

## THE HUMIC ACID AND ROOTSTOCK IN ENHANCING SALT TOLERANCE OF 'ANNA' APPLE SEEDLINGS

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### ABSTRACT

'Anna' apple plants (*Malus domestica* Borkh) budded on *M. communis* or MM106 rootstocks were observed under 0, 1000, and 2000 ppm saline irrigation water that contained equal parts of NaCl, CaCl<sub>2</sub>, MgSO<sub>4</sub> and NaHCO<sub>3</sub> salts. Also, the effect of humic acid treatments (soil, foliar, and soil + foliar application) on the growth parameters (shoot length, number of leaves, leaf area and leaf chlorophyll content), nutritional status (percentage of leaf dry matter and NPK content), root system growth (root length, number of roots, and dry matter of main and secondary roots), toxic ions (chloride and sodium leaf content), and proline amino acid was studied. Salinity treatments significantly decreased growth parameters, nutritional status and root system growth, and in the contrary, they increased toxic ions and proline amino acid content. *M. communis* rootstock tolerated salinity more than MM106 stock, since 'Anna' apple plants on *M. communis* grew better under different salinity doses than on MM106 roots. Moreover, humic acid application (especially soil treatment with 20 ml Actosol in 1 L of water per 35-cm-pot every other week from late June to Oct.15<sup>th</sup>) markedly minimized the harmful effect of salinity and enhanced apple salt tolerance.

### INTRODUCTION

Large land areas have been identified as being adversely affected by salinity (Lewis, 1984). Additional land area world-wide could be degraded by salinity. During the 21<sup>st</sup> century of the wide variety of crops grown, deciduous fruit species are considered to be among the most sensitive to salts (Maas & Hoffman, 1977 and Fathi & Catline, 1994). Accumulation of potentially toxic concentrations of sodium and chloride ions in leaves is dependent on the capability of the root system to exclude ions (Walker *et al.*, 1983). Rootstock varieties differ widely in their physiological characteristics associated with water and ion relations (Syvertsen & Graham, 1985). Salt tolerance and physiological consequences of toxic ions in the scion comes from short-term controlled environmental studies using young container grown seedlings or budded trees (Walker, 1986). Moreover, salinity has been found to be inversely related to plant dry weight and maximum root length with lesser effect on root than on top growth. It clearly decreased uptake of N, P, K, Ca, and S; increased uptake of Mg, Mn, Mo, B, Cl, and Na; while it had no influence on the uptake of Fe, Cu, and Zn (Liu and Cooper, 2002).

Humic acid (polymeric polyhydroxy acid) is the most significant component of organic substances in aquatic systems (Mecan and Petrovic, 1995). Liu & Cooper (2002) have showed that humic acid enhanced root growth of salt-stressed plants and improved salinity tolerance. Also, Hartwigsen & Evans (2000) indicated that humic acid significantly increased root fresh weight and total length of lateral roots.

Accordingly, this investigation was carried out to study the response of 'Anna' apple on two rootstocks, viz., *M. communis* and MM106 to irrigation with saline water and the role of rootstocks in increasing salt tolerance of the

scion. The possibility of using humic acid as a soil conditioner to reduce the harmful effects of salinity was also determined.

## **MATERIALS AND METHODS**

This investigation was conducted during the 2004 and 2005 seasons in the orchard of the Horticulture Research Institute, Agricultural Research Center, Giza. Treatments were applied to 'Anna' apple one-year-old seedlings of either *M. communis* or MM106 as rootstocks.

A split-split plot system in a randomized complete block design was used with three replicates. Each experimental unit consisted of 3 pots, each containing one seedling. Pots were 35 x 50 cm and were filled with a mixture of 15 kg sand + 100 g peatmoss. These pots were planted during February in the two seasons. The two rootstocks were allocated to the main plots. Watering was done using tap water until the end of June in each season. Thereafter, salinity treatments were applied until Oct 15<sup>th</sup> twice a week as sub-plots. They were 0, 1000, and 2000 ppm of a mixture of equal parts by weight of sodium chloride, calcium chloride, magnesium sulfate, and sodium bicarbonate salts. Sub-sub treatments were applied every other week during the same period, i.e., from July 1<sup>st</sup> to Oct. 15<sup>th</sup> using humic acid (in the form of Actosol) as follows: (a) soil application at the rate of 20 ml Actosol in 1 L water, (b) foliar application with 0.5 % Actosol solution + soil application as above, (c) foliar application as above and (d) control. Actosol is a commercial product that contains 2.9 % humic acid and 10-10-10 NPK. It is manufactured by Arctick Inc., Chentilly, VA, USA.

Foliage measurements included the following characters: (a) relative shoot length expressed as percentage of shoot length and relative number of leaves as percentage of number of leaves compared to control which were recorded in August, September and October of both tested seasons and (b) leaf area and leaf chlorophyll content as measured on Aug. 20<sup>th</sup> on 20 fully-expanded leaves per seedling and sampled from the middle of shoots. Leaf area was recorded using a CI203Area Meter (CID, Inc., USA), while a SPAD 502 chlorophyll meter (Minolta Corporation, Ramsey, N.J., USA) was used in recording chlorophyll readings.

Subsequently, in December of both seasons, measurements were made on the percentage of dry matter in vegetative growth, i.e. remaining leaves and stems, and in main and secondary roots.

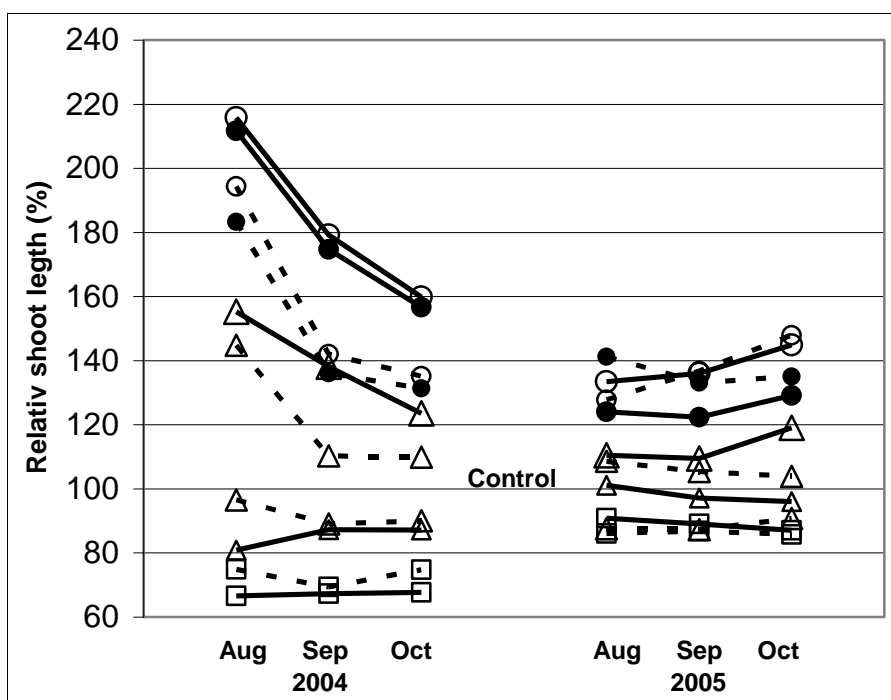
Chemical analysis was made on leaf samples to determine some mineral elements content. Samples were taken from an intermediate position on scion shoots in August. Leaves were first washed several times with tap water; then, they were washed with distilled water and 0.1 N HCl, dried at 70 °C, till constant weight and finely ground. Samples, 0.5 g each, were digested using H<sub>2</sub>SO<sub>4</sub>-H<sub>2</sub>O<sub>2</sub> as described by Cottenie (1980). Then, extracts were prepared for chemical analysis as described by Jackson (1973). Nitrogen was determined according to the modified Kjeldahl method as described by A.O.A.C. (1975). Phosphorus content was colorimetrically estimated according to Troug and Meyer (1939). Wet digestion was used for the determination of potassium as described by Piper (1950) using a flame

photometer according to Brown and Lilleland (1946). Sodium was determined by using flame photometer (Brown and Lilleland, 1946). Chloride content was assessed according to the methods of Higinbotham *et al.* (1967). Proline content was then colorimetrically estimated at 520 nm according to Bates *et al.* (1973). The obtained data were statistically analysed according to Snedecor & Cochran (1990). Mean separation was calculated using LSD values at the 5 % level.

## RESULTS

### Growth Parameters:

Growth parameters of 'Anna' apple plants budded on MM106 or *M. communis* rootstocks included shoot length (Fig.1), number of leaves (Fig.2), leaf area, and leaf chlorophyll content (Table 1).



**Fig. (1):** Relative shoot length (expressed as percentage of length in the control) as affected by humic acid treatments (O soil,  $\Delta$  foliar and  $\bullet$  soil + foliar) and salinity treatments ( $\times$  1000 and  $\square$  2000 ppm) for Anna apple budded on MM.106 (-) or *Malus communis* (-----).

Concerning the effect of humic acid, it markedly, enhanced 'Anna' apple plants to produce longer shoots, more leaves with larger expansion and with higher chlorophyll content, i.e., leaf area was 27.1, 54.2, 43.7, and 51.9 cm<sup>2</sup> and chlorophyll leaf content was 40.4, 48.5, 41.7, and 41.5 SPAD readings in the treatments: control, soil, foliar, and soil + foliar humic acid

application, respectively (table 1). The differences were mostly significant. However, soil treatment usually was better, i.e., it enhanced shoot length to 215.8 % and number of leaves to 330.0 % of the respective measurements in the control treatment.

Regarding the effect of rootstock, *M. communis* apple stock recorded better shoot length in the 1<sup>st</sup> season, leaf area in the 2004 season, and chlorophyll content in both season of the study; while MM 106 stock recorded better shoot length and leaf area in the 2005 season.

Salinity treatments significantly decreased shoot length, which declined from 65.2 % to 67.3 %, and then to 67.7 % as salinity level increased, compared to control (100 %), during Aug., Sep., and Oct., respectively.

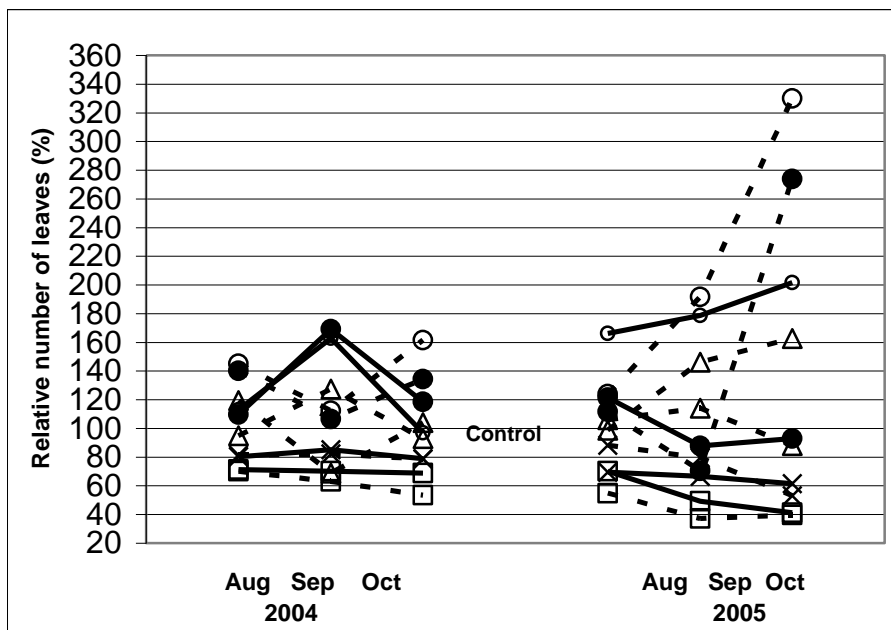


Fig. (2): Relative number of leaves (expressed as percentage of number in the control) as affected by humic acid treatments (O soil, Δ foliar and ● soil + foliar) and salinity treatments (× 1000 and □ 2000 ppm) for Anna apple budded on MM.106 ( - ) or *Malus communis* (-----) .

**Nutritional status:**

We assessed the percentage of leaf content of dry matter (Table 1) and NPK elements (Table 2) as a guide to the nutritional status. Humic acid treatments helped and improved the nutritional status than control even under salinity stress. Meanwhile, soil application was the most effective, as it raised leaves dry matter to 63.5%, N to 2.53 %, P to 0.36 %, and K to 1.43 % comparing to control (46.7 %, 1.16 %, 0.21 %, and 0.94 %, respectively).

T1

Furthermore, *Malus communis* apple rootstock enhanced 'Anna' apple leaves to deposit more dry matter (53.0 %), N (1.9%) and K (1.35 %) than MM106 stock (51.2 %, 1.063% and 1.13 %, respectively). However, MM106 resulted in higher P (0.27 %).

It is quite clear that leaf dry matter and NPK content effectively depressed as a function of salinity, i.e. dry matter decreased from 53.9 % to 52.1 %, then to 50.3 % (season 2004, table 1), nitrogen from 2.55 % to 1.9 %, then to 1.3 %, phosphorous from 0.3 % to 0.22 %, then to 0.20 % as well as potassium from 1.52 % to 1.6 %, then to 1.05 % (season 2005, table 2) when salinity increased from 0 to 1000, then to 2000 ppm respectively. Statistical analysis confirmed most of these differences.

Concerning the interaction effect, *M. communis* rootstock affected less by the deleterious salinity effect, while responded better to humic acid treatments.

#### **Root system growth:**

Root system growth (Figs. 3 and 4) included observation of root length (Table 3) and number of roots (Table 4) separated according to diameter to < 0.5, 0.5-1.5 and > 1.5 cm. We also assessed the percentage of dry matter of main and secondary roots (Table 5) as a remark of root system growth. When humic acid was applied to salt-treated plants, it was noticed that root length and number as well as dry matter of main and secondary roots significantly improved and effectively overcompensated the negative effect of salinity. The most effective treatment was soil application which produced longer roots (15.33, 32.25 and 22.0 cm) with more number (0.88, 8.43 and 18.93) and higher dry matter (56.6 % and 44.5 %) than the other treatments or control.

Though the root growth of 'Anna' apple budded on *Malus communis* or MM106 was restricted by salinization, *M. communis* roots resisted this conditions to some extent by producing extra root hairs and side branches (figs 3 and 4). This condition helps in improving root-soil contact; therefore, enabling the root system to absorb minerals and water at relatively higher rates which finally affect plant growth.

Likewise, saline irrigation water had a limiting effect on root elongation (root length) and branching (number of roots) as well as root nutritional status (dry matter) where the three root categories gradually decreased as salinity dose increased.

#### **Toxic ions content and proline:**

Humic acid treatments could successfully minimize the deleterious effect of saline irrigation water. Soil application was the most effective treatment where it limited the accumulation of Na<sup>+</sup> to 0.08 %, Cl<sup>-</sup> to 0.014 % and proline to 0.011 mg/g, comparing to control (1.40 %, 0.017 % and 0.039 mg/g, respectively) or other humic acid treatments.

However, this increase of toxic ions and proline (Table 6) was more pronounced with MM106 rootstock than with *M. communis* stock. Moreover, there is as important evidence that the roots of *M. communis* stock successfully relayed less quantities of toxic ions and proline to 'Anna' leaves.

T2





t4



t6

**Fig. (3): Effect of salinity and humic acid treatment (soil application) on root growth of 'Anna' apple on *Malus communis* rootstock at 2000ppm salinity.**

**Fig. (4): Effect of salinity and humic acid treatment (soil application) on root growth of 'Anna' apple on MM106 rootstock at 2000ppm salinity.**

This condition may explain the better growth parameters, nutritional status and root system growth of 'Anna' apple plants on *M. communis* than on MM106 apple even under salinity conditions.

The concentration of sodium (0.41%, 0.64 %, and 0.66%), chloride (0.011 %, 0.019 %, and 0.022 %) and the proline amino acid (0.005, 0.025 and 0.055 mg/g) gradually increased with increasing salt level from 0 to 1000, then to 2000 ppm, respectively.

## DISCUSSION AND CONCLUSION

From the above mentioned results, we can conclude that salinity of irrigation water exerted harmful effect on plant growth parameters (percentage of growth rate as shoot length and percentage of metabolism conductivity as number of leaves, leaf area and leaf chlorophyll content). Salinity adversely affected nutritional status where percentage of leaf dry matter and NPK content declined due to salt accumulation in both 'Anna' apple plants budded on *M. communis* or MM106 rootstocks and in the soil. Saline irrigation water had a restrictive effect on root elongation (root length) and branching (number of roots) as well as root nutritional status (dry matter of main and secondary roots). As salinity level increased a subsequent increase was observed in leaf Na<sup>+</sup>, Cl<sup>-</sup> and amino acid proline content, especially at high concentration of salts (2000 ppm). These results are in line with those obtained by Sweidan *et al.* (1992) on almond and peach rootstocks, El-Shall & Fathi (1993) as well as El-Shall *et al.* (1993) on 'Anna' apple, Fathi (1994) on apples, Fathi & Catlin (1994) on some *Prunus* rootstocks, Hussein (1998) and Salem *et al.* (1999) on apples, and Hussein (2004) on pears who reported that salinity exhibited severe effects by causing more branch stunting, more leaf shedding and reduction in chlorophyll content and nutrient elements absorption; subsequently, depressing plant nutritional status.

Taking rootstocks into consideration, MM106 apple rootstock recorded less growth parameters and nutritional status accompanied with less perforated root system and more toxic ions and proline amino acid. However, the nutritional status shortage showed by MM106 may be the result of: (1) the increase in Na<sup>+</sup> uptake which caused ionic imbalance in the plant by depressing N, P and K uptake (Farugge, 1968) and (2) the increase in pH of the root medium thus making many nutrients unavailable to the plants (Russell, 1982). On the other hand, *M. communis* roots effectively can resist salinity conditions to some extent by producing extra root hairs and side branches. This condition helps in improving the contact between roots and soil. Consequently, the root system gains the ability to absorb minerals and water at relatively higher rates, which finally affect plant growth as has been reported by (El-Shall and Fathi (1993), El-Shall *et al.* (1993) and Fathi (1994) on some apple stocks. Moreover, Hussein (1998) and Salem *et al.* (1999) on apples disclosed that *M. communis* roots successfully relayed lesser quantities of toxic ions (Na<sup>+</sup> and Cl<sup>-</sup>) to 'Anna' apple leaves; thereby, enhancing better growth.

When humic acid was applied to salt treated plants, growth parameters, nutritional status and root system growth were better and effectively overcame the deleterious effects of saline irrigation water. However, soil application was the most effective one. Besides, other reporters explained that humic substances increased dry matter of foliage and roots, promoted lateral root growth and N uptake rate (Tattini *et al.*, 1991), contributed to the nutritional regulation and adaptability of apple trees and enhanced photosynthesis and accumulation of nutrients (Jianguo *et al.*, 1998). In addition, Fathi *et al.* (2002) obtained the best significant results with 'Desert Red' peach by spraying trees with 5 pp GA<sub>3</sub> + 0.2 % K-Humate.

So, we can recommend apple nursery growers to (1) avoid irrigating apples seedlings with saline water more than 1000 ppm, (2) use the rootstock *M. communis* especially under salinity conditions, and (3) use soil application of humic acid at the rate of 20 ml Actosol (10-10-10 NPK)/1 L of water to each seedling pot every other week during the period from end of June till Oct.15<sup>th</sup> to minimize the harmful effects of salinity

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دور حامض الهيوميك والاصل فى زيادة مقاومة شتلات التفاح انا لملوحة ماء  
الرى  
فوزية محمد عيسى، مصطفى أحمد فتحى وسعد عبد الواحد الشال  
معهد بحوث البساتين-مركز البحوث الزراعية

شملت الدراسة نباتات بعمر سنة من صنف التفاح انا مطعومة على اصل مالص كميونس  
أو م.م ١٠٦ ومزروعة فى أكياس رويت بماء مالح بتركيز: صفر أو ١٠٠٠ أو ٢٠٠٠ جزء فى  
المليون يحتوى على أملاح: كلوريد الصوديوم + كلوريد الكالسيوم + كبريتات الماغنسيوم +  
بيكربونات الصوديوم.

كما تمت دراسة تأثير معاملة النباتات بحامض الهيوميك (معاملة التربة أو الرش على  
المجموع الخضرى أو هما معاً) على صفات النمو الخضرى (طول الفرع، وعدد الأوراق، ومساحة  
الورقة، ومحتوى الأوراق من الكلوروفيل) والحالة الغذائية للنبات (% للمادة الجافة فى الأوراق،  
ومحتوى الأوراق من عناصر النيتروجين والفوسفور والبوتاسيوم) وكذلك نمو المجموع الجدرى  
للنبات (طول وعدد الجذور، والنسبة المئوية للمادة الجافة فى الجذور الرئيسية والفرعية) وأيضاً  
محتوى الأوراق من عنصرى الصوديوم والكلور والحامض الأمينى برولين.  
أظهرت النتائج أن معاملات الملوحة قللت بوضوح النمو الخضرى والحالة الغذائية للنبات  
وكذلك نمو المجموع الجدرى، لكنها ساعدت على زيادة محتوى الأوراق من عنصرى الصوديوم  
والكلور والحامض الأمينى برولين.

أصل التفاح مالص كميونس كان أكثر تحملاً للملوحة حيث شجع كل مظاهر النمو فى  
نباتات التفاح انا كما أعاق امتصاصها للعناصر السامة (الصوديوم والكلور) عنه فى حالة استخدام  
الأصل م.م ١٠٦.

معاملة النباتات بحامض الهيوميك (خصوصاً معاملة التربة ب-٢٠ مل أكتوسول (٢,٩ %  
هيوميك أسيد، و ١٠-١٠-١٠ NPK) فى لتر ماء مرة كل أسبوعين من أواخر يونيو حتى منتصف  
أكتوبر) قللت بوضوح التأثيرات الضارة للملوحة على نباتات التفاح على الأصلين موضوع الدراسة.





Table (1): Effect of humic acid (A) and salinity of irrigation water (C) on leaf area, percentage of dry matter and leaf chlorophyll content of apple seedlings (B): on *Malus communis* (Mc) and MM106 rootstocks

(A)	(C)	Leaf area (cm <sup>2</sup> )						% Dry matter						Chlorophyll (SPAD reading)					
		2004 season			2005 season			2004 season			2005 season			2004 season			2005 season		
		MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)
Control	0	21.9	26.1	24.0	27.3	33.2	30.3	52.6	52.2	52.4	52.2	55.2	53.7	43.4	45.9	44.7	38.8	42.9	40.9
	1000	17.4	25.9	21.7	24.8	30.5	27.7	44.4	49.8	47.1	48.5	50.9	49.7	33.6	45.6	39.6	39.5	38.0	38.8
	2000	15.8	21.6	18.7	22.6	24.4	23.5	39.3	41.9	40.6	49.2	54.0	51.6	33.7	40.4	37.1	27.3	30.8	29.1
Ave. (A x B)		18.4	24.5	Ave (A) 21.5	24.9	29.4	Ave (A) 27.1	45.4	48.0	Ave (A) 46.7	50.0	53.4	Ave (A) 51.7	36.9	44.0	Ave (A) 40.4	35.2	37.2	Ave (A) 36.2
Soil	0	34.3	36.7	35.5	54.9	56.1	55.5	61.4	66.2	63.8	62.6	67.7	65.2	49.4	50.4	49.9	48.2	47.8	48.0
	1000	33.5	35.6	34.6	54.3	54.1	54.2	64.3	67.8	66.1	64.4	63.9	64.2	47.4	48.1	47.8	41.5	46.1	43.8
	2000	31.8	31.3	31.6	53.3	52.3	52.8	58.0	63.2	60.6	61.1	62.0	61.6	48.1	47.8	48.0	36.6	43.7	40.2
Ave. (A x B)		33.2	34.5	Ave (A) 33.9	54.2	54.2	Ave (A) 54.2	61.2	65.7	Ave (A) 63.5	62.7	64.5	Ave (A) 63.6	48.3	48.8	Ave (A) 48.5	42.1	45.9	Ave (A) 44.0
Foliar	0	30.9	34.6	32.8	56.1	33.9	45.0	50.7	49.3	50.0	52.8	52.2	52.5	40.2	47.5	43.9	38.8	43.6	41.2
	1000	26.3	32.4	29.4	51.5	37.9	44.7	47.2	48.2	47.7	47.5	48.8	48.2	39.7	43.3	41.5	38.9	43.7	41.3
	2000	24.9	30.9	27.9	48.7	33.8	41.3	50.2	52.0	51.1	48.0	49.3	48.7	36.5	42.8	39.7	34.3	42.8	38.6
Ave. (A x B)		27.4	32.6	Ave (A) 30.0	52.1	35.2	Ave (A) 43.7	49.4	49.8	Ave (A) 49.6	49.4	50.1	Ave (A) 49.8	38.8	44.5	Ave (A) 41.7	37.3	43.4	Ave (A) 40.4
Soil foliar	0	30.5	32.8	31.7	57.8	51.2	54.5	49.2	49.5	49.4	57.3	62.4	59.9	42.7	46.9	44.8	43.2	46.8	45.0
	1000	27.8	32.8	30.3	55.6	48.8	52.2	48.7	46.2	47.5	56.6	52.8	54.7	40.8	44.3	42.6	42.4	43.3	42.9
	2000	28.0	30.9	29.5	51.4	46.6	49.0	48.1	49.8	49.0	55.6	54.9	55.3	38.6	35.5	37.1	30.6	38.6	34.6
Ave. (A x B)		28.8	32.2	Ave (A) 30.5	54.9	48.9	Ave (A) 51.9	48.7	48.5	Ave (A) 48.6	56.5	56.7	Ave (A) 56.6	40.7	42.2	Ave (A) 41.5	38.7	42.9	Ave (A) 40.8
Ave. (B x C)	0	29.4	32.6	Ave (C) 31.0	49.0	43.6	Ave (C) 46.3	53.5	54.3	Ave (C) 53.9	56.2	59.4	Ave (C) 57.8	43.9	47.7	Ave (C) 45.8	42.3	45.3	Ave (C) 43.8
	1000	26.3	31.7	29.0	46.6	42.8	44.7	51.2	53.0	52.1	54.2	54.1	54.2	40.4	45.3	42.9	40.6	42.8	41.7
	2000	25.1	28.7	26.9	44.0	39.3	41.7	48.9	51.7	50.3	53.5	55.1	54.3	39.2	41.6	40.4	32.2	39.0	35.6
Ave. (B)		26.9	31.0		46.5	41.9		51.2	53.0		54.6	56.2		41.2	44.9		38.4	42.4	

LSD at 5% for:

Humic acid (A)	2.51	3.12	4.22	3.14	3.14	2.76
Apple rootstock (B)	2.18	2.70	3.65	2.72	2.72	2.39
Salinity (C)	2.18	2.70	3.65	2.72	2.72	2.39
A x B	3.55	4.41	5.96	4.45	4.45	3.91
A x C	4.35	5.41	7.30	5.45	5.45	4.78
B x C	3.08	3.82	5.16	3.85	3.85	3.38
A x B x C	6.16	7.64	10.33	7.70	7.70	6.76

Table (2): Effect of humic acid (A) and salinity of irrigation water (C) on percentage of leaf content of nitrogen (N), phosphorus (P) and potassium (K) of apple seedlings (B): on *Malus communis* (Mc) and MM106 rootstocks

(A)	(C)	N (%)						P (%)						K (%)					
		2004 season			2005 season			2004 season			2005 season			2004 season			2005 season		
		MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)
Control	0	1.40	1.64	1.52	1.90	2.03	1.97	0.19	0.20	0.20	0.20	0.22	0.21	0.61	1.13	0.87	1.61	1.60	1.61
	1000	0.06	1.44	0.75	1.15	1.14	1.15	0.18	0.21	0.20	0.20	0.23	0.22	0.48	0.68	0.58	0.51	0.71	0.61
	2000	1.14	1.29	1.22	0.93	1.00	0.97	0.30	0.14	0.22	0.27	0.16	0.22	0.54	0.50	0.52	0.65	0.53	0.59
Ave. (A x B)		0.87	1.46	Ave (A) 1.16	1.33	1.39	Ave (A) 1.36	0.22	0.18	Ave (A) 0.20	0.22	0.20	Ave (A) 0.21	0.54	0.77	Ave (A) 0.66	0.92	0.95	Ave (A) 0.94
Soil	0	2.94	3.19	3.07	2.99	3.10	3.05	0.29	0.33	0.31	0.81	0.34	0.58	1.08	1.60	1.34	1.58	1.60	1.59
	1000	2.67	2.58	2.63	2.79	2.77	2.78	0.22	0.24	0.23	0.44	0.24	0.34	0.23	0.55	0.39	1.21	1.58	1.40
	2000	2.00	1.80	1.90	1.98	1.90	1.94	0.16	0.16	0.16	0.17	0.16	0.17	0.08	0.51	0.30	1.08	1.54	1.31
Ave. (A x B)		2.54	2.52	Ave (A) 2.53	2.59	2.59	Ave (A) 2.59	0.22	0.24	Ave (A) 0.23	0.47	0.25	Ave (A) 0.36	0.46	0.89	Ave (A) 0.68	1.29	1.57	Ave (A) 1.43
Foliar	0	1.23	1.69	1.46	2.33	2.79	2.56	0.21	0.25	0.23	0.22	0.24	0.23	0.58	0.78	0.68	1.31	1.48	1.40
	1000	1.23	2.07	1.65	1.41	2.17	1.79	0.15	0.16	0.16	0.16	0.17	0.17	0.50	0.42	0.46	1.20	1.42	1.31
	2000	0.92	0.87	0.90	1.11	0.77	0.94	0.16	0.14	0.15	0.17	0.15	0.16	0.49	0.47	0.48	0.52	1.48	1.00
Ave. (A x B)		1.13	1.54	Ave (A) 1.34	1.62	1.91	Ave (A) 1.76	0.17	0.18	Ave (A) 0.18	0.18	0.19	Ave (A) 0.19	0.52	0.56	Ave (A) 0.54	1.01	1.46	Ave (A) 1.24
Soil foliar	0	2.84	2.77	2.81	2.39	2.86	2.63	0.20	0.17	0.19	0.20	0.19	0.20	1.44	1.50	1.47	1.44	1.53	1.49
	1000	1.92	1.93	1.93	2.33	2.11	2.22	0.17	0.16	0.17	0.17	0.17	0.65	0.78	0.72	1.22	1.36	1.29	
	2000	1.23	1.58	1.41	1.33	1.39	1.36	0.24	0.26	0.25	0.23	0.25	0.24	0.53	0.37	0.45	1.20	1.37	1.29
Ave. (A x B)		2.00	2.09	Ave (A) 2.05	2.02	2.12	Ave (A) 2.07	0.20	0.20	Ave (A) 0.20	0.20	0.20	Ave (A) 0.20	0.87	0.88	Ave (A) 0.88	1.29	1.42	Ave (A) 1.35
Ave. (B x C)	0	2.10	2.32	Ave (C) 2.21	2.40	2.70	Ave (C) 2.55	0.22	0.24	Ave (C) 0.23	0.36	0.25	Ave (C) 0.31	0.93	1.25	Ave (C) 1.09	1.49	1.55	Ave (C) 1.52
	1000	1.47	2.01	1.74	1.92	2.05	1.99	0.18	0.19	0.19	0.24	0.20	0.22	0.47	0.61	0.54	1.04	1.27	1.16
	2000	1.32	1.39	1.36	1.34	1.27	1.31	0.22	0.18	0.20	0.21	0.18	0.20	0.41	0.46	0.44	0.86	1.23	1.05
Ave. (B)		1.63	1.91		1.89	2.00		0.21	0.20		0.27	0.21		0.60	0.77		1.13	1.35	

LSD at 5% for:

Humic acid (A)	0.25	0.27	0.002	0.002	0.088	0.138
Apple rootstock (B)	0.21	0.23	0.002	0.002	0.076	0.120
Salinity (C)	0.21	0.23	0.002	0.002	0.076	0.120
A x B	0.35	0.38	0.003	0.003	0.124	0.195
A x C	0.43	0.47	0.004	0.004	0.152	0.239
B x C	0.30	0.33	0.003	0.003	0.108	0.169
A x B x C	0.61	0.66	0.005	0.005	0.215	0.338

Table (3): Effect of humic acid (A) and salinity of irrigation water (C) on root length (>1.5, 1.5-0.5 and < 0.5 cm) of apple seedlings (B): on *Malus communis* (Mc) and MM106 rootstocks.

(A)	(C)	> 1.5 cm						1.5-0.5 cm						<0.5 cm					
		2004 season			2005 season			2004 season			2005 season			2004 season			2005 season		
		MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)
Control	0	8.80	15.80	12.30	0.01	8.20	4.11	16.1	16.8	16.5	13.3	17.3	15.3	10.6	12.4	11.5	10.0	8.2	9.1
	1000	5.00	9.30	7.15	0.01	10.70	5.36	13.7	15.1	14.4	13.4	15.5	14.5	12.1	9.5	10.8	9.3	9.7	9.5
	2000	3.20	15.30	9.25	0.01	7.30	3.66	10.1	11.5	10.8	11.2	15.2	13.2	8.5	10.2	9.4	9.7	13.2	11.5
Ave. (A x B)		5.67	13.47	Ave (A) 9.57	0.01	8.73	Ave (A) 4.37	13.3	14.5	Ave (A) 13.9	12.6	16.0	Ave (A) 14.3	10.4	10.7	Ave (A) 10.6	9.7	10.4	Ave (A) 10.0
Soil	0	12.30	19.00	15.65	0.01	0.01	0.01	36.6	32.1	34.4	34.7	33.6	34.2	17.1	18.5	17.8	21.5	26.5	24.0
	1000	7.20	19.30	13.25	0.01	0.01	0.01	13.0	21.6	17.3	31.8	32.6	32.2	12.5	15.6	14.1	19.9	23.8	21.9
	2000	15.50	18.70	17.10	0.01	19.70	9.86	13.2	14.3	13.8	30.1	30.7	30.4	9.8	10.9	10.4	17.6	22.5	20.1
Ave. (A x B)		11.67	19.00	Ave (A) 15.33	0.01	6.57	Ave (A) 3.30	20.9	22.7	Ave (A) 21.8	32.2	32.3	Ave (A) 32.3	13.1	15.0	Ave (A) 14.1	19.7	24.3	Ave (A) 22.0
Foliar	0	15.70	15.00	15.35	0.01	0.01	0.01	15.5	19.4	17.5	15.0	17.3	16.2	12.8	13.8	13.3	11.7	14.9	13.3
	1000	8.40	11.80	10.10	6.00	0.01	3.01	17.9	18.2	18.1	19.7	10.5	15.1	12.8	13.6	13.2	11.2	13.3	12.3
	2000	12.80	13.00	12.90	0.01	0.01	0.01	11.3	15.0	13.2	13.6	13.3	13.5	10.4	11.9	11.2	10.1	12.8	11.5
Ave. (A x B)		12.30	13.27	Ave (A) 12.78	2.01	0.01	Ave (A) 1.01	14.9	17.5	Ave (A) 16.2	16.1	13.7	Ave (A) 14.9	12.0	13.1	Ave (A) 12.6	11.0	13.7	Ave (A) 12.3
Soil foliar	0	7.60	19.50	13.55	0.01	0.01	0.01	33.2	36.3	34.8	32.6	35.7	34.2	12.5	15.7	14.1	22.8	22.0	22.4
	1000	14.70	12.70	13.70	0.01	0.01	0.01	23.2	33.1	28.2	30.4	36.0	33.2	10.0	11.6	10.8	19.0	20.8	19.9
	2000	19.30	14.30	16.80	0.01	0.01	0.01	22.2	29.2	25.7	28.0	30.6	29.3	9.0	11.8	10.4	9.6	12.4	11.0
Ave. (A x B)		13.87	15.50	Ave (A) 14.68	0.01	0.01	Ave (A) 0.01	26.2	32.9	Ave (A) 29.5	30.3	34.1	Ave (A) 32.2	10.5	13.0	Ave (A) 11.8	17.1	18.4	Ave (A) 17.8
Ave. (B x C)	0	11.10	17.33	Ave (C) 14.22	0.01	2.06	Ave (C) 1.04	25.4	26.2	Ave (C) 25.8	23.9	25.9	Ave (C) 24.9	13.3	15.1	Ave (C) 14.2	16.5	17.9	Ave (C) 17.2
	1000	8.83	13.27	11.05	1.51	2.68	2.10	17.0	22.0	19.5	23.8	23.7	23.8	11.9	12.6	12.3	14.9	16.9	15.9
	2000	12.70	15.33	14.02	0.01	6.76	3.39	14.2	17.5	15.9	20.7	22.5	21.6	9.4	11.2	10.3	11.8	15.2	13.5
Ave. (B)		10.88	15.31		0.51	3.83		18.9	21.9		22.8	24.0		11.5	13.0		14.4	16.7	

LSD at 5% for:

Humic acid (A)	0.071	0.002	1.87	2.65	1.94	2.77
Apple rootstock (B)	0.061	0.002	1.62	2.30	1.68	2.40
Salinity (C)	0.061	0.002	1.62	2.30	1.68	2.40
A x B	0.100	0.003	2.65	3.75	2.75	3.91
A x C	0.122	0.004	3.24	4.59	3.37	4.79
B x C	0.087	0.003	2.29	3.25	2.38	3.39
A x B x C	0.173	0.005	4.58	6.49	4.76	6.78

Table (4): Effect of humic acid (A) and salinity of irrigation water (C) on number of root (>1.5, 1.5-0.5 and < 0.5 cm) of apple seedlings (B): on *Malus communis* (Mc) and MM106 rootstocks.

(A)	(C)	> 1.5 cm						1.5-0.5 cm						<0.5 cm					
		2004 season			2005 season			2004 season			2005 season			2004 season			2005 season		
		MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)
Control	0	0.70	1.00	0.85	0.01	0.01	0.01	3.90	3.80	3.85	3.80	4.00	3.90	9.40	10.70	10.05	9.50	12.00	10.75
	1000	0.70	0.70	0.70	0.01	1.30	0.66	3.30	3.30	3.30	3.70	3.20	3.45	8.00	10.00	9.00	6.80	11.80	9.30
	2000	0.30	0.70	0.50	0.01	0.01	0.01	3.00	2.80	2.90	4.30	3.10	3.70	6.60	8.00	7.30	4.70	9.20	6.95
Ave. (A x B)		0.57	0.80	Ave (A) 0.68	0.01	0.44	Ave (A) 0.23	3.40	3.30	Ave (A) 3.35	3.93	3.43	Ave (A) 3.68	8.00	9.57	Ave (A) 8.78	7.00	11.00	Ave (A) 9.00
Soil	0	1.00	1.00	1.00	0.01	0.70	0.36	6.30	6.80	6.55	8.80	8.50	8.65	16.70	19.30	18.00	19.00	23.10	21.05
	1000	0.30	1.00	0.65	0.01	0.30	0.16	4.50	3.50	4.00	8.50	9.00	8.75	16.50	18.00	17.25	13.20	21.30	17.25
	2000	1.00	1.00	1.00	0.01	1.00	0.51	4.50	3.00	3.75	7.80	8.00	7.90	14.50	16.00	15.25	16.90	20.10	18.50
Ave. (A x B)		0.77	1.00	Ave (A) 0.88	0.01	0.67	Ave (A) 0.34	5.10	4.43	Ave (A) 4.77	8.37	8.50	Ave (A) 8.43	15.90	17.77	Ave (A) 16.83	16.37	21.50	Ave (A) 18.93
Foliar	0	1.00	0.70	0.85	0.01	1.30	0.66	4.70	4.50	4.60	6.30	6.50	6.40	14.00	16.50	15.25	17.50	22.50	20.00
	1000	0.30	1.00	0.65	0.30	0.30	0.30	4.30	2.30	3.30	8.20	4.20	6.20	13.70	13.00	13.35	12.80	14.00	13.40
	2000	1.00	0.70	0.85	0.01	0.30	0.16	3.50	3.00	3.25	2.70	3.00	2.85	12.80	10.00	11.40	14.30	17.80	16.05
Ave. (A x B)		0.77	0.80	Ave (A) 0.78	0.11	0.63	Ave (A) 0.37	4.17	3.27	Ave (A) 3.72	5.73	4.57	Ave (A) 5.15	13.50	13.17	Ave (A) 13.33	14.87	18.10	Ave (A) 16.48
Soil foliar	0	0.70	1.00	0.85	0.01	0.30	0.16	4.80	5.00	4.90	7.30	8.30	7.80	16.50	17.00	16.75	21.20	23.20	22.20
	1000	0.70	0.70	0.70	0.01	1.00	0.51	4.30	4.80	4.55	7.70	8.00	7.85	13.00	16.00	14.50	20.30	21.50	20.90
	2000	0.70	0.70	0.70	0.01	0.70	0.36	4.30	4.70	4.50	7.50	7.70	7.60	14.00	15.80	14.90	17.70	20.30	19.00
Ave. (A x B)		0.70	0.80	Ave (A) 0.75	0.01	0.67	Ave (A) 0.34	4.47	4.83	Ave (A) 4.65	7.50	8.00	Ave (A) 7.75	14.50	16.27	Ave (A) 15.38	19.73	21.67	Ave (A) 20.70
Ave. (B x C)	0	0.85	0.93	Ave (C) 0.89	0.01	0.58	Ave (C) 0.30	4.93	5.03	Ave (C) 4.98	6.55	6.83	Ave (C) 6.69	14.15	15.88	Ave (C) 15.02	16.80	20.20	Ave (C) 18.50
	1000	0.50	0.85	0.68	0.08	0.73	0.41	4.10	3.48	3.79	7.03	6.10	6.57	12.80	14.25	13.53	13.27	17.15	15.21
	2000	0.75	0.78	0.77	0.01	0.50	0.26	3.83	3.38	3.61	5.58	5.45	5.52	11.98	12.45	12.22	13.40	16.85	15.13
Ave. (B)		0.70	0.85		0.03	0.60		4.29	3.96		6.39	6.13		12.98	14.19		14.49	18.07	

LSD at 5% for:

Humic acid (A)	0.010	0.007	0.75	0.94	2.14	1.98
Apple rootstock (B)	0.006	0.006	0.65	0.81	1.86	1.71
Salinity (C)	0.008	0.006	0.65	0.81	1.86	1.71
A x B	0.010	0.010	1.06	1.32	3.03	2.80
A x C	0.017	0.012	1.30	1.62	3.71	3.43
B x C	0.012	0.008	0.92	1.15	2.63	2.42
A x B x C	0.023	0.017	1.84	2.29	5.25	4.84

Table (5): Effect of humic acid (A) and salinity of irrigation water (C) on percentage of dry matter in main and secondary roots of apple seedlings (B): on *Malus communis* (Mc) and MM106 rootstocks.

(A)	(C)	Main roots (%)						Secondary roots (%)					
		2004 season			2005 season			2004 season			2005 season		
		MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)
Control	0	42.5	50.0	46.3	48.1	51.3	49.7	29.8	30.4	30.1	33.0	37.0	35.0
	1000	45.5	46.1	45.8	45.2	46.9	46.1	31.5	28.2	29.9	26.3	34.2	30.3
	2000	39.9	40.6	40.3	40.3	41.6	41.0	27.5	28.2	27.9	23.2	29.6	26.4
Ave. (A x B)		42.6	45.6	Ave (A) 44.1	44.5	46.6	Ave (A) 45.6	29.6	28.9	Ave (A) 29.3	27.5	33.6	Ave (A) 30.6
Soil	0	55.4	58.3	56.9	60.5	62.0	61.3	46.1	49.4	47.8	43.0	47.4	45.2
	1000	41.3	47.6	44.5	54.4	56.8	55.6	43.4	45.6	44.5	39.9	45.1	42.5
	2000	46.4	45.9	46.2	52.5	53.3	52.9	40.9	41.8	41.4	40.4	37.1	38.8
Ave. (A x B)		47.7	50.6	Ave (A) 49.2	55.8	57.4	Ave (A) 56.6	43.5	45.6	Ave (A) 44.5	41.1	43.2	Ave (A) 42.2
Foliar	0	47.7	49.0	48.4	44.9	44.4	44.7	37.5	41.5	39.5	36.1	37.1	36.6
	1000	46.1	50.4	48.3	43.9	45.6	44.8	36.6	38.4	37.5	26.5	30.4	28.5
	2000	43.0	46.7	44.9	40.9	41.8	41.4	30.6	29.8	30.2	21.8	27.0	24.4
Ave. (A x B)		45.6	48.7	Ave (A) 47.2	43.2	43.9	Ave (A) 43.6	34.9	36.6	Ave (A) 35.7	28.1	31.5	Ave (A) 29.8
Soil + foliar	0	48.1	52.5	50.3	52.0	55.8	53.9	33.5	42.0	37.8	37.6	42.7	40.2
	1000	42.9	47.8	45.4	48.1	48.3	48.2	33.3	37.3	35.3	34.0	36.0	35.0
	2000	41.7	45.8	43.8	41.7	45.4	43.6	26.1	29.1	27.6	26.6	25.8	26.2
Ave. (A x B)		44.2	48.7	Ave (A) 46.5	47.3	49.8	Ave (A) 48.6	31.0	36.1	Ave (A) 33.6	32.7	34.8	Ave (A) 33.8
Ave. (B x C)	0	48.4	52.5	Ave (C) 50.5	51.4	53.4	Ave (C) 52.4	36.7	40.8	Ave (C) 38.8	37.4	41.1	Ave (C) 39.3
	1000	44.0	48.0	46.0	47.9	49.4	48.7	36.2	37.4	36.8	31.7	36.4	34.1
	2000	42.8	44.8	43.8	43.9	45.5	44.7	31.3	32.2	31.8	28.0	29.9	29.0
Ave. (B)		45.1	48.4		47.7	49.4		34.7	36.8		32.4	35.8	

LSD at 5 % for:

Humic acid (A)	3.35	4.27	2.33	2.03
Apple rootstock (B)	2.90	3.70	2.02	1.76
Salinity (C)	2.90	3.70	2.02	1.76
A x B	4.74	6.04	3.30	2.87
A x C	5.80	7.40	4.04	3.52
B x C	4.10	5.23	2.86	2.49
A x B x C	8.20	10.46	5.72	4.98

Table (6): Effect of humic acid (A) and salinity of irrigation water (C) on proline amino acid percentage of sodium (Na) and chloride (Cl) of apple seedlings (B): on *Malus communis* (Mc) and MM106 rootstocks.

(A)	(C)	Proline (mg/g)						Na (%)						Cl (%)					
		2004 season			2005 season			2004 season			2005 season			2004 season			2005 season		
		MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)	MM106	Mc	Ave (AxC)
Control	0	0.007	0.006	0.007	0.007	0.006	0.007	0.76	0.75	0.76	0.80	0.78	0.79	0.009	0.010	0.010	0.009	0.013	0.011
	1000	0.043	0.031	0.037	0.009	0.006	0.008	1.55	1.55	1.55	1.71	1.69	1.70	0.021	0.019	0.020	0.021	0.010	0.016
	2000	0.083	0.064	0.074	0.090	0.083	0.087	1.61	1.57	1.59	1.73	1.71	1.72	0.025	0.025	0.025	0.025	0.025	0.025
Ave. (A x B)		0.044	0.034	Ave (A) 0.039	0.035	0.032	Ave (A) 0.034	1.31	1.29	Ave (A) 1.30	1.41	1.39	Ave (A) 1.40	0.018	0.018	Ave (A) 0.018	0.018	0.016	Ave (A) 0.017
Soil	0	0.007	0.007	0.007	0.008	0.006	0.007	0.08	0.05	0.07	0.09	0.05	0.07	0.014	0.014	0.014	0.016	0.006	0.011
	1000	0.008	0.015	0.012	0.008	0.016	0.012	0.09	0.07	0.08	0.10	0.06	0.08	0.019	0.028	0.024	0.016	0.013	0.015
	2000	0.014	0.017	0.016	0.012	0.039	0.026	0.11	0.08	0.10	0.11	0.07	0.09	0.019	0.020	0.020	0.019	0.013	0.016
Ave. (A x B)		0.010	0.013	Ave (A) 0.011	0.009	0.020	Ave (A) 0.015	0.09	0.07	Ave (A) 0.08	0.10	0.06	Ave (A) 0.08	0.017	0.021	Ave (A) 0.019	0.017	0.011	Ave (A) 0.014
Foliar	0	0.001	0.004	0.003	0.001	0.003	0.002	0.54	0.52	0.53	0.68	0.64	0.66	0.010	0.006	0.008	0.011	0.011	0.011
	1000	0.039	0.041	0.040	0.026	0.022	0.024	0.56	0.53	0.55	0.69	0.66	0.68	0.018	0.010	0.014	0.020	0.013	0.017
	2000	0.075	0.069	0.072	0.064	0.061	0.063	0.58	0.06	0.32	0.71	0.68	0.70	0.026	0.022	0.024	0.026	0.022	0.024
Ave. (A x B)		0.038	0.038	Ave (A) 0.038	0.030	0.029	Ave (A) 0.030	0.56	0.54	Ave (A) 0.55	0.69	0.66	Ave (A) 0.68	0.018	0.013	Ave (A) 0.015	0.019	0.015	Ave (A) 0.017
Soil foliar	0	0.006	0.005	0.006	0.005	0.005	0.005	0.09	0.07	0.08	0.12	0.09	0.11	0.014	0.010	0.012	0.017	0.010	0.014
	1000	0.031	0.023	0.027	0.036	0.039	0.038	0.10	0.07	0.09	0.11	0.11	0.11	0.015	0.018	0.017	0.015	0.015	0.015
	2000	0.039	0.051	0.045	0.048	0.039	0.044	0.12	0.08	0.10	0.13	0.12	0.13	0.015	0.020	0.018	0.030	0.018	0.024
Ave. (A x B)		0.025	0.026	Ave (A) 0.026	0.030	0.028	Ave (A) 0.029	0.10	0.07	Ave (A) 0.09	0.12	0.10	Ave (A) 0.11	0.015	0.016	Ave (A) 0.015	0.021	0.014	Ave (A) 0.018
Ave. (B x C)	0	0.005	0.006	Ave (C) 0.006	0.005	0.005	Ave (C) 0.005	0.37	0.35	Ave (C) 0.36	0.42	0.39	Ave (C) 0.41	0.012	0.010	Ave (C) 0.011	0.013	0.010	Ave (C) 0.012
	1000	0.030	0.027	0.029	0.020	0.021	0.025	0.58	0.56	0.57	0.65	0.63	0.64	0.018	0.019	0.019	0.018	0.013	0.016
	2000	0.053	0.050	0.052	0.054	0.056	0.055	0.60	0.58	0.59	0.67	0.65	0.66	0.021	0.022	0.022	0.025	0.020	0.023
Ave. (B)		0.029	0.028		0.026	0.027		0.52	0.50		0.58	0.56		0.017	0.017		0.019	0.014	

LSD at 5% for:

Humic acid (A)	0.007	0.007	0.008	0.007	0.002	0.002
Apple rootstock (B)	0.006	0.006	0.007	0.006	0.002	0.002
Salinity (C)	0.006	0.006	0.007	0.006	0.002	0.002
A x B	0.010	0.010	0.011	0.010	0.003	0.003
A x C	0.013	0.013	0.013	0.012	0.004	0.004
B x C	0.009	0.009	0.009	0.008	0.003	0.003
A x B x C	0.018	0.018	0.019	0.017	0.006	0.006