



Full Length Article

Exploring the Potential of Quinoa Accessions for Salt Tolerance in Soilless Culture

Muhammad Aamir Saleem^{1*}, Shahzad Maqsood Ahmed Basra¹, Irfan Afzal¹, Hafeez-ur-Rehman¹, Shahid Iqbal¹, M. Sohail Saddiq¹ and Safina Naz²

¹Department of Agronomy, University of Agriculture, Faisalabad, Pakistan

²Department of Horticulture, Bahauddin Zakariya University, Multan, Pakistan

*For correspondence: aamirsaleem28@gmail.com

Abstract

Pakistan has more than 6 million hectares of salt affected land and quinoa is tested as a facultative halophyte having super food characteristics. A hydroponic study was conducted to explore salt tolerance of two quinoa accessions, Ames-13737 (Q7) and PI-634919 (Q9) under a range of NaCl levels (0, 100, 200 and 300 mM) in wirehouse during 2016. Plant nursery was raised, and at four leaf stage, the seedlings were transferred to plastic tubs containing 20 L half strength Hoagland's solution as nutrient source. Salinity was developed incrementally to avoid osmotic shock after two days of transplanting. Results showed that growth (shoot length, root length and dry weight/plant) reduced drastically in Q9 by increasing the salinity level as compared to Q7. However, a comparable growth was observed in Q-7 at 0 and 100 mM i.e. the dry weight at 0 and 100 mM salinity were 0.8643 g and 0.8125 g, respectively. Furthermore, genotype Q7 was found better than Q9 by producing 10% more dry weight (0.2790 g) at the highest salinity level which was linked to 57% more leaf K⁺ accumulation (28.64 mg K⁺ g⁻¹ dry weight) as compared to Q9 (12.54 mg K⁺ g⁻¹ dry weight). It is concluded that tolerance of Q7 to salt stress might be due to more absorption of K⁺ by the roots at increased Na⁺ level. © 2017 Friends Science Publishers

Keywords: Hydroponics; Quinoa; Salt stress; Potassium; Growth

Introduction

Increasing salinity has become a global threat to agriculture, particularly in arid and semi-arid regions including Pakistan (Panta *et al.*, 2014). According to FAO and ITPS report (2015), more than 100 countries have salt affected soils and their global extent is evaluated at about 1 billion hectares. Martinez-Beltran and Manzur (2005) reported that irrigated area of 0.25–0.5 million hectares becomes unproductive globally due to salinity buildup every year. These soils considerably decrease the growth and development of plants and thus are considered problematic soils. In Pakistan, salinity buildup usually occurs by the application of saline water. On an average, 26% of irrigated area in Pakistan is salt affected (Shahid *et al.*, 2013). Irrigated area of estimated 6 million hectares is affected by soil salinity resulting in a loss of 62% in agricultural returns in Pakistan (Kazmi *et al.*, 2012). As most of the crop plants are salt sensitive non-halophytes (Greenway and Munns, 1980; Koyro and Huchzermeyer, 1999), the losses are incalculable. Primary salinity is caused due to the disintegration of primary minerals from the surface rocks (weathering). Munns and Tester (2008) reported another cause of primary salinity which is the deposition of salts carried in water and wind near coastal areas. Secondary salinity, which is caused by

anthropogenic activities is especially important in Pakistan, where saline underground water used for irrigation causes the buildup of salts. According to a report, about 23% of irrigated lands in Pakistan are being deteriorated due to saline intrusion from un-irrigated lands (FAO and ITPS, 2015).

Harmful effects of salinity include decreased water availability due to increased osmotic stress usually triggered by high amounts of dissolved salts in the soil, toxicity of ions due to increased concentrations of sodium, magnesium and chloride ions, production of reactive oxygen species (ROS) and mineral imbalances especially potassium deficiency (Munns and Tester, 2008; Shabala and Cuin, 2008). Thus, major physiological phenomena like photosynthesis, lipid metabolism and protein synthesis are hampered (Heuer, 2005), which have severe effect on growth and performance of plants. Elevated Na⁺ concentrations in cytosol triggers cell death due to membrane disintegration (Shabala, 2009). Thus, it is need of the day to counter and alleviate the harmful effects of salinity on arable crop production. It is necessary to introduce new approaches to tackle these issues. Different solutions to increasing salinity have been introduced i.e. reclamation of salt affected areas, breeding, use of salt tolerant germplasm etc. (Yilmaz *et al.*, 2004). One potential

and climate resilient strategy is the introduction of those new crops, which can tolerate high levels of salinity in soil and may allow irrigation with saline water *i.e.*, the use of halophytic crop plants (Koyro *et al.*, 2008).

Family Amaranthaceae is famous for having potential halophytes. Quinoa (*Chenopodium quinoa* Willd.) is one of the most potential members of this family. It is a facultative halophyte with its germplasm being able to handle salinity even at the levels that exist in sea water of 400 mM NaCl (Jacobsen *et al.*, 2001, 2003; Hariadi *et al.*, 2011). Pearsall (1992) reported quinoa cultivation in the Andes region for over 7000 years, which is known for its poor soil conditions and harsh climate. Thus, quinoa can acclimatize accordingly to tolerate frequent drought, frost and other harsh conditions (Jacobsen *et al.*, 2009). Quinoa not only has outstanding ability to tolerate stress environments but it is also considered a super-food for its nutrition. Quinoa grain has exceptional composition of vital amino acids, is abundant in vitamins (A, B₂ and E) and contains many minerals like copper, magnesium, iron, calcium, lithium and zinc, and it also proves to be an excellent source of carbohydrates and fatty acids crucial for human nourishment (Repo-Carrasco *et al.*, 2003). Due to low input requirement but high nutrition, the crop has also been nominated by FAO to guarantee food security in the 21st century (FAO, 1998). Despite the potential of quinoa as a high nutrition, low input and climate resilient crop and the work of many scientists, there are still some efforts to be put in the adaptation and screening of salt tolerant germplasm in Pakistan.

Large amount of energy is required for the biosynthesis of organic solutes causing yield losses (Shabala and Shabala, 2011). Thus, facultative halophytes like quinoa may balance their turgor by compartmentalizing Na⁺ and Cl⁻ in cell vacuoles of shoot and produce compatible solutes just for cytosolic osmotic adjustment (Flowers and Colmer, 2008; Shabala and Mackay, 2011). It is believed that this sequestration of sodium ions is attained by tonoplast Na⁺/H⁺ antiporters (Flowers and Colmer, 2008) and pyrophosphatase (Guo *et al.*, 2006; Krebs *et al.*, 2010). Furthermore, Na⁺ and Cl⁻ ions stored in cell vacuoles serves as cheap osmolyte to maintain cell turgor by replacing potassium in halophytes (Flowers *et al.*, 1977; Glenn *et al.*, 1999).

Mannitol, proline and *myo*-inositol are the compatible solutes present in quinoa (Ruffino *et al.*, 2010) with the ability to scavenge ROS (Szabados and Saviouré, 2010). Tolerance of quinoa to salinity may be credited to its efficient retention of K⁺ (Ruffino *et al.*, 2010). Salinity caused a decrease in transpiration and thus gas exchange in quinoa (Sanchez *et al.*, 2003). It has been reported that quinoa reduces stomatal density and cuticular pores under salinity (Razzaghi *et al.*, 2011), which may adjust water use efficiency in these conditions (Orsini *et al.*, 2011; Shabala *et al.*, 2012).

Although, quinoa is a facultative halophyte but still

huge diversity exists in its germplasm for salinity tolerance under different salinity and climatic conditions. Hence, the proposed study was conducted with objective to identify salt tolerant quinoa genotype on the basis of morphological, physiological, biochemical responses at different salinity levels in local climatic conditions.

Materials and Methods

Plant Material and Growth Conditions

The plant material used for the experiment consisted of two quinoa genotypes Q7 and Q9 originated from New Mexico (USA) and Chile respectively. These genotypes were screened and multiplied by Alternate Crops Lab, Department of Agronomy, University of Agriculture, Faisalabad, Pakistan.

Nursery of both genotypes was grown in a wire house at the experimental station of University of Agriculture, Faisalabad, Pakistan (31.4180° N, 73.0790° E) with average temperature of 12.35°C and relative humidity ranging from 46% to 85%. Nursery was grown in plastic bags containing about 1.5 kg of an equal mixture of sand, soil (loam) and leaf compost. 15 to 20 seeds were sown per bag and plants were watered every second day. Transplanting was done to hydroponic culture after 22 days at two to four-leaf stage.

Hydroponic Culture

Seedlings were transplanted into plastic tubs containing growth medium of half strength Hoagland's solution. Seedlings were grown under normal hydroponic conditions to minimize transplantation shock for two days. Seedlings were raised using float hydroponic technique in which porous thermopore sheets were used to support plants and strips of thin (1.3 cm wide) foam were used to fix seedlings in the pores. Four tubs for each variety were used. Air pumps were used to aerate the roots of the plants and growth medium and salts were changed fortnightly. Temperature recorded during the period was 19±2°C and relative humidity was ranging from 35% to 79%.

Salt Treatment

Salt treatment was given two days after transplanting seedlings. To avoid osmotic shock, salt (NaCl) was applied in nutrient medium incrementally in phases of 100 mM NaCl per day. In all, there were four treatments with 3 replicates each: Control (0), 100, 200 and 300 mM NaCl which was equal to 0, 20, 40 and 60% seawater salinity (Eisa *et al.*, 2012).

Sampling

Four weeks after transplanting, five plants were chosen randomly for shoot length, root length, total fresh weight and total dry weight. Leaf and root samples for ionic analysis were also taken and reserved after drying. Leaf

samples of plants were harvested and immediately stored at -40°C for the quantitative biochemical analysis.

Chlorophyll Content Index (CCI) and Gaseous Exchange Parameters

CCI of the young fully expanded leaves was measured with chlorophyll index meter. Two weeks after imposition of salinity, three plants from each replication were chosen randomly to measure gaseous exchange parameters (net photosynthesis rate, stomatal conductance and transpiration rate) of fourth uppermost young fully expanded leaves. These parameters were recorded using an infrared gas analyzer LCA-4 (ADC, England) under different salinity levels at saturating radiation of 580 $\mu\text{mol photon m}^{-2} \text{s}^{-1}$.

Antioxidants

Free proline content of young fully expanded leaves was spectrophotometrically (UV 4000, O.R.I., Germany) determined at 520 nm by the using the method described by Bates *et al.* (1973). Quantitative estimation of leaf ascorbic acid was done by using the method described by Kampfenkel *et al.* (1995). Carotenoids were calculated by the method and equation described by Arnon (1949).

Membrane Thermostability Index

Four uppermost fully expanded leaves of both genotypes were cut from each replication. Leaves were divided into two parts from each treatment to be used as control and heat treated. The leaves were placed in two test tubes with 10 mL deionized water and 15 mL water was added again. One test tube was placed at 45°C for 1 h in water bath while the other was kept at room temperature. Then conductivity readings were taken as the test tubes cooled down to room temperature using an EC meter for control and heat treated samples. Leaf membrane thermostability was estimated by using equations by Blum and Ebercon (1981).

Ionic Analysis

Third leaf and portion of primary roots were taken to analyze ionic contents in leaves and roots and flame photometry (Sherwood Model 360, Flame Photometer) was used to determine concentrations of Na^+ and K^+ (Yuri *et al.*, 2009). Selective absorption of K^+ vs Na^+ from the medium by roots (SA) and the selective transport of K^+ vs Na^+ from root to shoot (ST) was calculated by using the following equations (Debez *et al.*, 2010):

$$SA = \frac{[K/(K + Na)]_{\text{root}}}{[K/(K + Na)]_{\text{medium}}}$$

$$ST = \frac{[K/(K + Na)]_{\text{leaf}}}{[K/(K + Na)]_{\text{root}}}$$

Statistical Analysis

The experiment was conducted in Completely Randomized Design with factorial arrangement. Data collected on all parameters were analyzed statistically to compare means (Steel *et al.*, 1997) using the computer statistical program “Statistix 8.1”, while Tukey’s HSD (Honest Significant Difference) test at 5% level of probability was applied to distinguish significant treatments.

Results

Growth Parameters

Data presented in Table 1 depict that interactive effect of salinity levels and genotypes was highly significant ($p < 0.001$). Q9 plants exhibited maximum shoot length under nonsaline conditions but as the salinity level increased, shoot length in Q9 decreased and least shoot length was observed at 60% sea water salinity in genotype Q9. While in Q7 plants, there was no significant ($p > 0.05$) reduction in shoot length by the addition of NaCl up to 100 mM, however, as the salinity level increased over 100 mM, the shoot length decreased significantly ($p < 0.001$). It was observed that decrease in shoot length in Q7 was not as intense as in the plants of Q9.

Control plants produced maximum root length, however, significantly ($p < 0.001$) shorter roots were noted with increasing salt regimes in both genotypes, however, genotype Q9 produced more root length at control while genotype Q7 increased more root length than Q9 at 100 and 200 mM NaCl concentration.

Although Q9 plants gained more fresh weight at control, but a highly significant ($p < 0.001$) decline in total fresh weight was recorded in Q9 at increasing salinity level as compared to Q7, which also decreased with increased salinity level. Maximum dry weight was recorded from Q9 plants at control and then it decreased significantly ($p < 0.001$) at each salinity level while different trend was observed in Q7. Dry weights of Q7 plants were almost equal at control and 100 mM salinity level which then decreased significantly ($p < 0.05$) as the salinity level was increased to 200 and 300 mM.

Biochemical Parameters

Different trends of leaf chlorophyll content index were observed at different salinity levels in both the genotypes (shown in Table 2 and Fig. 1). Chlorophyll index in leaves was found to be increased significantly ($p < 0.001$) with increasing water salinity and maximum index was found at 200 mM NaCl level in both genotypes. No significant interaction ($p > 0.05$) in leaf ascorbic acid was found among the genotypes under increased salinity levels. Same trend was observed in the concentrations of free proline which was also found non-significant ($p > 0.05$). Concentration of carotenoids was also non-significant ($p > 0.05$) among the

Table 1: Influence of salt stress on the growth characteristics of quinoa genotypes

Parameters	Shoot Length (cm)		Root Length (cm)		Fresh weight (g)		Dry weight (g)	
	Q7	Q9	Q7	Q9	Q7	Q9	Q7	Q9
Control	28.94 ^b	36.72 ^a	31.64 ^a	24.86 ^b	8.731 ^{ab}	10.9 ^a	0.864 ^b	1.240 ^a
100 mM	28.82 ^b	27.14 ^b	24.16 ^b	21.14 ^{bc}	7.611 ^{bc}	5.833 ^{cd}	0.812 ^b	0.659 ^{bc}
200 mM	20.56 ^c	18.1 ^{cd}	20.48 ^{bc}	17.92 ^{cd}	5.569 ^{cd}	4.061 ^{de}	0.584 ^{bc}	0.477 ^{cd}
300 mM	15.48 ^{de}	12.9 ^{ce}	14.5 ^{de}	11.84 ^e	2.786 ^e	2.274 ^e	0.279 ^d	0.253 ^d

Values with identical letters vary non-significantly ($p > 0.05$)

Table 2: Comparison of biochemical parameters from leaves of the quinoa genotypes under nonsaline and saline conditions. (fwt. indicates Fresh Weight)

Parameters	Chlorophyll Index		Free proline ($\mu\text{mol g}^{-1}$ fwt.)		Ascorbic acid ($\mu\text{g g}^{-1}$ f.wt.)		Carotenoids ($\mu\text{g mL}^{-1}$ f.wt.)	
	Q7	Q9	Q7	Q9	Q7	Q9	Q7	Q9
Control	37.96 ^e	32.64 ^d	0.005	0.006	1.646	1.840	4.886	5.138
100 mM	48.86 ^{ab}	45.40 ^b	0.008	0.004	1.662	1.913	4.743	4.873
200 mM	52.28 ^a	49.78 ^a	0.006	0.004	1.921	1.947	5.153	5.105
300 mM	50.88 ^a	49.15 ^a	0.006	0.006	1.859	1.907	4.601	4.560

Values with identical letters vary non-significantly ($p > 0.05$)

treatments and genotypes (Table 2).

Maximum leaf membrane thermostability was observed in control of Q7 plants (Fig. 2). Membrane thermal stability index slightly increased at 100 mM in leaves of Q7 plants, then it started decreasing significantly ($p < 0.001$) as the salinity level increased. While in Q9 plants, the membrane thermostability index significantly decreased ($p < 0.05$) at 100 mM NaCl and remained almost same till salinity at 200 mM and then it declined at 300 mM concentration. Overall membrane stability in Q7 leaves was significantly ($p < 0.05$) greater than Q9 up to 100 mM then it decreased consistently in both the genotypes.

Gaseous Exchange Parameters

Highly significant ($p < 0.001$) decrease in photosynthetic rates was recorded for every increase in salinity level in both genotypes. Same trend was observed in stomatal conductance which was also found to be significantly ($p < 0.001$) decreased in both genotypes at increasing NaCl concentrations (Fig. 3a).

Another parameter was transpiration rate of plants, in which maximum transpiration rate was observed in non-saline conditions in genotype Q9 and then it decreased significantly ($p < 0.05$) at each level. At 100 mM, transpiration rate in Q9 was recorded to be significantly ($p < 0.05$) higher than Q7 which then decreased at higher salinity levels (Fig. 3b). As for the genotype Q7, the transpiration rate increased non-significantly at salinity levels up till 200 mM and then, it decreased significantly at 300 mM.

Ionic Analysis

K^+ over Na^+ absorption ratio (SA) of roots significantly

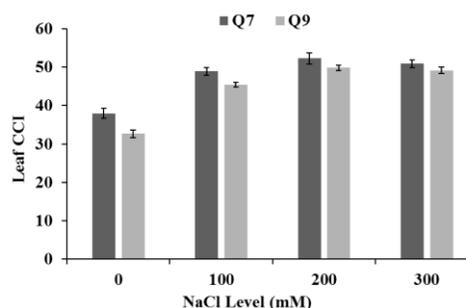


Fig. 1: Effect of increasing salinity level on leaf chlorophyll content index of quinoa genotypes

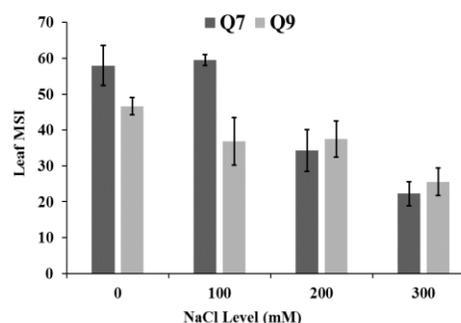


Fig. 2: Effect of salinity on leaf membrane stability index (MSI) of two quinoa genotypes

($p < 0.001$) increased from the medium in both genotypes as the salinity level increased (Fig. 4). However, Q7 plants showed higher absorption of K^+ as compared to Q9 and maximum absorption of Q7 plants was recorded at 60% sea water salinity. Similar linear increase in SA was also recorded in Q9 plants as well.

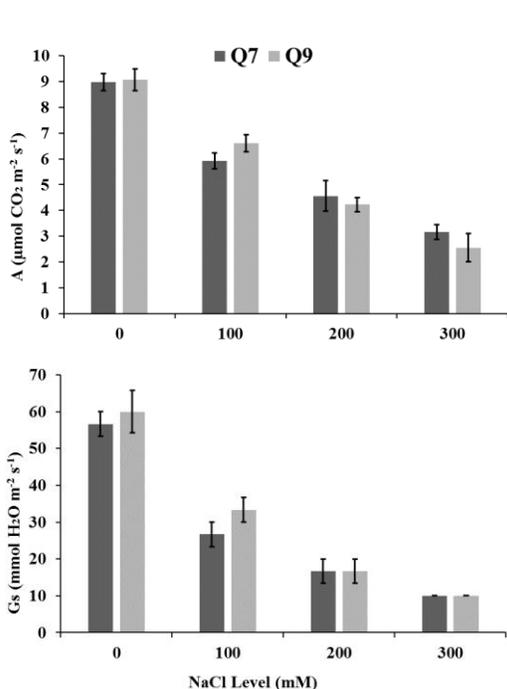


Fig. 3a: Effect of increasing salinity level on photosynthetic rate (A) and stomatal conductance (Gs) of quinoa genotypes

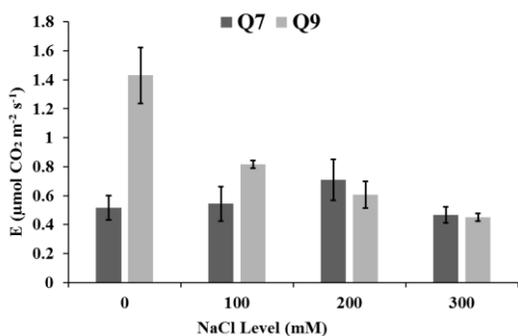


Fig. 3b: Effect of increasing salinity level on transpiration rate of quinoa genotypes

Different trends among genotypes were observed for selective transport ratio (ST) of K^+ vs Na^+ from root to leaves. In genotype Q7, as the salinity level increased, the selective transport of K^+ decreased significantly ($p < 0.001$) from root to leaves while in genotype Q9, the ratio increased at increasing salinity levels and maximum transport of K^+ was observed at 200 mM concentration in Q9 (Fig. 4).

In leaves of Q7 K^+ decreased significantly ($p < 0.001$) at all salinity levels as compared to control. The decrease was linear until 200 mM and then at 300 mM, the K^+ concentration in Q7 plants increased and was significantly ($p < 0.001$) more than the concentration at 200 mM (Fig. 5). As for genotype Q9, the potassium concentration in leaves increased with increasing salinity up to 200 mM which

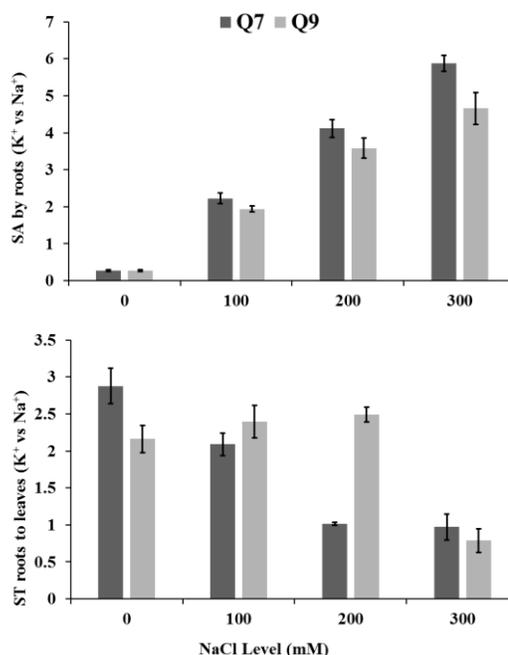


Fig. 4: Selective Absorption (SA) of K^+ vs Na^+ by the roots of quinoa from the medium and Selective Transport (ST) of K^+ vs Na^+ from roots to leaves

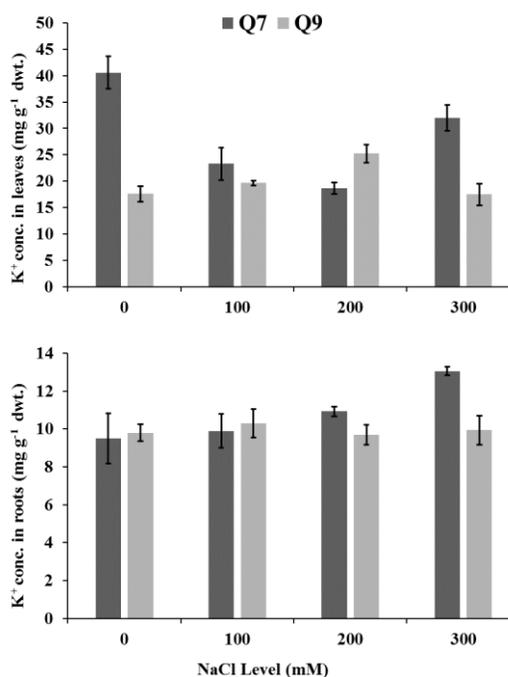


Fig. 5: Effect of increasing salinity level on K^+ concentration in leaf and root of quinoa genotypes

decreased significantly ($p < 0.001$) at 300 mM. In roots, the behavior was contrary to that noted in leaves. As the salinity level increased to 300 mM, there was a significant ($p < 0.05$) rise in K^+ concentration in roots of Q7 plants. However, in

Q9 plants, all the salt treatments were statistically similar ($p > 0.05$) to control (Fig. 5).

Na^+ concentrations in leaves increased significantly ($p < 0.001$) in Q7 plants with increasing salinity levels and the concentration of Na^+ in Q7 plants was significantly ($p < 0.05$) higher than Q9 plant. Maximum Na^+ concentration in leaves was recorded in Q7 plants at 300 mM NaCl concentration. As for the Na^+ concentration in roots, there was no significant ($p > 0.05$) change among genotypes and the concentration increased significantly ($p < 0.001$) with increasing salinity and maximum sodium concentration in roots was found at 300 mM in both genotypes (Interactions of Na^+ concentrations in leaves and roots can be observed in Fig 6).

Discussion

Growth of quinoa genotype Q7 was unaffected by the NaCl up to the 100 mM except for root length which declined at increasing NaCl concentrations. Soil salinity may induce drought stress due to an increase in extracellular solute concentration which may cause a decline in water potential resulting in loss of cellular turgor (Taiz *et al.*, 2015). So, the decline in growth parameters especially fresh and dry weights of the plants under increasing salinity levels is linked to a rise in osmotic potential in water stressed plants (Turner, 1981) which may be due to high extra cellular solute concentration or decreased volumes of cell under drought. Jacobsen *et al.* (2009) reported that minute thick walled cells of quinoa plants acclimated to water losses under drought do not lose turgor even under severe water limitations. Plant height in quinoa is one of the most sensitive characteristics under salinity (Jacobsen *et al.*, 2001). In our study, the variation in height of both genotypes in response to salinity was distinct. Q7 plants maintained height upto 100 mM NaCl level then onwards, there was a continuous reduction in shoot length, while in Q9, the plant height declined at all saline treatments.

Genotype Q7 maintained plant growth up to 100 mM NaCl concentration and then salinity affected its growth at further levels, which may be due to capability of quinoa plants to maintain water status even under saline environment while Q9 plants showed sensitivity even at 100 mM salinity. This decrease in growth may be linked with photosynthesis rate and stomatal conductance, which have been found to reduce under high salinity stress (Razzaghi *et al.*, 2011). Similar situation was noted in our study except for transpiration rate in Q7 plants in which it increased at 200 mM salinity then it declined at further salinity level. A severe decline in photosynthesis activity was correlated with a significant reduction in stomatal conductance and high levels of Na^+ accumulation in leaf tissues, which also strongly decrease photosynthetic capacity of plants. Decrease in photosynthetic capacity might be due to the decreased activity of photosynthetically active enzymes like RubisCO, which decreases under salinity (Rivelli *et*

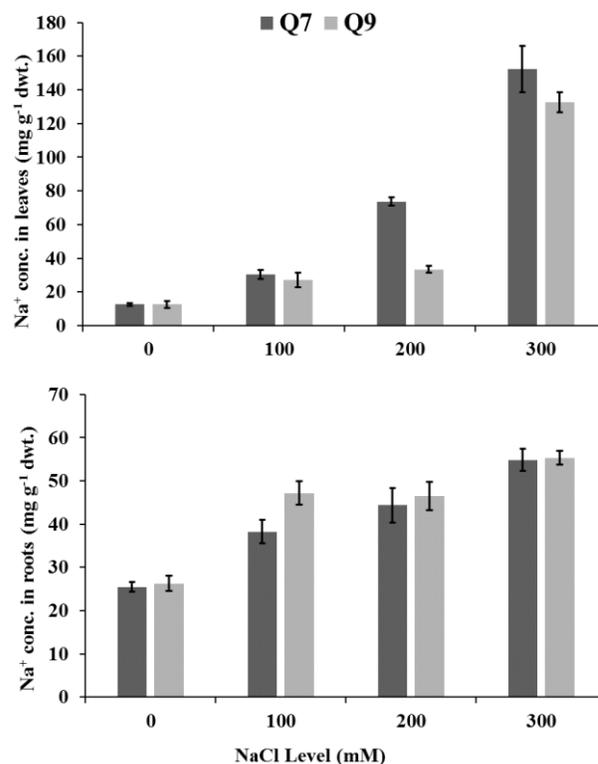


Fig. 6: Increasing concentrations of Na^+ in leaves and roots

et al., 2002). As salinity is the main reason for drop in gas exchange, it is also responsible for the decline in transpiration of quinoa plants (Sanchez *et al.*, 2003). Although, transpiration rates were found to be decreased in Q9 plants with increasing salinity, similar rates were observed in Q7 plants up to 200 mM salinity level. This behavior shows the tolerance of quinoa to highly saline environments by acquiring salt ions and regulating leaf osmotic potential, thus sustaining turgor and transpiration (Jacobsen *et al.*, 2003).

Antioxidants i.e., free proline, ascorbic acid and carotenoids were observed in leaves at all concentrations of NaCl. There was no significant difference from control in the concentrations of ascorbic acid and carotenoids under salinity. Although, free proline concentration increased at 100 mM, it was found to be decreasing with increased salinity. Chen *et al.* (2007) reported K^+ to be the main contributor in cytoplasmic osmolality in salt tolerant barley while free proline and glycinebetaine compensated for reduced cytosolic K^+ levels. These organic osmolytes may be involved indirectly in osmotic adjustment by modifying K^+ movement across membranes thus limiting sodium induced efflux of this inorganic osmolyte (Cuin and Shabala, 2007). Current and previous observations of high K^+ levels in quinoa under stress together with inadequate free proline, ascorbic acid and carotenoids buildup corroborate with the concept. Thus, it can be said that inorganic ions like sodium and potassium are mainly

responsible for the osmotic adjustment in quinoa.

Salinity induced reduction in chlorophyll content has been observed in NaCl susceptible plants (Jamil *et al.*, 2007). However, in salinity tolerant plants, the chlorophyll contents have been found to increase under salt stress (Khan *et al.*, 2009). Hence, improved chlorophyll contents in both genotypes showed the ability of quinoa to thrive under severe salt stress.

Plant growth and CO₂ exchange rate may be reduced by several factors but salinity induced ion exchange is debatably most important among these factors (Adolf *et al.*, 2012). Competition is found between K⁺ and Na⁺ due to similarities in their physiochemical properties and sodium is known to compete with potassium for most of the binding sites (Kronzucker, 2013). Hence, a sharply reduced leaf K⁺: Na⁺ ratio by an accumulation of high levels of Na⁺ was observed in both genotypes. Loss of K⁺ from leaves under salinity may cause programmed cell death by activating enzymes like proteases (Shabala, 2009) thus increasing drop of older leaves (data not given).

Na⁺ increased significantly with increasing salinity in the leaves of Q7 plants, while the abrupt rise of Na⁺ concentration was found only in 300 mM concentration. Same trend was observed in genotype Q9 as well. As for the K⁺ content in leaves, it decreased with increasing salinity levels and at 300 mM, there was a significant improvement in its contents in genotype Q7. Orsini *et al.* (2011) reported that K⁺ concentrations declined at levels starting from 150 mM and increased at higher salinity levels. In the genotype Q9, K concentration increased up till 200 mM and then decreased at 300 mM. The increase in Na⁺ concentration may be due to fact that quinoa plants accumulate Na⁺ which is readily available for cytosolic osmotic adjustment and sustaining turgor pressure, so to manage a suitable K⁺: Na⁺ concentration in leaves, the increased Na⁺ uptake should be followed by enhanced K⁺ transport from root to shoot (Cuin *et al.*, 2009). Potassium is believed to activate more than 50 enzymes including RUBISCO and those involved with chlorophyll biosynthesis (Shabala, 2003). Therefore, the specific roles of K⁺ cannot be replaced by Na⁺. It was found from our study that the selective absorption of K⁺ over Na⁺ from medium was enhanced in both genotypes as NaCl level increased while its transport to leaves by roots decreased considerably in Q7 and slightly increased in Q9 reaching maximum at 200 mM. The decrease in transport in Q7 may be due to Na⁺ acting as an osmolyte.

Conclusion

Taking all the parameters into consideration, 100 mM salinity level may be regarded as an optimum level of salinity as the plants in both genotypes were least affected at this level. Furthermore, Q7 is more salt tolerant than Q9 as most of the parameters (especially growth) were scarcely or not affected with the application of 100 mM NaCl, thus

generally confirming the halophytic behavior of quinoa. Genotype Q9 produced best results under nonsaline conditions so it can be used for arable crop production. However, since Q7 was least affected by moderate salt stress, it can be regarded better genotype in moderately saline soils.

References

- Adolf, V.I., S. Shabala, M.N. Andersen, F. Razzaghi and S.E. Jacobsen, 2012. Varietal differences of quinoa's tolerance to saline conditions. *Plant Soil*, 357: 117–129
- Arnon, D.T., 1949. Copper enzyme in isolated chloroplasts polyphenol oxidase in *Beta vulgaris*. *Plant Physiol.*, 24: 1–15
- Bates, L.S., R.P. Waldren and I.D. Teare, 1973. Rapid determination of free proline for water stress studies. *Plant Soil*, 39: 205–207
- Blum, A. and A. Ebercon, 1981. Cell membrane stability as a measure of drought and heat tolerance in wheat. *Crop Sci.*, 21: 43–47
- Chen, Z., T.A. Cuin, M. Zhou, A. Twomey, B.P. Naidu and S. Shabala, 2007. Compatible solute accumulation and stress-mitigating effects in barley genotypes contrasting in their salt tolerance. *J. Exp. Bot.*, 58: 4245–4255
- Cuin, T.A. and S. Shabala, 2007. Potassium efflux channels mediate Arabidopsis root responses to reactive oxygen species and the mitigating effect of compatible solutes. *Plant Cell Environ.*, 30: 875–885
- Cuin, T.A., Y. Tian, S.A. Betts, R. Chalmandrier and S. Shabala, 2009. Ionic relations and osmotic adjustment in durum and bread wheat under saline conditions. *Funct. Plant Biol.*, 36: 1110–1119
- Debez, A., D. Saadaoul, I. Slama, B. Huchzermeyer and C. Abdely, 2010. Responses of *Batis Maritima* plants challenged with upto two-fold seawater NaCl salinity. *J. Plant Nutr. Soil Sci.*, 173: 291–299
- Eisa, S., S. Hussin, N. Geissler and H.W. Koyro, 2012. Effect of NaCl salinity on water relations, photosynthesis and chemical composition of quinoa (*Chenopodium quinoa* Willd.) as a potential cash crop halophyte. *Aust. J. Crop Sci.*, 6: 357–368
- FAO and ITPS, 2015. *Status of the World's Soil Resources (SWSR) – Main Report*. Food and Agriculture Organization of the United Nations and Intergovernmental Technical Panel on Soils, Rome, Italy
- FAO, 1998. *Under-utilized Andean Food Crops*. FAO, Rome, Italy
- Flowers, T.J. and T.D. Colmer, 2008. Salinity tolerance in halophytes. *New Phytol.*, 179: 945–963
- Flowers, T.T., R.F. Troke and A.R. Yeo, 1977. The mechanism of salt tolerance in halophytes. *Annu. Rev. Plant Physiol.*, 28: 89–91
- Glenn, E.P., J.J. Brown and E. Blumwald, 1999. Salt tolerance and crop potential of halophytes. *Crit. Rev. Plant Sci.*, 18: 227–255
- Greenway, H. and R. Munns, 1980. Mechanisms of salt tolerance in non-halophytes. *Annu. Rev. Plant Physiol.*, 31: 149–190
- Guo, S.L., H.B. Yin, X. Zhang, F.Y. Zhao, P.H. Li, S.H. Chen, Y.X. Zhao and H. Zhang, 2006. Molecular cloning and characterization of a vacuolar H⁺ pyrophosphatase gene, SSVP, from the halophyte *Suaeda salsa* and its overexpression increases salt and drought tolerance of arabidopsis. *Plant Mol. Biol.*, 60: 41–50
- Hariadi, Y., K. Marandon, Y. Tian, S.E. Jacobsen and S. Shabala, 2011. Ionic and osmotic relations in quinoa (*Chenopodium quinoa* Willd.) plant grown at various salinity levels. *J. Exp. Bot.*, 62: 185–193
- Heuer, B., 2005. Photosynthetic carbon metabolism of crops under salt stress. In: *Handbook of Photosynthesis*, pp: 779–792. M. Pessaraki (ed.). Taylor & Francis Group, Boca Raton, Florida, USA
- Jacobsen, S.E., A. Mujica and C. Jensen, 2003. The resistance of quinoa (*Chenopodium quinoa* Willd.) to adverse abiotic factors. *Food Rev. Int.*, 19: 99–109
- Jacobsen, S.E., F. Liu and C.R. Jensen, 2009. Does root-sourced ABA play a role for regulation of stomata under drought in quinoa (*Chenopodium quinoa* Willd.). *Sci. Hort.*, 122: 281–287
- Jacobsen, S.E., H. Quispe and A. Mujica, 2001. *Quinoa: An Alternative Crop for Saline Soils in the Andes, Scientists and Farmer-partners in Research for the 21st Century*, pp: 403–408. CIP Program Report 1999–2000

- Jamil, M., S.U. Rehman, K.J. Lee, J.M. Kim, H.S. Kim and E.S. Rha, 2007. Salinity reduced growth PS2 photochemistry and chlorophyll content in radish. *Sci. Agric.*, 64: 111–118
- Kazmi, S.I., M.W. Ertsen and M.R. Asi, 2012. The impact of conjunctive use of canal and tube well water in Lagar irrigated area, Pakistan. *Phys. Chem. Earth*, 47: 86–98
- Kampfenkel, K., M.V. Montagu and D. Inze, 1995. Extraction and determination of ascorbate and dehydroascorbate from plant tissues. *Anal. Biochem.*, 225: 165–167
- Khan, M.A., M.U. Shirazi, M.A. Khan, S.M. Mujtaba, E. Islam, S. Mumtaz, A. Shereen, R.U. Ansari and M.Y. Ashraf, 2009. Role of proline, K/Na ratio and chlorophyll content in salt tolerance of wheat (*Triticum aestivum* L.). *Pak. J. Bot.*, 41: 633–638
- Koyro, H.W. and B. Huchzermeyer, 1999. Salt and drought stress effects on metabolic regulation in maize. In: *Handbook of Plant and Crop Stress*, 2nd edition, pp: 843–878. Pessarakki, M. (ed.). Marcel Dekker, New York, USA
- Koyro, H.W., N. Geißler, S. Hussin and B. Huchzermeyer, 2008. Survival at extreme locations: life strategies of halophytes – the long way from system ecology, whole plant physiology, cell biochemistry and molecular aspects back to sustainable utilization at field sites. In: *Biosaline Agriculture and High Salinity Tolerance*, pp: 1–20. Abdelly, C., M. Öztürk, M. Ashraf and C. Grignon (eds.). Birkhäuser Verlag, Switzerland
- Krebs, M., D. Beyhl, E. Görlich, K.A. Al-Raschid, Y.D. Stierhof, R. Hedrich and K. Schumacher, 2010. Arabidopsis V-ATPase activity at the tonoplast is required for efficient nutrient storage but not for sodium accumulation. *Proc. Natl. Acad. Sci.*, 102: 3251–3256
- Kronzucker, H.J., D. Coskun, L.M. Schulze, J.R. Wong and D.T. Britto, 2013. Sodium as nutrient and toxicant. *Plant Soil*, 369: 1–23
- Martinez-Beltran, J. and C.L. Manzur, 2005. *Overview of Salinity Problems in the World and FAO Strategies to Address the Problem*, pp: 311–313. Proceedings of the international salinity forum, Riverside, California, USA
- Munns, R. and M. Tester, 2008. Mechanisms of salinity tolerance. *Annu. Rev. Plant Biol.*, 59: 651–681
- Orsini, F., M. Accorsi, G. Gianquinto, G. Dinelli, F. Antognoni, K.B.R. Carrasco, E.A. Martinez, M. Alnayef, I. Marotti, S. Bosi and S. Biondi, 2011. Beyond the ionic and osmotic response to salinity in *Chenopodium Quinoa*: functional elements of successful halophytism. *Funct. Plant Biol.*, 38: 818–831
- Panta, S., T. Flowers, P. Lane, R. Doyle, G. Haros and S. Shabala, 2014. Halophyte agriculture: success stories. *Env. Exp. Bot.*, 107: 71–83
- Pearsall, D., 1992. The origins of plant cultivation in South America. In: *The Origins of Agriculture. An International Perspective*, pp: 173–205. Cowan, C.W. and P.J. Watson (eds.). Smithsonian Institution Press, Washington, London, UK
- Razzaghi, F., S.H. Ahmadi, V.I. Adolf, C.R. Jensen, S.E. Jacobsen and M.N. Andersen, 2011. Water relations and transpiration of quinoa (*Chenopodium quinoa* Willd.) under salinity and soil drying. *J. Agron. Crop Sci.*, 197: 348–360
- Rivelli, A.R., S. Lovelli and M. Perniola, 2002. effect of salinity on gas exchange, water relations and growth of sunflower (*Helianthus annuus*). *Funct. Plant Biol.*, 29: 1405–1415
- Repo-Carrasco, R., C. Espinoza and S.E. Jacobsen, 2003. Nutritional value and use of the andean crops quinoa (*Chenopodium Quinoa*) and ka'niwa (*Chenopodium Pallidicaule*). *Food Rev. Int.*, 19: 179–189
- Ruffino, A.M.C., M. Rosa, M. Hilal, J.A. González and F.E. Prado, 2010. The role of cotyledon metabolism in the establishment of quinoa (*Chenopodium quinoa*) seedlings growing under salinity. *Plant Soil*, 326: 213–224
- Sanchez, H.B., R. Lemeur, P.V. Damme and S.E. Jacobsen, 2003. Ecophysiological analysis of drought and salinity stress in quinoa (*Chenopodium quinoa* Willd.). *Food Rev. Int.*, 19: 111–119
- Shabala, S., 2003. Regulation of potassium transport in leaves: from molecular to tissue level. *Ann. Bot.*, 92: 627–634
- Shabala, S., 2009. Salinity and programmed cell death: unravelling mechanisms for ion specific signalling. *J. Exp. Bot.*, 60: 709–711
- Shabala, S. and A. Mackay, 2011. Ion transport in halophytes. *Adv. Bot. Res.*, 57: 151–199
- Shabala, S. and L. Shabala, 2011. Ion transport and osmotic adjustment in plants and bacteria. *Biomol. Concepts*, 2: 407–419
- Shabala, S. and T.A. Cuin, 2008. potassium transport and plant salt tolerance. *Physiol. Plant.*, 133: 651–669
- Shahid, S.A., 2013. Developments in Soil Salinity Assessment, Modeling, Mapping and Monitoring from Regional to Submicroscopic Scales. In: *Developments in Soil Salinity Assessment and Reclamation*, pp: 3–43. Springer, New York, USA
- Steel, R.C.D., J.H. Torrie and D.A. Deeky, 1996. *Principles and Procedures of Statistics a Biometric Approach*, 3rd edition, pp: 400–428. McGraw Hill Book Co. New York, USA
- Szabados, L. and A. Savouré, 2010: Proline: a multifunctional amino acid. *Trends Plant. Sci.*, 15: 89–97
- Taiz, L., E. Zeiger, I.M. Moller and A. Murphy, 2015. *Plant Physiology and Development*, 6th edition. Sinauer Associates Publishers, Sunderland, Massachusetts, USA
- Turner, N.C., 1981. Techniques and experimental approaches for the measurement of plant water status. *Plant Soil*, 58: 339–366
- Yilmaz, K., I.E. Akinci and S. Akincim, 2004. Effect of salt stress on growth and Na, K contents of pepper (*Capsicum annum* L.) in germination and seedling stages. *Pak. J. Biol. Sci.*, 7: 606–610
- Yuri, S., P. Langridge and M. Tester, 2009. Salinity tolerance and sodium exclusion in genus *Triticum*. *Breed. Sci.*, 59: 671–678

(Received 26 November 2016; Accepted 17 December 2016)