ORIGINAL ARTICLE

Influence of NaCl salinity on growth analysis of strawberry cv. Camarosa

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In order to study of salinity effect on growth analysis of strawberry, a greenhouse experiment was conducted in Vali-e-Asr University of Rafsanjan in 2010. This study was carried out RCBD design with 4 replications to determine the influence of salinity (30, 60, 90 Mmol and control with distilled water) on strawberry growth analysis. Results indicated that relative growth rate (RGR), crop growth rate (CGR), leaf area ratio (LAR) and dry matter accumulation were decreased with increasing salinity. The lowest RGR, CGR and LAR were observed in 90 Mmol NaCl salinity. Results also indicated that maximum dry matter accumulations were observed in 1050, 1200 and 1400 degree days in 30, 60 and 90 Mmol NaCl salinity, respectively. Water salinity more than 30 Mmol NaCl L⁻¹ will decreased fresh fruit yield more than 50 percent in hydroponics strawberry production. Dry mass partitioning in NaCl-stressed plants was in favor of crown and petioles and at expense of root, stem and leaf whereas leaf, stem and root DM progressively declined with an increase in salinity.

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Strawberry is considered as a NaCl salinity sensitive species, but differences between cvs. Exist (Keutgen and Pawelzik 2009; Kaya et al. 2002). Negative influences of NaCl salinity on strawberry plant growth and fruit productivity have been reported by Keutgen and Pawelzik 2009, D'Anna et al. (2003), Awang and Atherton (1995a,b) and Awang et al. (1993). The salt stress results in the development of leaf necrosis and accelerated leaf senescence, thus reducing photosynthetic capability of the plants. In consequence, assimilation of carbohydrates available for fruit production is reduced (Saied et al. 2005; Keutgen and Keutgen 2003; Giuffrida et al. 2001). Selection of an optimal planting and harvest date will require a better understanding of the growth that occur during maturation and of the feeding value of the crop at different stages of development (Mikkelsen 1981). Nutritive value has been related to accumulate growing degree days (GDD), crop age in days, leaf stem ratio, stand height and codified to morphological stage (McMaster and Wilhelm 1997). The GDD, involving the sum of daily mean temperature above a given base temperature, is widely used with crops to predict relative maturity during the growing season (Bourgeois et al. 2000; Mikkelsen 1981). A base temperature from 0 to 5° C was generally adequate for all strawberry cultivars to compute growing degree days between sowing and maturity, although a base temperature of 4°C was often reported to be the most satisfactory for strawberry (Bernadine et al. 1988). Specific leaf area (SLA), an indicator of leaf thickness, has often been observed to be reduced under salinity conditions (Giuffrida et al. 2001). Decrease in SLA in salt stressed plants maybe due to the different sensitivity of photosynthesis and leaf area expansion to soil salinity. Salinity stress affects leaf expansion earlier than photosynthesis (Lambers et al. 2008; Kaya et al. 2002). Reduction of SLA is assumed to be a way to improve water use efficiency (WUE) (Rahimi et al. 2011). This is because thicker leaves usually have a higher density of chlorophyll and proteins per unit leaf area and, hence, have a greater photosynthetic capacity than thinner leaves. Information on dry matter partitioning between various plant parts is an essential step in the

development of crop simulation models (Bourgeois et al. 2000). Furthermore, the value of many agricultural experiments could be greatly enhanced if data about growth and the partitioning of this growth were available. It would allow better interpretation of the results within the context of processes and resource exploitation (McMaster and Wilhelm 1997). Little information can be found in the literature on the changes in yield potential and growth analysis of strawberries planted in greenhouse. The objectives of this study were to investigate the effects of salinity stress on dry matter partitioning, specific leaf area (SLA), relative growth rate (RGR) and crop growth rate (CGR) of strawberry cv. Camarrosa, and to evaluate the significance of changes in these features for plant performance during salinity stress.

MATERIALS AND METHODS

Plant material and growth conditions

Experiment was conducted from the end of October to the mid of June during 2009 and 2010 in Iran, with strawberry (Fragaria Rafsanjan, ananassa cv. Camarosa). Cold stored strawberry each had a well-developed crown with 8-10 mm diameter were established at the mid of November to the mid of June during 2009 and 2010 in 15 L black plastic bags and fixed under an open hydroponics system. Day/light temperature ranged between 23-28/14-18 °C. The bags were watered two or three times a day depending on greenhouse temperature by mineral nutrients with 200mL of modified Hoagland solution (Table 1) per plant with a complete nutrient solution and pH of solution adjusted in 6.5 using nitric and sulfuric acids. Three weeks after planting, NaCl treatments were initiated with 200mL of the solutions containing 0, 30, 60 or 90 Mmol NaCl L⁻¹ seven times a week to each plant. The EC and pH of drainage water from pots were

checked every week in the drainage water from the pots and an additional 200mL of dematerialized was applied to minimize EC and pH changes in the root zone. Four different salinity levels of 0, 30, 60 90 Mmol NaCl L⁻¹ (equal 1.7, 3.5, 5 and 7.4 dS.m⁻¹) were used in four replications. The salinity treatments were arranged in a completely randomized design with 3 replicates per treatment and 12 plants per unit of the experiment. Surpluses of solutions were allowed to pass the containers to ensure NaCl stress in the root medium at a given concentration, but to avoid anoxia by water logging. Varying osmotic potentials in the root medium temporarily may facilitate water uptake and, hence, strengthen the effect of NaCl stress while reducing those of water stress, thus allowing focusing onto the salt-specific impacts. To improve fruit quality, runners were removed at emergence. Fruits were harvested at the optimum stage of physiological maturity, when 90% of the fruit surface had reached a fully red color. At the end of each experiment, 5 plants were randomly collected from each treatment every 15 days intervals 30 DAT to measure Shoot dry matter and leaf area. The leaf area was measured using a leaf area-meter (Li-Cor, Moldel Li- 1300, USA) by the deduction of necrotic leaf area from total area of the leaf.

Plant growth

According to Lambers et al. (2008), leaf area ratio (LAR) was defined as the amount of leaf area per unit of plant dry mass. It was the product of specific leaf area (SLA), the amount of leaf area per unit of leaf dry mass. Net assimilation rate (NAR) of the plants represented the net increment in plant dry mass per unit of mean leaf area during the experiment. Relative growth rate of plants (RGR) was calculated by multiplying LAR and NAR. Fruit fresh weight per plant was calculated. RGR=1/DM (Δ DM/10GDD) CGR=RGR×DM NAR=RGR/LAR LAR= (LA₂/ W₂ + LA₁/ W₁)/2 SLA= (LA₂/ w₂ + LA₁/ w₁)/2

Statistical analysis

The data were analyzed over the experiment and harvest maturity through regression analysis and via an analysis of variance (ANOVA), using SAS (v 9.0 USA). All the parameter analyses were performed in duplicate and averaged for the statistical analyses. The resulting data were averaged over the replicates before regression analysis and used as individual observations for ANOVA. The data were regressed on the growing degree days (GDD) with a 4°C base temperature (Mikkelsen, 1981) and on the age in days from the seeding date, as independent variables. The accumulated GDD was calculated using the following formula for each year: P {[(Tmax + Tmin)/2] 4° C};

where Tmax and Tmin are the daily maximum and minimum temperatures, respectively, in degree Celsius. The average temperature [(Tmax + Tmin)/2] was set equal to 4°C if less than 4°C (McMaster and Wilhelm 1997). Linear and quadratic regressions were compared using the Draper and Smith (1998) stepwise selection procedure to select the best regression model at the 0.05 probability level. The best equation for each parameter was selected using coefficient of determination and root mean square error (RMSE). All the determination coefficients (R^2) reported in this paper were adjusted for degrees of freedom. When no differences were found a single regression line was determined for the pooled

data. The yield data were analyzed via ANOVA, with their significance reported at a

0.05 probability level using the general linear model of SAS.

	mg.l ⁻¹		mg.l ⁻¹
Mg	50	Ν	180
Κ	270	PO_4	65
Fe	5	SO_4	67
MnSO ₄ , 4H ₂ O	1	(NH ₄) ₆ MoO ₂₄ , 6H ₂ O	0.06
ZnSO ₄ , 7H ₂ O	0.2	В	0.08

Table 1 Composition of modified Hoagland solution (pH 6.5) for soilless culture of plant



Figure 1. Leaf area (LA) as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.



Figure 2. Total dry matter (DM) as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.



Figure 3. Leaf area ratio (LAR) as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.

RESULTS AND DISCUSSION

Growth analysis

Salinity significantly affected leaf area expansion in this cultivar. In all treatment except 90 Mmol NaCl, Total leaf area per plant increased with stage of growing (700 °C of GDD) to fruiting stage (1150 °C of GDD) from about 400 to 900, 730 and 680 CM² in 0, 30 and 60 Mmol NaCl, Respectively (Fig 1). The asymmetric logistic peak curve shown in Fig. 1 provided a good fit to the data obtained in all the growth strawberry (Fig.1). Leaf area of the whole plant decreased in all treatment with advancing maturity with different trends and with high coefficient of determination. It is clear that this cultivar of strawberry will decreased LA with advancing GDD from 1400 degree days in all treatments. Curves describing changes in LAI reached a maximum around fruiting stage and then decreased until maturity, due to the senescence and death of leaves (Fig. 1). The variable responsible for the differences was the thermal time needed to reach

the maximum LA (Fig. 1). The immediate response of salt stress is reduction in the rate of leaf surface expansion leading to cessation of expansion as salt concentration increases (Khan et al. 2000). Salt stress also results in a considerable decrease in the fresh and dry weights of leaves, stems, and roots (Parida et al. 2004; Flower et al. 1977). Most likely salt toxicity was responsible for the occurrence of this leaf necrosis. The maximum rate of increase in leaf area per plant, which determines the rate of increase in the photosynthetic capacity of the plant, took place at 1200 GDD from sowing (Fig 1). Total dry matter (DM) of strawberry plant increased with advancing growth from 9 gr/plant to 14 gr/plant in 0 and 30 Mmol NaCl treatments in the same GDD (around 1300 °C of GDD). Results also indicated that DM were significantly decreased in 60 and 90 Mmol/NaCl compared with 0 and 30 Mmol NaCl, corresponding to 1100 and 1500 GDD (Fig 2). In all salinity treatment except 90 Mmol NaCl with linear model, DM changes with quadratic model. The peak

value of DM in control and 30 Mmol NaCl treatments were observed in 1300 GDD which were 45 and 26% higher than 60 and 90 Mmol NaCl. Total dry weight accumulation of plants was not noticeable inhibited at low salinities (30 Mmol NaCl), but it was significantly inhibited at higher salinity. Curves decreasing after reaching a maximum, such as the curves describing changes in leaf area per plant, Total dry matter (DM) per plant and Curves not decreasing after reaching a maximum, such as the patterns of the changes in DM (Fig. 1 and 2). Thermal time from sowing to the maximum rate dry weight accumulation among different salinity ranged from 1100 GDD to 1300 GDD. On average, in control and 30 Mmol NaCl needed 109 more GDD to reach their maximum rate dry weight accumulation than 60 and 90 Mmol NaCl did. This result was expected, since it is known that salinity have a lower rate of development than control (Khan et al. 2000) and have a longer cycle (Kaya et al. 2002). Leaf area ratio (LAR) and Specific leaf area (SLA) significantly decreased in cv. Camarosa at 60 and 90 Mmol NaCl L⁻¹. It means that this cultivar of strawberry developed leaf thickness with increasing salinity while leaf area per plant significantly decreased by increasing salinity (Fig. 3, 4). In all salinity treatment, SLA and LAR with linear model decreased with increasing salinity while SLA with linear and LAR with quadratic model were increased in control. The peak value of SLA and LAR were observed in 1800 °C and 1500 of GDD in control plant, respectively. SLA and LAR of plants were not noticeable decresed at low salinities (30 Mmol NaCl), but it was significantly decreased at higher salinity. The crop growth rates (CGR) were first increased until 1100 °C of GDD in control and 30 Mmol NaCl with different trends all in quadratic model. It is progressively decline from the flowering stage (870 GDD) in 60 and 90 Mmol

with advancing maturity with the same trends and with high coefficient of determination (Fig 5). It is photosynthesis clear that and drv matter accumulation will decreased with advancing salinity levels and also advancing GDD from 1150 degree day in all treatments in this cultivar of strawberry. It would be due to higher respiration and more upper leaves shadow. The relative growth rate (RGR) of shoots and net assimilation rate (NAR) were calculated with advancing maturity to determine the optimal growth stage. In All treatment, RGR and NAR were progressively linear declined with advancing maturity and increasing salinity in this cultivar due to decreasing more dry matter accumulation, reduction of leaf area and CGR in addition to an increase of necrotic leaf area (Fig 6, 7). Low values in RGR, NAR and CGR in response to salinity are could also be associated with a decrease in photosynthesis rates, an increase in respiration rates, or an increase in relative of nonphotosynthesis tissues participating in respiration (Turhan and Eris, 2004). This result is consistent with numerous studies having shown a negative effect of salinity on RGR and NAR (Parida et al. 2005; D'Anna et al. 2003). In our experiments, cv. Camarosa was characterized by a large necrotic leaf area due to NaCl stress, which was accompanied by lower leaf area (Fig. 1). These results are well in line with those of Saied et al. (2005) and Turhan and Eris (2004). In this study, salinity variability found could be associated with plant phonology, since in most cases it was due to differences between GDD. Such was the case for the significant salinity variation found for the time needed to reach (i) the maximum LAI, (ii) CGR, RGR and (iii) the maximum dry weight accumulation in the whole plant. The differences found in this study for the length of dry matter accumulation phase between different salinity were expected, since the length of this phase depends on the photoperiod and temperature requirements of the crop (Bourgeois et al. 2000). The remarkable finding of this work is that the asymmetric logistic peak curve described logistic curve fitted accurately to asymptotic relationships (CGR, Dry weight) and to peak-type relationships (LA and dry weight of whole plant). This may make growth analysis, which is generally expensive and time consuming, simpler and more accurate (Keutgen and Pawelzik 2009). Consequently, for strawberry seedlings growing in saline conditions, NAR is the most important parameter reflecting differences in RGR, whereas LAR and SLA are of secondary importance. NAR may then be regarded as a reliable indicator of salinity stress and salinity tolerance in strawberry in hydroponic culture.



Figure 4 Specific leaf area (SLA) as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.



Figure 5 Relative area rate (RGR) as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.

JOURNAL OF STRESS PHYSIOLOGY & BIOCHEMISTRY Vol. 7 No. 4 2011



Figure 6 Crop growth rate (CGR) as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.



Figure 7 Net assimilation rate (NAR) as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.

The most reduced DM organ plants in this plant was observed by 62% in stem DM per plant following by Leaf and root DM with 45% and 48%, respectively. The least DM was observed in petioles DM (Fig. 8). Total DM per plant was reduced by 63% in this strawberry cultivar at 90 Mmol NaCl L-1. The root to shoot ratio of DM did not change under NaCl salinity and different substrate and their interaction, While NaCl stress significantly decreased root, leaf and stem DM (Fig. 8). In consequence, dry mass partitioning in NaCl-stressed plants was in favor of crown and petioles and at expense of root, stem and leaf whereas leaf, stem and root DM progressively declined with an increase in salinity (Fig. 5). These results are well in line with those of Saied et al. (2005) and Turhan and Eris (2004).

Fruit yield

The physiological maturity of the fruit was commenced in 72, 69, 58 and 54 days from

transplanting in 0, 30, 60 and 90 Mmol NaCl L-1, respectively corresponding to 935, 905, 854 and 838 of GDD. Fruit yield per plant was significantly affected by salinity. Among the different salinity levels, the highest fruit yield (250±32.5 g.plant⁻¹)

was found in control followed by 30 Mmol NaCl. In general, water salinity more than 30 Mmol NaCl L⁻¹ will decreased fresh fruit yield more than 50 percent in hydroponics strawberry production (Fig 9).



Figure 8 Dry mass distribution as a function of the growing degree days (GDD) during growth for strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.



Figure 9 Fruit yield of strawberry cv. Camarosa at 0, 30, 60 and 90 Mmol NaCl.

CONCLUSION

Obviously, plant growth was significantly reduced in cv. Camarosa and this decrease was due to a reduction of leaf area, in addition to an increase of necrotic leaf area. In our experiments, cv. Camarosa was characterized by a large necrotic leaf area due to NaCl stress, which was accompanied by lower water content of the leaf (Fig. 6). Specific leaf area (SLA) and Leaf area ratio (LAR) significantly decreased in cv. Camarosa at 60 and 90Mmol. The presence of NaCl more than 30 Mmol NaCl in the root medium induced a noticeable decreased in RGR, CGR which cause decreased LA, DM, RGR and CGR. Results also showed that the presence of NaCl in the root medium induced a decreased in RGR, CGR. Obviously, plant growth was significantly reduced in cv. Camarosa and this decrease was due to a reduction of leaf area. Water salinity more than 30 Mmol NaCl L⁻¹ will decreased fresh fruit yield more than 50 percent in hydroponics strawberry production. Dry mass partitioning in NaCl-stressed plants was in favor of crown and petioles and at expense of root, stem and leaf whereas leaf, stem and root DM progressively declined with an increase in salinity.

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