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EFFECT OF ASCORBIC AND SALICYLIC ACID ON LEAF AREA, N,P,K CONTENT AS WELL AS YIELD AND ITS COMPONENTS OF MANGO (*Mangifera indica* L.) TREES

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ABSTRACT

A field study was carried out at the Horticulture Research Farm of El-Baramon, Dakahlia, governorate, Egypt aimed to study the effects of salicylic acic (SA) and ascorbic acid (AsA) on mango trees cv Fagri kalan. Leaf area and its mineral contents as growth parameter, yield and its were evaluated. Data indicated that spraying treatments increased productivity of mango trees and enhanced their fruit quality, in the two seasons Salicylic acid and AsA, when applied alone, produced low productivity of Fagri kalan mango compared to their combination. Spraying with 2MmSA+1Mm AsA gave the best values in this respect. Moreover, spraying with this treatment gave the highest values of TSS and total sugars in the fruits content and lowest value in acidity comparing with other interactions treatments used in both seasons. Further investigation are need to study the effects of AsA and SA on natural defiance system on some biotic and abiotic stress condition and to trace the involvement of metabolic processes in resistance to many disorder in mango.

Keywords: *Mangifera indica* L.; cv Fagri kalan; Salicylic acid(SA), Ascorbic acid(AsA), metabolic process; Fruit quality

INTRODUCTION

Mango (*Mangifera indica* L.) is one of the most important fruits grown in tropical and subtropical country. In Egypt, the cultivated area reached, 84204 feddan (FAO 2011) with many cultivars such as Ewais, Sediek, Zebda Succary, Keitt and others. More than 40% of this area exists in Ismailia governorate which the main cultivar planted is succary Abiad Mango yield worldwide are generally poor, ranging from four to nine t/ha in the major production countries (Oothuyse, 1993). Growth of mango is not continuous but it occurs as intermittent, short lasting flushes of shoots from apical or lateral buds. Vegetative growth occurs up to three or four times per year on individual stems, depending upon cultivars and growth conditions. Terminal or panicles are initiated in dormant apical buds on stems that developed vegetative from lateral buds following the previous flowering seasons (Litz, 1997).

Several compounds with different biochemical effects have been tried to improve plant growth and development on various plant species and mostly applied as foliar spray, from these compounds ascorbic acid (Dehghan, *et al*., 2011), SA (Hayat *et al*., 2010). The signal molecules salicylic acid (SA), jasmonic acid (JA) and methyl jasmonate (MeJA) are endogenous plant growth substances that play key roles in plant growth and development, and responses to environmental stresses. These signal molecules are involved in some signal transduction systems, which induce particular enzymes catalyzing biosynthetic reactions to form defense

compounds such as polyphenols, alkaloids or pathogenesis-related (PR) proteins (Van Loon, 1995) and enhancement of the level of chlorophyll and carotenoid pigments, photosynthetic rate and modifying the activity of some of the important enzymes are other roles assigned to SA (Hayat and Ahmad, 2007).

The metabolic aspects of plants, supplied with SA or its derivatives, shifted to a varied degree, depending on the plant type and the mode of application of SA. The application of SA (20 mg ml-1) to the foliage of the plants of Brassica napus improved the chlorophyll contents (Ghai et al., 2002). Because AA also serves as an important cofactor in the biosynthesis of many plant hormones, including ethylene (ET), JA, salicylic acid (SA), abscissic acid (ABA) and gibberellic acid (GA3); one has to assume that the endogenous level of AA will affect not only biosynthesis, but also the levels and the signalling of these hormones under stressful circumstances. (El-Mashad and Mohamed 2011).

On the other hand, it was found that salicylic acid (ortho-hydroxy. Benzoic acid) and its close analogues enhanced the leaf area and dry mass production in different plant species (Khan et al., 2003). It is distributed in the whole plant kingdom and found to participates in the regulation of stomatal closure, nutrient uptake, chlorophyll and protein synthesis, inhibition of ethylene biosynthesis, transpiration, and photosynthesis grapevine leaves (Wang et al., 2010). To induce growth and productivity of mango trees an investigated strategy shoule be considered for the different growth phases to minimizing the biannual bearing phenomenon (a high yield in a year is followed by a low yield in the subsequent year)noticed in mango trees.

So far however, little effort was focused on the enhancement of growth and yield of mango by SA and AsA treatments. The effects of these compound on mango is less understood. Therefore, the objective of this work was to evaluate the effects of AsA and SA acids on growth, NPK content as well as yield and its components of mango (*Mangifera indica* L.) cv. Fagri Kalan, growing in the spring growth cycle.

MATERIALS AND METHODS

The present investigation was carried out during the two growing seasons of 2014 and 2015 on mango (*Mangifera indica* L.) cv. Fagri Kalan trees grown at the Horticulture Research Farm of El-Baramon, Dakahlia, governorate, Egypt. Analysis of the soil surface and subsurface was done (Wilde *et al*., 1985) and the data are presented (Table 1). The texture of the soil is clay loam with a water table depth not less than 1.5 meters.

Table 1 . Analysis of the experimental soil.

Characters	Values of the soil samples		
	0-30 cm	30-60 cm	60-120 cm
Sand (coarse+fine)%	13.92	13.74	14.54
Silt %	20.37	20.13	19.47
Clay %	63.29	64.74	64.29
E.C.(1:2:5extract) dmS^{-1}	1.29	1.27	1.34
pH 1:2:5 suspension	7.92	7.83	7.79
Organic matter %	1.37	1.42	1.46
Available N%	0.19	0.17	0.16
" P %	17.35	17.26	17.12
" K %	573	567	558
" Zn ppm	0.25	0.22	1.22
" Fe ppm	1.96	1.7	1.65
" Mn ppm	0.5	0.4	0.4
$CaCo_3$	0.19	0.13	0.13
Field capacity %	15.3	15.7	15.8
Permanent wilting point%	7.4	7.6	7.7

Mango trees (Fagri Kalan cv.) 8 years old, similar vigor, height and planted at 5x5m were selected. Common horticultural practices as recommended by the Ministing of Agric, Egypt except application of any antioxidants treatments, were carried out as usual. Eight treatments from one antioxidant (Ascorbic acid) and three levels of salicylic acid (Sigma Company) as well as their combinations were used as spraying treatments and compared. Randomized block design system with 3 replicates per treatments, three trees per each replicate were used. The treatments were:

- Control (plants sprayed with distilled water).
- Spraying at ascorbic acid at 1mM.
- Spraying at salicylic acid with 1mM.
- Spraying at salicylic acid with 2 mM.
- Spraying at salicylic acid with 3 mM.
- Spraying at salicylic acid with 1mM + ascorbic acid at 1mM .
- Spraying at salicylic acid with 2mM + ascorbic acid at 1mM .
- Spraying at salicylic acid with 3mM + ascorbic acid at 1mM .

At the starting of spring growth cycle in each season, the application of all treatments was took place at three physiological stages. The 1st spraying was done before blooming (1st week of March) and the second just after fruit setting (1st week of May) and the 3rd was sprayed at one month later (1st week of Jun). Salicylic and was dissolved in ethyl alcohol whereas ascorbic acid in distilled water. Tween 20 as a wetting agent was added to all spraying solutions at 0.05% and the spraying was done till runoff.

Growth and physiological analysis

At beginning of fruit growth (2 weeks from the last spraying date), sixteen new shoots were chosen. Four labeled branches (four shoots for each direction) were taken for growth and physiological parameters. Leaf area as growth parameter was estimated using the equation given by Ahmed and Morsy (1999) as follows:

Leaf area (cm^2) = 0.70 (leaf length x leaf width) – 1.06

Fifty mature leaves from non-fruiting shoots were taken for the determination of nitrogen, phosphorus and potassium concentrations according to the methods described by Bremner and Mulvaney (1982), A.O.A.C (1995) and Chapman and Pratt (1982) respectively.

Harvesting from each treated mango tree was took place periodically, three times, 20 days intervals, from 1st August, till of 10th September. At each harvesting time, fruit yield was measured during the two growing season. Yield, expressed in weight (kg) per tree was calculated. At the third harvesting time (Sept. 10th) ten fruits from each tree were taken for determination physical and chemical characters: These include, fruit weight (g) and thickness (cm.); percentage of pulp; edible (pulp weight, g) to non edible portions (peel and seed weights, g); percentage of total soluble solids; percentage of total acidity(as g citric acid/ 100 ml juice) according to (A.O.A.C., 1985); percentages of total sugars were determined by (Amberger, 1954), and ascorbic acid content in the juice (as mg/ 100 ml juice) by titration against 2-6 dichloro phenol endophenol (A.O.A.C. 1985**)**.

Statistical analysis:

All data were subjected to analysis of variance (Snedecor and Cochran 1980) using SAS system (2003).

RESULTS AND DISCUSSION

a) Leaf area and its mineral contents

Table (2) indicated that, all the treatments of salicylic acid (SA) and ascorbic acid (AsA) significantly increased leaf area as well as N, P and K percentages compared with the control in both seasons. The highest values were obtained with sprayed 2mM SA+1mM ascorbic acid followed by 1mM SA+1mM ascorbic comparing with other treatments used in the two growing seasons. The lowest values were recorded in the control treatment in both seasons. Therefore, using salicylic acid and ascorbic acid in a descending order was significantly preferable in improving leaf area as well as N, P and K content. The interaction treatments showed highest values in this respect when used spraying with the two antioxidants at 1 mM. Similar results were reported by (Badran and Ahmed 2009) on mango with sprayed antioxidants at 500 to 2000 ppm. The controlling effect of antioxidants on different disorders may be another reason (Orth *et al.,.* 1993). In *Arabidopsis*, SA may aid in the induction of antioxidant defenses and maintenance of the glutathione pool (Sharma *et al*., 1996), both possible indications of its involvement in plant protection against oxidative stress (Rao and Davis, 1999). Larqué and Martin (2007) suggested that SA could play an important role in the bioproductivity of plants and that could be linked to the observed effect of promoting the root length of plants. Salicylic acid is synthesized from chorismate by means of isochorismate synthase in chloroplasts and the salicylic acid synthesized by this pathway is responsible for providing local and systemic acquired resistance in plants (Wildermuth et al., 2001). This protection from oxidative damage was mediated by an increased activity of various antioxidant enzymes. (Zhou et al., 2009) The promotive effect of these antioxidants on nutritional status of the trees was supported by the

results of Ahmed (2001) on Taimour mango trees; Abo- ElKomsan *et al.,* (2003) on Balady orange trees; Hamad (2004) on some mango cvs, Gamal (2006) on Washington Navel orange trees and Ali – Ragaa (2008) on Balady mandarin trees.

Table (2). Effect of SA and ascorbic acid on leaf area cm^2/ branche and N, P, K percentages (%) of Fagri Klan mango trees during the two growing season 2014 and 2015.

Treatments	Leaf area (cm2)		N %		P %		K %	
	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015
control	59.3	59.7	1.06	1.09	0.23	0.25	0.58	0.57
1Mm ascorbic acid	63.4.	63.5	1.12	1.17	0.26	0.29	0.63	0.65
1Mm SA	65.6	64.8	1.17	1.12	0.28	0.31	0.66	0.67
2Mm SA	67.8	68.2	1.26	1.29	0.31	0.34	0.71	0.73
3Mm SA	66.7	66.9	1.19	1.12	0.29	0.32	0.68	0.67
1MmSA+1Mm ascorbic	68.4	96.5	1.25	1.28	0.32	0.38	0.73	0.75
2MmSA+1Mm ascorbic	71.5	72.7	1.32	1.35	0.34	0.41	0.78	0.79
3MmSA+1Mm ascorbic	66.2	67.8	1.26	1.29	0.31	0.35	0.71	0.72
LSD at 5%	1.04	1.25	0.21	0.11	0.02	0.02	0.03	0.03

b) Yield and its components

It is clear from the data in Table (3) that a remarkable and significant strengthen on physical and chemical quality of the fruits in the two growing seasons due to SA and Ascorbic acid treatments. Fruit weight was the highest within all treatments with significantly increments than control**.** Salicylic acid treatments enhanced fruit set and total fruit number per tree compared with the control. Averaged across the application methods, the highest fruit set observed with the application of salicylic acid plus ascorbic acid compared with the other treatments specific control trees. The most effective treatment on total fruit numbers at harvesting was 2 mM SA+1mM ascorbic.

Using salicylic acid has significant promotion on fruit quality of Fagri kalan mango trees. The best results with regard to fruit quality from the economical point of view were obtained when the trees received SA *via* leaves at 2mM. Undesirable effects on fruit quality were recorded on trees did not receive these antioxidants. The same direction was observed on different characters of the Fruit; fruit weight, yield/tree kg and thickness, pulp %, edible to non- edible portions during the two growing seasons. In horticultural species, the effect reported is the increase of yield without affecting the quality of the fruits. It is proposed that the increase in bioproductivity is mainly due to the positive effect of SA on root length and its density. (Shaaban and Mahmoud 2012, Larqué and Martin 2007). SA is one of the growth regulators which is now in demand by the producers to enhance the papaya production by about 20% (Herrera-Tuz, 2004; Martin-Mex et al., 2005). Moreover, in other plants, grown in green house and in open conditions, the lower

concentration (10^{-6} M) of SA increased the fruit yield from 90 to 120 ton/ha in commercial plantations without affecting the quality of fruits.

Table (3) : Effect of SA and ascorbic acid on yield / tree (kg), fruit weight (g), fruit thickness (cm), pulp % and edible to non edible portions of Fagr Klan mango trees during the two growing season 2014 and 2015.

Treatments	Yield/tree kg		Fruit weight (g)		Fruit thickness (cm)		Pulp %		Edible to non edible portions	
	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015
Contro	57.4	59.2	247.6	249.7	5.3	5.5	68.4	69.1	1.8	1.9
1Mmascorbic acid	68.5	71.3	257.5	261.3	6.5	6.7	69.6	69.8	1.9	2.1
1Mm SA	78.7	79.8	268.4	272.5	6.8	6.9	71.6	71.7	2.2	2.3
2Mm SA	94.2	98.5	294.7	298.4	7.1	7.4	72.4	72.7	2.5	2.5
3Mm SA	78.9	81.2	279.4	283.6	6.9	7.1	71.2	71.5	2.4	2.3
1MmSA+1Mm ascorbic	102.6	105.6	321.6	334.2	7.4	7.7	73.3	73.6	2.7	2.8
2MmSA+1Mm ascorbic	121.4	128.3	377.8	381.2	7.9	8.2	74.5	74.7	2.9	3.2
3MmSA+1Mm ascorbic	98.6	99.3	311.2	313.4	7.1	7.3	72.1	72.3	2.6	2.8
LSD at 5%	1.52	1.49	7.6	7.9	0.31	0.27	0.99	1.11	0.06	0.047

c) Quality of mango fruit (physical and chemical characteristics)

Fruit TSS content increased significantly with salicylic acid than ascorbic acid treatments during the two seasons (Table 3). Moreover, spraying with 2mM SA+ 1mM ascorbic gave the highest significant increase in fruit TSS comparing with other treatments in both seasons. Lower acidity content comparing with control fruits during the study in the two seasons and the lowest values were recorded with the treatment 2mM SA+ 1 mM ascorbic but the values of total sugar increase as a result of spraying with all treatments more than control. Data presented in the same table reveal that in both seasons the highest sugars values were obtained in the fruits harvested from trees sprayed with 2mM SA+ 1 mM ascorbic followed by 1mM SA+ 1 mM ascorbic. Fruit total soluble solids , total sugars and ascorbic acid content increased significantly with SA treatments than free during the two seasons (Table 3). Moreover, with spraying 1Mm ascorbic acid supplementation gave the highest significant increasing in fruit all characters comparing with other treatments in both seasons. The interaction between SA and ascorbic acid gave highest values in this respect. Fruit retention yield/ tree, fruit weight and thickness, edible to non- edible portions, total soluble solids total and reducing sugars and ascorbic acid content and decreased total acidity compared to the control treatment. Similar results were reported by Badran and Ahmed (2009). Qaiser Hayat *et al.,* (2010) reported that the effect of exogenous application of SA on bio-productivity, growth, photosynthesis, plant water relations, various enzyme activities and its effect on the plants exposed to various biotic and abiotic stresses has also been discussed.

Jiankang -Cao *et al.,* (2006) suggested that, the SA spray applied to the trees around 30 days after full flowering notably enhanced accumulation of hydrogen peroxide in the young fruit. Meanwhile, activities of defense enzymes, including peroxidase, phenylalanine ammonia-lyase (PAL), chitinase or b-1,3-glucanase in the young fruit from SA-treated trees. The promoting effect of these antioxidant compounds on yield was confirmed by the results of Ahmed (2001) and Hammam *et al.,* (2001) on Hindy Bissinara mango trees, Ahmed (2001) on Taimour mango trees and Gamal (2006) on Washington Navel orange trees. Indeed, it has been demonstrated that application of SA on mango fruit before harvest (Zainuri *et al.,* 2001), or acibenzolar-S-methyl, a functional analogue of SA, on melon plant prior to flowering (Huang *et al.,* 2000) could effectively reduce postharvest diseases in fruit. Jiankang Cao *et al.,* (2006) found that treatment of SA sprays on the trees may provide further protection against postharvest disease of Ya Li pear fruit in practice and could be used as an alternative and economical approach to reduce application of chemical fungicides. Fruit firmness, total soluble solids and titratable acidity content were not obviously affected by organic acid (OA) and SA treatments. It was suggested that the effect of OA or SA on mango CI probably attributed to more reducing status of ascorbate and glutathione (Zhou *et al.,* 2009). SA can decrease the degradation rate of starch to soluble sugar in pulp of fruit, enhance the firmness and decrease fruits decay index, and in turn SA improves preservation of fruits. It's suggested that SA could evidently enhance antioxidation capacity and preservation effect of harvested banana fruits by increasing the ability of eliminating reactive oxygen species in fruit (Larqué-Saavedra and R. Martin-Mex 2007).

Table (4): Effect of SA and ascorbic acid on some physical and chemical aspects, (total soluble solid, total acidity, total sugars percentage and ascorbic acid content) of the fruits of Fagri Kalan mango trees during 2014 and 2015 seasons.

Treatments	TSS %		Total acidity %		Total sugars %		Ascorbic acid content mg/100mljuice	
	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015	1 st 2014	2 nd 2015
Contro	11.5	11.6	1.62	1.64	7.92	8.12	29.6	28.7
1Mm ascorbic acid	11.8	11.8	1.57	1.56	8.17	8.32	29.9	28.9
1Mm SA	12.2	12.4	1.46	1.44	8.23	8.43	28.4	27.8
2Mm SA	12.7	12.6	1.37	1.36	8.57	8.74	28.7	28.4
3Mm SA	12.3	12.5	1.42	1.4	8.21	8.42	27.6	28.5
1MmSA+1Mm ascorbic	12.9	13.1	1.31	1.31	8.93	9.22	31.6	32.5
2MmSA+1Mm ascorbic	13.2	13.3	1.26	1.24	9.46	9.54	33.2	34.3
3MmSA+1Mm ascorbic	12.8	12.9	1.29	1.31	8.78	8.86	30.3	31.4
LSD at 5%	0.11	0.12	0.02	0.01	0.71	0.8	0.17	0.15

CONCLUSION

In conclusion, results from this study suggest that AsA and SA are a potent stimulator for induction mango growth as well as fruit yield and improving its components. The treatments affected on certain physical and chemical attributes of mango fruits. Further studies are needed to evaluate the effects of AsA and SA on natural defiance system against stress condition and to trace the involvement of metabolic processes in resistance to this disorder in mango.

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تأثير حمض الساليسيليك و الاسكوربيك أو أيهما على المساحة الورقية، ومحتوى الأوراق من عناصر النيتروجين والفسفور و البوتاسيوم و كذلك على المحصول و مكوناته لأشجار المانجو صنف فجركلان

حنان أحمد محمد رشاد الحسينى
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أجريت التجربة بالمزرعة البحثية لمعهد بحوث البساتين بالبرامون - مركز البحوث الزراعية محافظة الدقهلية، مصر. لدراسة تأثيرالرش بحمض السالسليك (SA) وحمض الاسكوربيك (AsA) أو أيهما على أشجار المانجو صنف فجركلان على مساحة الورقة ومحتوياتها من العناصر المعدنية الكبرى، و المحصول و صفات جودة ثماره. و أوضحت النتائج أن جميع المعاملات ادت الى رفع إنتاجية أشجار المانجو وتحسين نوعية ثمارها مقارنة بمعاملة الكنترول و ذلك خلال موسمى النمو. و لقد كان استخدام مزيج من حمضى السالسليك والاسكوربيك افضل بكثيرمن استخدام أى منهما على حده. و أعطت المعاملة ١مللى مول حمض اسكوربيك + ٢مللى مول حمض سالسليك أفضل القيم من حيث الإنتاجية ونوعية الثمار في الموسمين. كما سبقت هذه المعاملة إرتفاع قيمة المواد الصلبة الكلية بالثمرة و محتواها السكرى مع إنخفاض درجة حموضتها مقارنة بالمعاملات الأخرى المستخدمة في كلا الموسمين. و يمكن التوصية باستخدام خليط من حمض السالسليك و الاسكوربيك معا كمنشطات حفزية قوية للنمو و المحصول فى المانجو صنف فجركلان مع تحسين مكونات الثمرة الفيزيائية والكيميائية. على ان يجرى مزيد من الدراسة لتقييم اثار إستخدام المعاملات على نظام المقاومة الطبيعية لأشجار المانجو لظروف الإجهاد المسببة تدهور الاشجار تحت مثل هذه الظروف