



## *Duranta repens* salinity tolerance and foliar application with proline and ascorbic acid

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### Abstract

*Duranta repens* L. is an evergreen shrub used for landscape gardens and hedges in some world regions. Due to the deficit of non-saline irrigation water, and fewer ornamental plants that encounter these obstacles; impair landscaping and gardening of these areas are expected. Therefore, Pot experiments were conducted during 2017 and 2018 seasons to investigate the impact of saline water which assigned as a main plot and the combinations of proline (Pr)+ ascorbic acid (AA) were assigned as a sub plot. The obtained results indicated that plants survival percentage was 100% till 2 g/l NaCl, while at 3-5 g/l NaCl the survival percentage was nil. The treatment of 1 g/l NaCl; caused a significant increases of studied plant vegetative growth characters in the first season compare to control treatment and 2 g/l NaCl. While higher significant values of such traits were recorded in the second season. However, control treatment showed some higher values for some other attributes during both seasons. In the second season, highly significant content of proline and leaf green color degree, Na and Cl (%) were resulted from the plants treated with 2 g/l NaCl. In terms of foliar application treatments, the gained results showed that 2 g/l Pr +0.3 g/l AA and control treatment; resulted in higher or lower significant values of vegetative growth traits and leaf green color degree and water content, respectively during both seasons. Higher significant proline, Na and Cl (%) contents were achieved by control treatment and due to apply the combined treatment of 1.0 g/l Pr + 0.2 g/l AA. Pertaining the interaction effect between the tested dependent variables, the presented results exhibited that the best results of vegetative growth traits and water content were obtained owing to irrigation the plants with 1 g/l NaCl in the first season and control treatment in the second one in combination with 2 g/l Pr + 0.3 g/l AA. While, higher significant percentages of carbohydrates, N, P and K; resulted from irrigation the plants with control treatment and spraying with 1 g/l Pr + 0.2 g/l AA; meanwhile, irrigation the plants with 2 g/l NaCl without foliar spraying; gave rise to higher significant proline content and Na and Cl (%). On the other side, 2 g/l NaCl in combination with 2 g/l Pr + 0.3 g/l AA treatment; achieved higher significant value of leaf green color degree during both seasons.

**Keywords:** *Duranta repens*, saline irrigation, proline, ascorbic acid, NaCl

### 1. Introduction

Ornamental plants have profound positive impacts on the global environment *via* several means such as: reducing the CO<sub>2</sub> concentration and heat accumulation as well as scavenge the air pollutants (whether dust or chemicals). On the other extreme, ornamental plants are the most important using objects in the tourist cities for many purposes such as opening gardens, golf spaces, interior designs, etc. Among them known *Duranta erecta* or *Duranta ellisia* Jacq, Golden dewdrop (Fam. Verbenaceae) which is, also, known takes the form of a scrambling shrub that sometimes takes the scrambling shrub or rarely a small tree. It is producing, commercially, from seeds, it is popular ornamental used for accent plants and hedges in tropical and subtropical parts of the world because of its profuse displays of flowers and fruits cuttings or by layering, its flowering begins in late summer and continue through autumn <sup>[1]</sup>. *Duranta erecta* plants survive at least 15 years, it has a moderate growth rate, usually 0.5 m/year for the first few years. It is sometimes grown in greenhouses in too cold areas for natural plants.

Most of the Egyptian lands are located in the arid region, where irrigation water is limited and usage of saline water is the probable alternative, which is common in most of the

new reclaimed lands <sup>[2]</sup>. The tolerance of various ornamental plants to drought and saline stresses is very important for landscaping and gardening in these areas. Physiologically, under saline and water stresses plants closed their stomata in order to protect the plants from dehydration. However, closing of stomata, also, restricts the exchange of carbon dioxide and oxygen between outside atmospheric air and its internal tissues. This condition decreases the uptake of nutrients through plant and slows down metabolism in plant and decreases the chances of plant survival <sup>[3]</sup>.

In order to overcome this constrain, several attempts have been investigated as the usage of vitamins to encounter the harmful effects of salinity stress on growth and productivity of many plant species is highly recommended <sup>[4]</sup>. For instance, ascorbic acid has been applied to improve of plant growth under these conditions due to its antioxidant activity and protecting plants from damage due to abiotic stress <sup>[5]</sup>. The amino acid proline is the most studied compound responding to salinity stress and osmotic status and usually considered as an osmoprotectant agent being accumulated and involved in reducing the oxidative damage by scavenging and/or reducing the free radicals <sup>[6, 7, 8]</sup>. Hence, exogenous application of proline is known to induce abiotic stress tolerance in plants <sup>[9]</sup>.

The present study was conducted to evaluate the growth and chemical composition of *Duranta repens* under different levels of NaCl, and its salt stress resistance with foliar application of both ascorbic and proline acids.

## 2. Materials and methods

Pots experiment was carried out at the Experimental Farm of the Faculty of Agric. Saba Basha, Alex. Univ. during both seasons of 2017 and 2018 to study the effects of different NaCl concentrations and various combinations of

proline plus ascorbic acid, as well as their interactions on vegetative growth traits and chemical constituents of *Duranta repens* L. shrub. *Duranta* transplants of six months old with 28±2cm in height were obtained from commercial nursery in Alex. Governorate. They were transplanted in plastic pots 30 cm in diameter on 1<sup>st</sup> march 2017 and 2018 seasons (one transplant/pot). The pots were filled with 7.5 kg of dried soil and the physical and chemical parameters of the used soil were determined according to [10] and shown in Table (1) [the same soil for both seasons].

**Table 1:** Physical and chemical analysis of the used soil

Particle size distribution (%)				Textural class	EC (dSm <sup>-1</sup> )	pH	CaCO <sub>3</sub> (%)	O.M (%)			SP (%)
Coarse sand	Fine sand	Silt	Clay	Sand clay							
3.19	71.53	10.21	15.07		1.28	7.75	2.91	0.92			45.3
Anions (meq/100g soil)								Available (mg/kg)			
Ca <sup>++</sup>	Mg <sup>++</sup>	Na <sup>+</sup>	K <sup>+</sup>	CO <sub>3</sub> <sup>--</sup>	HCO <sub>3</sub> <sup>-</sup>	Cl <sup>-</sup>	SO <sub>4</sub> <sup>-</sup>	N	P	K	
1.29	0.84	4.12	0.30	-	1.42	3.66	1.47	52.6	5.65	95.8	

The experiment was designed as a split plot design with 3 replicates, each one included 30 treatments which were combinations between 6 levels of saline water (control; tap water with 365 mg/l salts), 1, 2, 3, 4 and 5 g/l NaCl) as arranged in main plots and 5 treatments of proline [Pr] and ascorbic acids [AA] (control, 1.0 g/l proline+0.2 g/l ascorbic acid, 1g/l proline + 0.3 g/l ascorbic acid, 2 g/l proline + 0.2 g/l ascorbic acid and 2 g/l proline+ 0.3 g/l ascorbic acid) were assigned as subplots. Each treatment contained 3 plants, 90 plants for each season. The plants were irrigated with tap water for one month, then were irrigated with saline water from 1<sup>st</sup> April to 1<sup>st</sup> Nov. each season and the following data were recorded: 1) Survival %, 2) vegetative growth traits as plant height (cm), main stem diameter in cm (at 5 cm from soil surface), plant diameter [cm] (small diameter + large diameter/2), branches number/plant, leaf area (cm<sup>2</sup>) according to [11], fresh and dry weights of shoots and roots/plant (g) and shoot/root ratio, 3) chemical composition: leaf green color degree (SPAD units) using chlorophyll meter [12], water content% = (FW-DW/FW)X100 [13], N, P, K, Na, Cl and total carbohydrates% and proline (mg/g DW) contents were measured in the dried leaves as follows: N% by modified Micro-kjeldahl [14], K and Na% was estimated using Flame photometrically, using Jenway Flame photometer, Model corning 400 according to the modified method of [15], Cl (%) was determined photometrically [16], total carbohydrates (%) using method of [17] and proline content according to [18]. All data were statically analyzed according to the technique of analysis variance (ANOVA) and the Duncan's Multiple Rang Test method at 5% level was used to compare the difference between the means of treatment values to the methods described by [19].

## 3. Results and discussion

### 3.1 The effect of salinity levels, proline plus ascorbic acids concentrations and their interaction on survival% and vegetative growth traits of *Duranta repens*

After 8 months of irrigation with saline water and 7 times spraying by proline plus ascorbic acid at the tested concentrations, *Duranta repens* plants tolerated salinity up to 2 g/l NaCl, the survival %percent was 100% until such level. While, irrigated plants with 3, 4 and 5 g/l NaCl survival was nil (0.0%) at harvesting time on 1<sup>st</sup> Nov. during both seasons. Plant survival percentage was affected adversely as a result of salinity, also the salinity tolerance

depends on the interaction between salinity and other environmental factors [20]. Likewise, salinity affect all the major physical and biochemical processes that regulate growth such as photosynthesis, proline synthesis and lipid metabolism [21]. Similarly [22], found that buttonwood plants could be irrigated with water should not exceed 3500 ppm salt concentration to make the best vegetative and chemical characters.

Concerning the main effect of salinity on vegetative growth traits, results postulated in Table (2) indicated that the highest significant values of plant height, plant diameter, stem diameter branches number/plant, plant leaf area and fresh and dry weights of shoots and roots; resulted from applying 1.0 g/l NaCl to irrigation water in comparison to control treatment and due to applying 2.0 g/l NaCl in the first season only. While in the second season, the significantly ( $p \leq 0.01$ ) highest values of such traits were derived from the control treatment compare with either 1.0 or 2.0 g/l NaCl treatments. Higher significant ( $p \leq 0.01$ ) value of shoot/root ratio was achieved from control plants during both seasons. On the other side, the least significant values of vegetative growth traits mentioned earlier were achieved owing to apply 2.0 g/l NaCl during both seasons. This result may be taken place due to excessive Na<sup>+</sup> and Cl<sup>-</sup> concentrations which affect the absorption of many essential nutrients such as K, Ca, Mg and N [22]. This occurs through competitive interactions which affect the ionic selectivity of cell membranes) [23] and photosynthetic activity [24], reducing stomata opening and leading to decrease in intracellular CO<sub>2</sub> [21]. Salinity causes a reduction in photosynthetic rate *via* raising antioxidant enzymes and proline contents as stress responses to deal with increased levels of reactive oxygen species, leading to restrict most plants from differentiate or discriminate between K<sup>+</sup> and Na<sup>+</sup> ions, and in saline conditions uptake and accumulate high levels of Na<sup>+</sup> ions to the detriment of the necessary quantities of K<sup>+</sup> leading to loss of K<sup>+</sup> function dependent reactions and leading N<sup>+</sup> ions to induce toxicity [25]. The low level of NaCl (1g/l) caused a significant increase in the growth traits in the first season. This finding may refer to the interaction mode of action between salinity and other environmental factors [20]. This result was supported by [26] who showed that lower seawater ratios in *Moringa oleifera* irrigation water; gave rise to the best results for growth characters. Likewise, [27] illustrated that increasing salinity (6.25 ds.m<sup>-1</sup>) significantly reduced growth traits of *Jatropha curcas*.

**Table 2:** Average values of some vegetative growth traits of *Duranta repens* as affected by NaCl levels during both 2017 and 2018 seasons.

Traits	NaCl treatments (g/l)					
	Control		1.0 g/l		2.0 g/l	
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
Plant height (cm)	66.99B	87.10A	81.80C	68.27B	56.25A	58.95C
Plant diameter (cm)	46.03B	55.99A	52.67A	36.02B	37.06C	22.59C
Steam diameter (cm)	0.50B	0.65A	0.67A	0.45B	0.40C	0.40C
Branches number/plant	7.60B	9.93A	11.47A	6.80B	5.67C	5.73C
Plant leaf area (cm <sup>2</sup> )	62.93B	62.09A	71.83A	62.44A	61.44C	59.22B
Shoot fresh weight g/plant	44.68B	61.19A	79.19A	36.76B	27.47C	29.38C
Shoot dry weight g/plant	15.64B	20.78A	27.87A	11.97B	9.15C	10.42B
Roots fresh weight g/plant	13.86B	19.89A	27.70A	15.26B	9.42C	11.67C
Root dry weight g/plant	5.63B	6.75A	11.13A	5.42B	3.76C	4.22C
Shoot/root ratio	3.33A	2.94A	2.83B	2.88B	2.96C	2.81C

Means for each season followed by the same letter (s) in every consecutive horizontal boxes are not significantly different at 0.05 level according to Duncan's Multiple Rang Test.

In terms of the effect of foliar application by different combinations of proline and ascorbic acid, results in Table (3) exhibited that higher significant ( $p \leq 0.01$ ) values of plant height, plant diameter, stem diameter, branches number/plant, plant leaf area and fresh and dry weights of shoot and roots were recorded because of spraying plants with 2.0 g/l proline + 0.3 g/l ascorbic acid, while lower significant values of such traits were recorded owing to not spraying plants which gave rise to higher significant ( $p \leq 0.01$ ) shoot/root ratio during both seasons. But lower significant shoot/root ratio was recorded for the treatments consisted of 2.0 g/l proline+ 0.2 g/l ascorbic acid, and 2.0 g/l proline  $\leq$  + 0.3 g/l ascorbic acid during both seasons, respectively. The difference between the tested treatments reached the significant level in the most cases within both seasons. The promotive effect of 2.0 g/l proline +0.3 g/l ascorbic acid on vegetative growth parameters may be attributed to that ascorbic acid mode of action where plays an essential role in plant growth and development, have been implicated in many physiological responses [28]. As well as, it is a well-known that antioxidant and cellular reluctant with an intimate and complex role in the response of plants to O<sub>3</sub> [29]. Also, it acts as a primary substrate in the cyclic pathway of enzymatic detoxification of hydrogen peroxide. Improvement of ascorbate in plants will lead to the possibility of increasing ascorbate concentration in

plants by genetic manipulation. This will have benefits for tolerance of plants [30]. In addition, proline is a proteinogenic amino acid, highly essential for various vital metabolic processes within the plant tissues [31]. Also, [32] reported that proline at its optimal level has several functional roles such as osmoprotectant, turgor generation, storage of carbon and nitrogen, maintenance of the structure of proteins, maintenance of cytosolic pH, balance of redox status, acting as a part of stress signal, an inhibitor of lipid membrane peroxidation and antioxidant as effective quencher of ROS formed under stress conditions in plants. As well as, the effectiveness of applied proline depends on the type of plant species, development stage, rate of application and its concentration [33]. All these effects of ascorbic acid and proline reflected on vegetative growth traits. The exogenous application of proline at different concentrations had better effects on the growth characters in numerous studies as [24] on *Concarpus erectus* [35], on *Citrus sinensis* and [31] on *Azabidopsis thaliana*. In addition, [36] reveled that ascorbic acid at 200 ppm improved plant height, stem diameter, leaves number and dry weight of leaves, stem and root of *Khaya senegalinsis*. In addition, [37] concluded that ascorbic acid at 400 mg/l on three flax cultivars (Sakha 3, Giza 3 and Asiane) had better effects on the growth traits. This was supported by [38] on *Brassica rapa* mentioned that 100 mM ascorbic acid was more effective on vegetative growth characters.

**Table 3:** Average values of some vegetative growth traits of *Duranta repens* as affected by proline + ascorbic acid during both 2017 and 2018 seasons.

Traits	Proline (Pr)+ascorbic acid (AA) (g/l) treatments									
	0		1.0 g Pr +0.2 g AA		1.0 g Pr +0.3 g AA		2.0 g Pr +0.2 g AA		2 g P +0.3 g AA	
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
Plant height (cm)	61.34E	64.71E	65.03D	69.60D	68.14C	71.64C	72.09B	74.47B	61.34A	76.78A
Plant diameter (cm)	40.78E	32.85E	42.44D	35.89D	43.99C	38.58C	48.05B	40.88B	51.01A	42.78A
Steam diameter (cm)	0.44B	0.44D	0.48B	0.48C	0.54A	0.50C	0.55A	0.53B	0.58A	0.56A
Branches number/plant	6.56E	6.44D	7.44D	7.11C	8.22C	7.56BC	9.00B	8.00AB	10.00A	8.33A
Plant leaf area (cm <sup>2</sup> )	62.07E	58.27E	63.75D	58.27D	64.83C	58.27C	67.54B	58.27B	68.79A	58.27A
Shoot fresh weight g/plant	37.80E	38.03E	45.10D	40.05D	50.18C	42.40C	55.75B	44.79B	63.41A	46.95A
Shoot dry weight g/plant	13.17E	12.90B	15.54E	13.82AB	17.38C	14.51AB	19.51B	15.60A	22.16A	15.13A
Roots fresh weight g/plant	12.29E	12.40E	14.74D	14.16D	16.61C	15.82C	19.95B	17.34B	21.37A	18.32A
Root dry weight g/plant	4.79E	4.36D	5.85D	4.87C	6.68C	5.50B	8.08B	6.17A	8.80A	6.40A
Shoot/root ratio	3.20A	2.91A	3.18B	2.89B	2.98D	2.89B	2.82E	2.86C	3.02C	2.84D

Means for each season followed by the same letter (s) in every consecutive horizontal boxes are not significantly different at 0.05 level according to Duncan's Multiple Rang Test.

As for the interaction effect between NaCl levels and different treatments of proline + ascorbic acid, results in Table (4) showed that the different combinations between

NaCl and proline + ascorbic acid had remarkable significant effect on vegetative growth traits (plant height and diameter, stem diameter, branches number, plant leaf area and fresh

and dry weight of shoots and roots). So, higher significant ( $p \leq 0.01$ ) values of such traits were resulted from applying the treatments of 1.0 g/l NaCl combined with 2.0 g/l proline +0.3 g/l ascorbic acid in the first season and control (tap water) combined with 2.0 g/l proline + 0.3 g/l ascorbic acid in the second season, in addition to some other vegetative traits as stem diameter, branches number and leaf area in the two seasons. Meanwhile, the plants irrigated with 2.0 g/l NaCl without foliar spray (control) recorded the least significant values of such traits mentioned- earlier during both seasons. But the plants irrigated with tap water (control treatment) and were sprayed with 1.0 g/l proline +0.2 g/l ascorbic acid in the first season and those were not sprayed in the second one; gave rise to the highest significant shoot/root ratio. On the contrary, the plants irrigated with 1.0 g/l NaCl in the first season and 2.0 g/l NaCl in the second one combined with 2.0 g/l proline +0.3 g/l ascorbic acid; recorded lower significant shoot/root ratio. Similar results, more or less, were obtained by [34] on buttonwood where they found that foliar application the given plants with 0.5 and 1.0% proline; improved vegetative traits under 10% seawater [39]. Mentioned that supplementary proline significantly mitigated the adverse effects of salinity on *Olea europaea* cv., Chemlali, via the improvement of photosynthetic activity, relative water contents, chlorophyll, carotenoid and starch contents [40]. Reported that proline at 100 mg/l with humic acid at 50 kg/fed; improved significantly plant height, plant fresh and dry weights, technical stem length and number of branches/plant of flax plants grown under salinity conditions [41]. showed that exogenous application of ascorbic acid on *linonium stocksii* grown under saline conditions; improved growth traits and increased its salinity tolerance. Also [38], Mentioned that 100

mM ascorbic acid was more effective in reducing the salinity stress on *brassica repa*.

**3.2 Effect of salinity level, proline plus ascorbic acids concentrations and their interaction on chemical composition of *Duranta repens***

Regarding salinity level effect, data in Table (5) showed that either 1.0 or 2 g/l NaCl; caused a significant increase in leaf green color degree, proline content, Na (%) and Cl (%) in comparison to control treatment during both seasons; however, both levels, significantly, decreased water content, total carbohydrates, N, P and K(%) in the leaves when compared to the control treatment during both seasons. Notably, applying 2.0 g/l NaCl treatment was more effective in increase or decrease of such parameter than 1.0 g/l NaCl. These results may be taken place due to the excessive Na<sup>+</sup> and Cl<sup>-</sup> concentration in the soil which affect the absorption of many essential nutrients such as K, Ca, Mg, P and N [42]. In general, salt stress decreases total carbohydrates, but increased proline content [20]. Also, an increase the salinity of soil's water inhibits water uptake, water use efficiency and relative water content [43] and increases Na concentration in plant tissues [23]. Increasing Na<sup>+</sup> concentration in irrigation water decreases relative water content and increased Na<sup>+</sup> and Cl<sup>-</sup> contents in *Eucalytus comaldulensis* [44]. Additionally, Na, Cl and proline contents of leaves of walnut cultivars increased with increasing irrigation water salinity [45]. Leaves total carbohydrates, N, P and K (%) and total chlorophyll of *leucophyllum frutescens* were reduced but Na and Cl% increased because of increasing NaCl in irrigation water in a range of 3000-8000 ppm [46]. Recently [47], revealed that NaCl at 17-85 mM increased proline and Na content in *Carpinus betulus*.

**Table 5:** Chemical constituents of *Duranta repens* as affected by NaCl added to irrigation water during both 2017 and 2018 seasons.

Traits	NaCl treatments g/l					
	Control		1.0 g/l		2.0 g/l	
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
Proline content mg/g		6.98C		13.89B		19.85A
Na%	0.469C	0.449C	0.549B	0.520B	0.623A	0.592A
Cl%	1.41C	1.31C	1.72B	1.57B	2.03A	1.85A
N%	2.52A	2.66A	2.31B	2.16B	2.02C	1.67C
P%	0.276A	0.290A	0.246B	0.248B	0.228C	0.228C
K%	3.59A	3.43A	3.15B	3.05B	2.77C	2.66C
Total carbohydrates%	29.19A	28.40A	28.32B	26.91B	27.38C	25.63C
Leaf green color	30.44C	37.78B	33.23B	38.68A	37.85A	38.73A
Water content	67.62A	62.22A	60.78B	59.01B	61.91B	56.65C

Means for each season followed by the same letter (s) in every consecutive horizontal boxes are not significantly different at 0.05 level according to Duncan's Multiple Rang Test.

**Table 6:** Chemical constituents of *Duranta repens* as affected by proline + ascorbic acid during both 2017 and 2018 seasons.

Traits	Proline (Pr)+ascorbic acid (AA) [g/l] treatments									
	0		1.0 g Pr +0.2 g AA		1.0 g Pr +0.3 g AA		2.0 g Pr +0.2 g AA		2.0 g Pr +0.3 g AA	
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
Proline content mg/g	14.61A			12.77D		13.28C		13.40C		13.82B
Na%	0.577A	0.549A	0.517E	0.490E	0.532D	0.506D	0.546C	0.520C	0.562B	0.536B
Cl%	1.86A	1.72A	1.59E	1.46E	1.65D	1.51D	1.72C	1.57C	1.79B	1.61B
N%	2.10E	1.99E	2.45A	2.35A	2.38B	2.25B	2.29C	2.16C	2.18D	2.09D
P%	0.230E	0.240E	0.262A	0.270A	0.252B	0.263B	0.247C	0.255C	0.237D	0.246D
K%	3.01E	2.88E	3.34A	3.22A	3.25B	3.15B	3.17C	3.02C	3.09D	2.95D
Total Carbohydrates%	27.83E	26.48E	28.78A	27.48A	28.50B	27.21B	28.30C	26.99C	28.06D	26.48D
Leaf green color	30.58E	36.10E	31.57D	36.80D	32.75C	38.17C	36.54B	39.41B	37.75A	41.48A
Water content	60.93B	55.80E	61.83B	57.36D	63.32AB	59.00C	65.33A	60.94B	65.75A	63.37A

Means for each season followed by the same letter (s) in every two consecutive horizontal or vertical boxes are not significantly different at 0.05 level according to Duncan's Multiple Rang Test.

**Table 4:** Average values of some vegetative growth traits of *Duranta repens* as affected by interaction treatments between NaCl and proline + ascorbic acid during both 2017 and 2018 seasons.

Treatments		Plant height (cm)		Plant diameter (cm)		Stem diameter (cm)		Branches number/plant		Plant leaf area (cm <sup>2</sup> )		Shoot fresh weight g/plant		Shoot dry weight g/plant		Roots fresh weight g/plant		Root dry weight g/plant		Shoot/root ratio	
NaCl	Pr +AA	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
Control	0	59.40jk	78.31e	41.85k	49.86e	0.40e	0.56c	5.67fg	8.33b	59.46f	58.76f	30.26j	57.30d	10.42j	19.12c	8.46j	16.79e	3.23ij	5.57c	3.60b	2.98a
	1.0 g Pr +0.2 g AA	63.32i	86.40d	43.64i	53.82d	0.42e	0.63b	7.00ef	9.67a	60.53f	60.36ef	36.46i	58.65d	12.64i	20.08bc	10.26hi	18.51d	4.12h	6.47b	3.65a	2.95b
	1.0 g Pr +0.3 g AA	67.57h	87.56c	44.16h	56.88c	0.57cd	0.64b	7.33e	10.33a	62.52e	62.46cd	46.78h	61.33c	16.45h	20.74abc	15.45f	20.74c	5.83f	6.74b	3.11d	2.93b
	2.0 g Pr +0.2 g AA	70.26g	90.59b	49.16f	59.34b	0.52d	0.70a	8.67d	10.67a	65.59d	63.45bc	51.46g	63.43b	18.24g	21.62ab	17.44e	21.39b	6.85e	7.37a	2.98f	2.92bc
	2.0 g Pr +0.3 g AA	74.41e	92.67a	51.35d	60.03a	0.57cd	0.71a	9.33cd	10.67a	66.53d	65.43a	58.46f	65.22a	20.43f	22.33a	17.7e	22.37a	8.12d	7.57a	3.29c	2.92bc
1 g/L	0	72.20f	60.67k	46.85g	30.86j	0.57cd	0.40fg	9.00d	6.00efg	69.37c	59.58ef	61.57e	32.38i	21.64e	11.04e-h	21.06d	11.56i	8.20d	4.26e	2.93g	2.93b
	1.0 g Pr +0.2 g AA	77.36d	65.22i	49.66e	34.06i	0.63bc	0.43ef	10.33c	6.33cde	70.37bc	60.47ef	75.37d	34.30h	26.54d	11.82efg	26.38c	13.49f	10.46c	4.43e	2.87h	2.89cd
	1.0 g Pr +0.3 g AA	81.34c	68.61h	53.16c	36.83h	0.67ab	0.46e	11.67b	6.67cde	71.40b	62.50cd	78.3c	36.46g	27.87c	12.53de	25.69c	15.61f	10.63c	5.40c	2.90gh	2.89d
	2.0 g Pr +0.2 g AA	87.50b	72.19g	54.85b	38.14g	0.72a	0.47e	12.33b	7.33bcd	73.44a	64.49ab	85.35b	39.39f	29.85b	13.83d	31.06b	17.24e	12.82b	6.48b	2.74i	2.86de
	2.0 g Pr +0.3 g AA	90.61a	74.65f	58.85a	40.20f	0.75a	0.52d	14.00a	7.67bc	74.55a	65.15a	95.33a	41.31e	33.43a	10.66	34.33a	18.40d	13.55a	6.54b	2.72i	2.82f
2 g/L	0	52.43m	55.14n	33.65n	17.83o	0.36e	0.36h	5.00g	5.00g	57.37g	56.47g	21.55m	24.41l	7.43l	8.54i	7.37j	8.84k	2.94j	3.26f	3.09d	2.83ef
	1.0 g Pr +0.2 g AA	54.41l	57.18m	34.01n	19.81n	0.39e	0.38gh	5.00g	5.33fg	60.35f	58.74f	23.46l	27.19k	7.44l	9.56hi	7.59j	10.47j	2.98j	3.70f	3.02e	2.83e
	1.0 g Pr +0.3 g AA	55.51l	58.75l	34.67m	22.03m	0.39e	0.39fgh	5.67fg	5.67efg	60.57f	59.37ef	25.46k	29.41j	7.83k	10.25fgh	8.70ij	11.49i	3.59i	4.35e	2.92g	2.84ef
	2.0 g Pr +0.2 g AA	58.51k	60.64k	40.14l	25.16l	0.41e	0.43ef	6.00fg	6.00efg	63.61e	60.34ef	30.42j	31.54i	10.44j	11.35efg	11.35gh	13.39h	4.56g	4.67de	2.72i	2.78g
	2.0 g Pr +0.3 g AA	60.40j	63.03j	42.83j	28.10k	0.42e	0.44e	6.67ef	6.67cde	65.28d	61.17de	36.44i	34.33h	12.63i	12.41def	12.09g	14.18f	4.73g	5.09cd	3.03e	2.77g

Means for each season followed by the same letter (s) in every consecutive vertical boxes are not significantly different at 0.05 level according to Duncan's Multiple Rang Test.

**Table 7:** Chemical constituents of *Duranta repens* as affected by interaction treatments between NaCl and proline + ascorbic acid during both 2017 and 2018 seasons.

Treatment		Proline content g/g		Na%		Cl%		N%		P%		K%		Total carbohydrates%		Leaf green color degree		Water content	
NaCl	P+AA	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
Control	0		7.83j	0.495j	0.477k	1.55i	1.44i	2.33e	2.50e	0.261d	0.275d	3.40e	3.24e	28.74e	27.91e	27.85o	35.62l	67.70ab	57.84i
	1 g P +0.2 g AA		6.12n	0.442n	0.419o	1.28m	1.20l	2.71a	2.85a	0.293a	0.306a	3.79ab	3.63a	29.63a	28.89a	28.44n	36.01i	67.83ab	59.64g
	1 g P +0.3 g AA		6.55m	0.456m	0.436n	1.34l	1.25k	2.62b	2.74b	0.282b	0.298b	3.68	3.54b	29.44b	28.63b	29.53l	37.42g	67.96ab	62.12d
	2 g P +0.2 g AA		7.00l	0.469l	0.447m	1.42k	1.30j	2.52c	2.65c	0.278b	0.287c	3.58c	3.41c	29.20c	28.43c	32.44i	39.06d	70.08a	64.46b
	2 g P +0.3 g AA		7.40k	0.483k	0.464l	1.48j	1.33j	2.40d	2.58d	0.268c	0.281d	3.49d	3.34d	28.95d	28.15d	33.95h	40.76b	64.53abc	67.04a
1 g/L	0		14.91e	0.582e	0.552f	1.86e	1.72e	2.12gh	1.98j	0.231g	0.233jk	2.99i	2.90i	27.82i	26.41j	29.34m	36.25k	55.15e	55.84l
	1 g P +0.2 g AA		12.76i	0.517i	0.490j	1.58i	1.46hi	2.47c	2.34f	0.261d	0.262e	3.31f	3.22e	28.82de	27.43f	30.75k	37.12h	57.34de	57.10j
	1 g P +0.3 g AA		13.46h	0.534h	0.502i	1.65h	1.5h	2.40d	2.24g	0.254e	0.256f	3.22g	3.14f	28.57f	27.15g	31.64j	38.64f	60.76cd	58.74h
	2 g P +0.2 g AA		13.92g	0.547g	0.520h	1.71g	1.58g	2.32e	2.17h	0.246f	0.248g	3.18g	3.02g	28.31g	26.90h	36.23e	39.53c	63.00bc	60.29e
	2 g P +0.3 g AA		14.39f	0.565f	0.536g	1.78f	1.62f	2.21f	2.08i	0.236g	0.239hi	3.09h	2.95h	28.07h	26.65i	38.17c	41.84a	67.64ab	63.05c
2 g/L	0		21.09a	0.653a	0.618a	2.17a	1.99a	1.86k	1.49o	0.197j	0.213l	2.64m	2.51m	26.93l	25.13o	34.55g	36.43j	59.95cde	53.72n
	1 g P +0.2 g AA		19.43cd	0.592e	0.560e	1.90e	1.73e	2.18fg	1.85k	0.231g	0.242h	2.92j	2.81j	27.90hi	26.12k	35.53f	37.27gh	60.32cde	55.33m
	1 g P +0.3 g AA		19.83b	0.608d	0.579d	1.96d	1.78d	2.10h	1.76l	0.219h	0.235ij	2.85k	2.76k	27.50j	25.87l	37.08d	38.44e	61.24cd	56.12k
	2 g P +0.2 g AA		19.27d	0.623c	0.592c	2.04c	1.83c	2.03i	1.67m	0.216h	0.229k	2.75l	2.64l	27.40j	25.64m	40.95b	39.64c	62.92bc	58.07i
	2 g P +0.3 g AA		19.66bc	0.638b	0.608b	2.10b	1.89b	1.93j	1.60n	0.206i	0.218l	2.68m	2.55m	27.15k	25.38n	41.13a	41.85a	65.09abc	60.03f

Means for each season followed by the same letter (s) in every consecutive vertical boxes are not significantly different at 0.05 level according to Duncan's Multiple Rang Test.

Concerning proline plus ascorbic acid treatments, results in Table (6) pointed out that all combination treatments of proline + ascorbic acid; caused increments in chemical parameters in comparison to control treatment except for proline, Na and Cl (%) and the differences among the used treatments reached the significant level in all parameters during the two seasons. So, the significantly highest values of leaf green color degree and water contents were recorded due to spraying plants with 2.0 g/l proline +0.3 g/l ascorbic acid, while those of total carbohydrates, N, P and K (%) resulted from applying 1.0 g/l proline +0.2 g/l ascorbic acid treatment, but the significantly highest proline in the 2<sup>nd</sup> season, Na and Cl (%) was stemmed out control treatment in the two seasons. On the other hand, the least significant values of such parameters were resulted from control plants except for proline in the 2<sup>nd</sup> season, Na and Cl % were recorded owing to foliar spraying the plants with 1.0 g/l proline +0.2 g/l ascorbic acid in the both seasons. These results were supported by those of <sup>[36]</sup> on *Khaya senegalensis*, <sup>[48]</sup> on *Grevilla substa*, <sup>[40]</sup> on flax plants, <sup>[49]</sup> rate on chilli genotypes, and <sup>[37]</sup> on *Brassica rapa*.

With regard to interaction between salinity levels and proline plus ascorbic acid treatments, results in Table (7) had exhibited differential effects on the measured chemical parameters. Whereas, leaf green color degree demonstrated higher significant ( $p < 0.01$ ) value which resulted from applying the treatment comprise of 2.0 g/l NaCl in combination with 2.0 g/l proline + 0.3 g/l ascorbic acid, meanwhile the least significant value was recorded for control plants during both seasons. While the significantly highest water content was resulted from the control plants which sprayed by 2.0 g/l proline plus 0.2 g/l 0.3 g/l ascorbic acid in during both seasons, but the lower significant water content was recorded for plants received 2.0 g/l NaCl without foliar sprayed within both seasons. The irrigated plants with tap water and sprayed with 1.0 g/l proline +0.2 g/l ascorbic acid had significantly higher percentage of carbohydrates, N, P and K, while those received 2.0 g/l NaCl without foliar sprayed had lower significant percentage of such traits in the two seasons. The received plants 2.0 g/l NaCl and un-sprayed and those irrigated with tap water and sprayed with 1.0 g/l proline +0.2 g/l ascorbic acid resulted in higher and lower significant values of proline content, Na and Cl (%), respectively. These results may be attributed to either proline or ascorbic acid exerted significant functions against salinity stress <sup>[50, 51]</sup>. Also, the proline concentration of cell, tissues and plant organs are regulated by the interplay of biosynthesis, degradation and intra, as well as intercellular transport process <sup>[52]</sup>. Moreover, the lower accumulation of Na<sup>+</sup> ions in proline treated plants compared to their corresponding salinity treatment showed a profound positive effect of proline on the ability of roots to exclude the salt ions from the xylem sap and streaming to shoots. Similar performances were noticed on *Brassica rapa* <sup>[38]</sup>.

#### 4. Conclusion

*Duranta repens* is a sensitive plant to salinity, and in order to improve its tolerance to 2.0 g/l NaCl, it is recommended to be sprayed with 2.0 g/l proline + 0.3 g/l ascorbic acid 7 times with one-month interval.

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