than two meters deep. Surface irrigation system was carried out using Nile water.

The selected trees were subjected to the normal horticultural practices that already applied in the orchard except those dealing with inorganic, organic and biofertilization, application of EM, amino acids and different nutrients.

This investigation was consisted of ten treatments from two factors (A & B). The first factor (A) comprised from five treatments of inorganic, organic and effective microorganisms (EM) namely:

Table (1): Analysis of the soil at the trial location

Constituents	Values
Sand %	7.0
Silt %	13.5
Clay %	79.5
Texture	Clay
O.M. %	2.33
pH (1:2.5 extract)	8.11
E.C (1:2.5 extract) (mmhos/ cm/ 25° C)	0.95
CaCO ₃ %	1.22
Available N %	0.12
Available P (Olsen method, ppm)	6.5
Available K (ammonium acetate, ppm)	460

- a₁) Application of the suitable N (1000 g N/ tree) completely via inorganic N source (2985.1 g ammonium nitrate / tree) .
- a₂) Application of the suitable N via 75% inorganic (2239 g ammonium nitrate / tree) + 12.5 % organic N (50 kg farmyard manure / tree) + 50 ml EM/ tree.
- a₃) Application of the suitable N via 62.5% inorganic N (1866 g ammonium nitrate / tree) + 12.5 % organic N (50 kg farmyard manure/ tree) + 100 ml EM/ tree.
- a₄) Application of the suitable N via 50 % inorganic N (1493 g ammonium nitrate / tree) + 12.5 % organic N (50 kg farmyard manure / tree) + 200 ml EM/ tree .
- a₅) Application of the suitable N via 37.5 % inorganic N (1119.4 g ammonium nitrate / tree) + 12.5 % organic N (50 kg farmyard manure / tree) + 400 ml EM/ tree .

The second factor (B) included two factors from amino acids enriched with NPKMgZnMn, Fe and B namely b₁) untreated trees (sprayed with water) and b₂) foliar application of amino acids (tryptophan + methionene + arginine) at 0.1% besides eight nutrients namely NPKMg, Zn, Mn, Fe and B. Therefore, this experiment included ten treatments, each replicated three times, one tree per each. Organic N source namely farmyard manure (F.Y.M.) was added once at the last week of Feb. for both seasons in deep trenches (50 cm length x 50 cm width x 50 cm depth) around each tree (under tree canopy (Table 2). Effective microorganisms (E M) (each ml contains 10⁷ bacterial cells) was added once at the first week of March during both seasons. It was applied in shallow trenches 930 cm length x 20 cm width x 10 cm depth) according to the recommendation of general organization for Agriculture Equalization Foundation (GOAEF),

Ministry of Agric. and Reclamation. The inorganic N source namely ammonium nitrate (33.5 % N) was splitted into three equal batches at the first week of March, May and July. It was broadcasted around each tree under tree canopy at 50 cm far from trunk. Irrigation was conducted after the addition of chemical and biofertilizers in both seasons. All the selected trees (30 trees) received N at fixed rate namely 1000 g N / tree / year. The three amino acids (tryptophan, methionene and arginine) was applied at 0.1%. The mixture of nutrients applied with the three amino acids contains urea (46 % N), orthophosphoric acid (55% P₂O₅), potassium sulphate, (48 % K₂O), and magnesium sulphate (9.6 % Mg) each at 0.2% (2 g. / I water); chelated Fe , Zn and Mn each at 0.05% and boric acid at 0.05%. Nutrient mixture and amino acid were sprayed three times at the first week of March, June and August. Triton B as a wetting agent was added to all amino acid and nutrient solutions at 0.05%. Spraying was done till runoff (50 I water / tree). Untreated trees sprayed with water containing Triton B.

Table (2): Analysis of the tested farmyard manure

Parameter	Values
Cubic meter weight (kg.)	650.0
Moisture %	35.0
O.M %	24.0
pH (1 : 10)	8.70
EC (mmhos cm ⁻¹ / 25°C)	5.70
C/N	17.5
Total N %	0.25
Total P %	0.32
Total K %	0.92
Total Ca %	1.83
Total Mg %	1.00
Total Fe (ppm)	1490
Total Mn (ppm)	500
Total Zn (ppm)	55

Statistical analysis was done using randomized complete block design (RCBD) in split plot arrangement. The five inorganic, organic and EM treatments and the two amino acids enriched with nutrient treatments ranked the main and subplots, respectively.

During both seasons the following measurements were recorded:

- 1- Leaf area (cm²) (**Ahmed and Morsy, 1999**).
- 2- Chlorophylls a & b in the fresh leaves (as mg/ 100 g F.W.) (**Von- Wettstein, 1957**) and total chlorophylls was estimated by summation of chlorophylls a and b.
- 3- In early Sept. twenty leaves (3rd leaf from the shoot base) from non fruiting shoot of spring growth cycle (**Summer, 1985**) for each tree were

- selected for determination of percentage of N, P, K and Mg (**Wilde et al., 1985**).
- 4- Percentages of Initial fruit setting and fruit retention.
 - 5- Number of fruits per tree and yield/ tree (kg.) were calculated (1st week of April).
 - 6- Physical and chemical characters of the fruits namely fruit weight (g.), T.S.S.%, total acidity % (as g. citric acid/ 100 ml juice); T.S.S./acid, total and reducing sugars % and vitamin C content (mg/ 100 ml juice) (**A.O.A.C., 2000**).
 - 7- The juice content of nitrate and nitrite (ppm)(**Ridnour- Lisa et al., 2000**).
 - 8- Amounts of CO₂ (mg/ 100 g soil) (**Paul and Clark, 1996**), nitrogenase activity (mg/ dwt/ g soil / one hour) (**Thalman, 1968 and Paul and Clark, 1996**) and total counts of bacteria (cfu/ 1.0 g soil) (**Waksman, 1952 and Abd El- Malek and Ischac, 1968**) in the soil.

Statistical analysis was done and treatment means were compared using new L.S.D. at 5% (**Mead et al., 1993**).

3.Results

1- Leaf area:

Data in Table (3) clearly show that using N as 50 to 75% inorganic N + 12.5 % organic (50 kg F.Y.M.) + 50 to 200 ml EM (tree significantly enhanced the leaf area rather than using N completely via. Inorganic N or when percentage of inorganic N was reduced to 37.5%. The promotion on the leaf area was significantly associated with reducing inorganic N percentages from 100 to 50% and increasing the levels of EM from 0.0 to 200 ml/ tree. The maximum values were recorded on the trees that received N via 50 % inorganic N + 12.5 % organic N + 200 ml EM/ tree. Using N as 37.5 % inorganic N + 12.5 organic + 400 ml EM/ tree gave the lowest values.

Foliar application of amino acids enriched with nutrients at 0.1% gave the maximum values over the check treatment.

Fertilizing of the trees with N as 50% inorganic + 12.5 % organic + 200 ml EM/ tree + Spraying amino acids enriched with nutrients at 0.1 % resulted in the maximum values. These results were true during seasons.

2- Leaf chemical composition:

It is clear from the data in Tables (3, 4 & 5) that amending the trees with N as 50 to 75% inorganic N + 12.5 % organic N + 50 to 200 ml EM/ tree significantly was accompanied with enhancing total chlorophylls and percentages of N, P, K and Mg in the leaves relatively to using N as 100% inorganic or when N was added as inorganic N at percentages lower than 50%. The promotion was significantly associated with reducing percentages of inorganic N from 100 to 50%

and increasing EM levels from 0.0 to 200 ml EM/ tree. The maximum values were recorded on the trees that fertilized with N as 50% inorganic + 12.5 organic + 200 ml EM/ tree.

Leaf content of total chlorophylls N, P, K and Mg was significantly increased by foliar application of amino acids enriched with nutrients at 0.1% over the check treatment.

The maximum values were recorded on the trees that received N as 50% inorganic N + 12.5 % organic + 200 ml EM/ tree + foliar fertilized with amino acids enriched with nutrients at 0.1% . These results were true during both seasons.

3- Fruit setting and yield:

Data in Tables (6 & 7) obviously reveal that using N as 50 to 75 % inorganic N + 12.0 % organic N + 50 to 200 ml EM/ tree significantly was accompanied with improving percentages of initial fruit setting and fruit retention, number of fruits / tree and yield rather than using N as 100% inorganic N or when N was applied as 37.5 % inorganic N + 12.5 % organic N + 400 ml EM/ tree.

Spraying amino acids enriched with nutrients at 0.1% significantly was superior than using the check treatment in improving fruit setting and yield.

Nitrogen fertilization on the basis of 50% inorganic N + 12.5 % organic N + 200 ml EM/ tree in combined with spraying the trees with amino acids enriched with nutrients at 0.1% gave the maximum values. These results were true during both seasons.

4- Fruit quality:

It is clear from the data in Tables (8 to 12) that application of N as 37.5% to 75% inorganic + 12.5% organic N + 50 to 400 ml EM/ tree significantly was very effective in improving fruit quality in terms of increasing fruit weight, T.S.S. %, total and reducing sugars %, T.S.S./ acid and vitamin C content and decreasing total acidity % nitrate and nitrite in the juice rather than using N as 100% inorganic N. The promotion was significantly associated with reducing percentages of inorganic N from 100 to 37.5 % and increasing EM levels from 0.0 to 400 ml/ tree. The most pronounced effect on fruit quality was attributed to using N as 37.5% inorganic N + 12.5 % organic + 400 ml EM/ tree.

Subjected the trees to amino acids enriched with nutrients at 0.1% caused a significant promotion on fruit quality over the check treatment.

Supplying Valencia orange trees with N as 37.5% inorganic N + 12.5 organic N + 400 ml EM/ tree in combined with spraying the trees with amino acids enriched with nutrients gave the best results with regard to fruit quality. Similar results were obtained during both seasons.

5- Amounts of CO₂, nitrogenase activity and total counts of bacteria in the soil:

It is clear from the data in Tables (12 & 13) that supplying the trees with N as 37.5 to 75 % mineral N + 12.5 % organic + 50 to 400 ml EM/ tree significantly enhanced the amounts of CO₂, nitrogenase activity and total counts of bacteria in the soil comparing with using N as 100% inorganic . The promotion on these characters was significantly depended on reducing percentages of inorganic N from 100 to 37.5% and at the same time increasing levels of EM from 0.0 to 400 ml/ tree / year. The maximum values were recorded when N was applied as 37.5% inorganic + 12.5 % organic N + 400 ml EM/ tree. Using N completely via inorganic N gave the lowest values.

Foliar application of amino acids enriched with nutrients at 0.1 % failed significantly to show any effect on amounts of CO₂, nitrogenase activity and total counts of bacteria over the check treatment.

The highest values were recorded on the trees that fertilized with N as 37.5% inorganic + 12.5 organic N + 400 ml EM/ tree without foliar application of amino acids enriched with nutrients. These result were true during both seasons.

4. Discussion:

Table (3): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on leaf area in the spring growth cycle (cm²) and the total chlorophylls (mg/ 100 g F.W.) in the leaves of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Leaf area in the spring growth cycle (cm ²)						Total chlorophylls (mg/ 100 g F.W)					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	16.1	18.2	17.2	16.2	18.3	17.3	34.7	39.4	37.1	35.1	39.3	37.2
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	18.1	20.3	19.2	18.7	21.0	19.9	39.2	44.2	41.7	39.7	44.5	42.1
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	20.1	22.3	21.2	21.0	23.9	22.5	44.6	49.8	44.7	45.0	50.9	48.0
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	22.2	24.5	23.4	23.3	26.0	24.7	49.4	54.8	52.1	49.9	55.0	52.5
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	14.0	16.2	15.1	14.6	17.3	15.9	30.1	34.8	32.5	31.0	35.1	33.1
Mean (B)	18.1	20.3		18.8	21.3		39.6	43.6		40.1	45.0	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	1.3	1.0	2.2	1.4	1.2	2.7	1.9	1.8	4.0	2.1	2.0	4.5

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (4): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the percentages of N and P in the leaves of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Leaf N %						Leaf P %					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	1.81	1.92	1.87	1.85	1.95	1.90	0.14	0.17	0.16	0.13	0.16	0.15
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	1.91	2.01	1.96	2.00	2.11	2.06	0.18	0.22	0.20	0.17	0.20	0.19
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	2.05	2.17	2.11	2.11	2.22	2.17	0.22	0.26	0.24	0.23	0.27	0.25
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	2.20	2.31	2.26	2.21	2.33	2.27	0.26	0.31	0.29	0.28	0.32	0.30
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	1.74	1.82	1.78	1.71	1.82	1.77	0.11	0.13	0.12	0.10	0.13	0.12
Mean (B)	1.94	2.05		1.98	2.09		0.18	0.22		0.18	0.22	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	0.07	0.06	0.13	0.06	0.05	0.11	0.3	0.2	0.4	0.3	0.2	0.4

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (5): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the percentages of K and Mg in the leaves of Valencia orange trees during 2011/2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Leaf K %						Leaf Mg					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	1.11	1.17	1.14	1.13	1.20	1.17	0.29	0.36	0.33	0.30	0.35	0.33
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	1.20	1.28	1.24	1.25	1.32	1.29	0.36	0.46	0.41	0.41	0.47	0.44
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	1.30	1.40	1.35	1.35	1.42	1.39	0.43	0.52	0.48	0.50	0.57	0.54
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	1.49	1.55	1.52	1.55	1.63	1.59	0.51	0.61	0.56	0.57	0.66	0.62
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	1.05	1.10	1.08	1.04	1.12	1.08	0.23	0.31	0.27	0.24	0.30	0.27
Mean (B)	1.23	1.30		1.26	1.34		0.36	0.45		0.40	0.47	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	0.05	0.04	0.09	0.06	0.05	0.11	0.05	0.04	0.09	0.05	0.04	0.09

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (6): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the percentages of initial fruit setting and fruit retention of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Initial fruit setting %						Fruit retention %					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	14.7	15.8	15.3	15.0	16.3	15.7	0.88	0.99	0.94	0.95	0.99	0.97
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	15.9	17.0	16.5	16.1	17.4	16.8	0.94	1.06	1.00	1.01	1.15	1.08
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	17.1	18.8	18.0	17.3	18.5	17.9	1.00	1.27	1.14	1.11	1.33	1.22
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	18.3	21.5	19.9	18.5	19.8	19.2	1.08	1.40	1.24	1.20	1.45	1.33
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	13.0	14.6	13.8	13.8	15.2	14.5	0.80	0.90	0.85	0.82	0.89	0.86
Mean (B)	15.8	17.5		16.1	17.4		0.94	1.12		1.02	1.16	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	1.0	0.9	2.0	1.1	1.0	2.2	0.05	0.04	0.09	0.06	0.04	0.09

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (7): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the number of fruits per tree and yield per tree (kg.) of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Number of fruits / tree						Yield per tree (kg.)					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	311.0	341.0	326.0	313.0	345.0	329.0	43.9	50.9	47.2	43.8	51.1	47.5
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	341.0	375.0	358.0	345.0	381.0	363.0	53.0	62.3	57.7	52.8	62.5	57.7
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	381.0	415.0	398.0	391.0	431.0	411.0	64.4	73.5	69.0	65.3	75.4	70.4
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	401.0	441.0	421.0	416.0	471.0	443.5	72.2	83.3	77.8	74.0	88.1	81.1
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	151.0	171.0	161.0	161.0	181.0	171.0	28.5	33.7	31.1	30.1	35.1	32.6
Mean (B)	317.0	348.0		325.0	361.8		52.4	60.7		53.2	62.4	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	18.3	17.8	39.9	19.0	18.1	40.5	5.5	4.9	11.0	6.1	5.8	13.0

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (8): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the average fruit weight (g.) and T.S.S. in the fruits of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Av. Fruit weight (g.)						T.S.S. %					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	141.3	149.3	145.3	140.0	148.0	144.0	11.5	11.9	11.7	11.4	11.8	11.6
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	155.5	166.0	160.8	153.0	164.0	158.5	11.9	12.4	12.2	11.9	12.4	12.2
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	169.0	177.0	173.0	167.0	175.0	171.0	12.5	12.9	12.7	12.6	13.0	12.8
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	180.0	189.0	184.5	178.0	187.0	182.5	13.0	13.5	13.3	13.1	13.5	13.3
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	189.0	197.0	193.0	187.0	194.0	190.5	13.3	13.8	13.6	13.9	14.3	14.1
Mean (B)	167.0	175.7		165.0	173.6		12.4	12.9		12.6	13.0	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	7.1	6.8	15.2	6.8	6.5	4.6	0.3	0.2	0.4	0.4	0.3	0.7

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (9): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the percentage of total acidity and T.S.S. / acid in the fruits of Valencia orange trees during 2011/2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Total acidity %						T.S.S. / acid					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	1.437	1.410	1.424	1.425	1.401	1.413	8.0	8.4	8.2	8.0	8.4	8.2
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	1.410	1.380	1.395	1.391	1.351	1.371	8.4	9.0	8.7	8.6	9.2	8.9
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	1.371	1.341	1.356	1.361	1.321	1.341	9.1	9.6	9.4	9.3	9.8	9.6
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	1.316	1.276	1.296	1.311	1.271	1.291	9.9	10.6	10.3	10.0	10.6	10.3
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	1.281	1.210	1.456	1.270	1.231	1.251	10.4	11.4	10.9	10.9	11.6	11.3
Mean (B)	1.363	1.323		1.352	1.315		9.2	9.8		9.4	9.9	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	0.021	0.018	0.040	0.018	0.016	0.036	0.4	0.4	0.9	0.5	0.4	0.9

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (10): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the percentages of total and reducing sugars in the fruits of Valencia orange trees during 2011/2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Total sugars %						Reducing sugars %					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	7.1	8.0	7.6	7.2	8.2	7.7	3.1	3.5	3.3	3.3	3.8	3.6
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	7.8	8.7	8.3	7.9	8.9	8.4	3.6	4.1	3.9	3.9	4.4	4.2
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	8.7	9.6	9.2	8.8	9.9	9.4	4.1	4.6	4.4	4.9	5.4	5.2
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	9.5	10.5	10.0	9.6	9.7	9.7	4.6	5.1	4.9	5.6	6.1	5.9
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	10.7	11.6	11.2	10.9	11.9	11.4	5.1	5.6	5.4	6.0	6.5	6.3
Mean (B)	8.8	9.7		8.9	9.7		4.1	4.6		4.7	5.2	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	0.6	0.5	1.1	0.7	0.5	1.1	0.4	0.3	0.7	0.5	0.4	0.9

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (11): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the vitamin C content (mg/ 100 ml/ juice) and nitrate in the juice of the fruits of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Vitamin C(mg/ 100 ml juice)						Nitrate in the juice (ppm)					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	41.0	44.4	42.7	41.9	45.9	43.9	4.8	3.7	4.3	4.7	3.6	4.2
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	43.6	47.0	45.3	44.7	48.3	46.5	4.0	3.0	3.5	3.8	2.8	3.3
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	45.3	49.0	47.2	46.2	51.3	48.8	3.1	2.0	2.6	2.9	1.9	2.4
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	47.9	51.0	49.5	48.8	53.0	50.9	2.5	1.5	2.0	2.3	1.3	1.8
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	51.0	54.0	52.5	52.2	56.9	54.6	1.2	0.7	1.0	1.0	0.7	1.4
Mean (B)	45.8	49.1		46.8	51.1		3.1	2.2		3.1	2.1	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	1.8	1.6	3.6	1.7	1.5	3.4	0.5	0.4	0.9	0.6	0.4	0.9

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (12): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the nitrite in the juice of the fruits as well as CO₂ amount (mg/ 100 g soil) in the soil of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Nitrite in the juice (ppm)						CO ₂ (mg/ 100 g soil)					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	2.20	2.05	2.13	2.22	2.09	2.16	11.2	11.2	11.2	11.3	11.5	11.4
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	2.00	1.80	1.90	1.91	1.71	1.81	13.8	14.0	13.9	15.0	15.1	15.1
a ₃ N at 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	1.71	1.41	1.56	1.71	1.01	1.36	15.3	15.3	15.3	16.5	16.6	16.6
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	1.11	0.90	1.01	1.01	0.73	0.87	18.9	19.0	19.0	20.1	20.3	20.2
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	0.81	0.61	0.71	0.71	0.61	0.66	21.9	22.0	22.0	23.1	23.2	23.2
Mean (B)	1.57	1.35		1.51	1.23		16.2	16.3		17.2	17.3	
New L.S.D. at 5%	A	B	AB	A	B	AB	A	B	AB	A	B	AB
	0.11	0.10	0.22	0.12	0.11	0.25	1.1	NS	2.2	1.3	NS	2.3

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

Table (13): Effect of inorganic and organic N fertilization, biofertilization with EM and foliar application with nutrients on the nitrogenase activity (mg/ dwt/ g. soil) and total counts of bacteria (cfu/ g soil) in the soil orchard of Valencia orange trees during 2011/ 2012 and 2012 / 2013 seasons.

Inorganic and organic N as well as EM treatments (A)	Nitrogenase activity (mg/ dwt. g soil / one hour)						Total counts of bacteria (cfu/ g soil)					
	2011/ 2012			2012/ 2013			2011/ 2012			2012/ 2013		
	Amino acids enriched with nutrients (B)											
	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)	0.0 %	0.1 %	Mean (A)
a ₁ N as 100 % inorganic	0.111	0.112	0.112	0.115	0.116	0.116	6.1	6.1	6.1	6.5	6.6	6.6
a ₂ N as 75% inorg. + 12.5 org + 50 ml EM / tree	0.150	0.152	0.151	0.171	0.171	0.171	6.6	6.6	6.6	7.1	7.2	7.2
a ₃ N as 62.5 % inorg. + 12.5 % org. + 100 ml EM / tree	0.191	0.195	0.193	0.213	0.215	0.214	6.9	6.9	6.9	7.5	7.5	7.5
a ₄ N as 50% inorg. + 12.5 % org. + 200 ml EM/ tree	0.311	0.313	0.312	0.341	0.345	0.343	7.4	7.5	7.5	8.1	8.2	8.2
a ₅ as 37.5 % inorg. + 12.5 % org. + 400 ml EM/ tree	0.381	0.381	0.381	0.400	0.411	0.406	7.8	7.9	7.8	8.4	8.5	6.5
Mean (B)	0.229	0.231		0.248	0.252		7.6	7.0		7.5	7.6	
	A	B	AB	A	B	AB	A	B	AB	A	B	AB
New L.S.D. at 5%	0.011	NS	0.025	0.015	NS	0.027						

Inorg. = Inorganic N form (ammonium nitrate, 3.5 % N)

Org. = organic N form (Farmyard manure 0.25 % N)

The great merits of organic and biofertilization on fruiting of Valencia organic trees might be attributed to their positive action on enhancing soil fertility and the uptake of nutrients, nitrogenase activity and CO₂ amounts (Mengel *et al.*, 2001 and Kannaiyan, 2002).

These results are in agreement with those obtained by Abdelaal *et al.*, (2012); and Faraag (2013).

The beneficial effects of EM on growth and yield of Valencia orange trees was mainly attributed to its positive action on enhancing soil fertility and uptake of most nutrients, nitrogenase activity, total counts of bacteria and CO₂ amounts. (Kannaiyan, 2002). These results were supported by the findings of Ahmed – Samah (2011) and Ibrahiem (2012).

The important roles of amino acids in enhancing the biosynthesis of proteins and natural hormones (Davies, 1982) could result in enhancing fruiting of Valencia orange trees. The same trend was emphasized by the result of Hassan (2014); Ahmed *et al.*, (2014a) and (2014 b) and Sayed- Ola (2014) .

The essential regulatory effects of nutrients on enhancing the biosynthesis of organic foods, natural hormones, amino acids and plant pigments as well as cell division (Mengel *et al.*, 2001) surely reflected on improving productivity of Valencia orange trees. These results were confirmed by the results of Gamal (2013) and Ibrahiem and Al- Wasfy (2014) .

Conclusion:

For reducing pollution by nitrate and nitrite as well as improving yield and quality of Valencia orange trees, it is advised to use N (1000 g N/ tree) via 50% inorganic N + 12.5 % organic + 200 ml EM/ tree + carrying out three sprays of amino acids at 0.1 % enriched with NPK, Mg, Zn, Fe, Mn and B.

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