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The influence of salt stress on growth and biomass production of *Populus deltoides*

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Abstract

Salinity is one of the major abiotic stress in all over the world they are increased day by day due to the insufficient agriculture practices. Pakistan is mostly arid to semiarid country. Out of 23 million ha are cultivated lands, 6.8 million ha cultivated area are mostly affected by soil and water salinity. *Populus deltoides*, is the eastern cottonwood native to North America, growing throughout the eastern, central, and southwestern USA. *P. deltoides* is a tree that growing up to15–50m tall. The study was conducted to explore the salinity tolerance of forest species Eastern cottonwood (*Populus deltoids*) at Department of Forestry & Range Management, University of Agriculture, Faisalabad. Plant species was exposed to different level of salinity stress by using commercial salt NaCl (EC control, 2, 6 and 12 dSm^1) in earthen pots experiment to determine the different growth parameters. Results show that salinity have negative effect on biomass production but in EC $6dSm^1$ the growth parameters were significantly different from control treatment. However in EC $12dSm^1$ showed significantly decreased as compare to others treatments. Therefore, it is concluded that *Populus deltoides* well performed under the EC $6dm^1$.

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Introduction

Salt stress is an abiotic stress which affected more than 45 million hectares of irrigated land and they are important element to reduce the crop production around the worldwide (Hasanuzzaman *et al.*, 2014). Du to some economic influence associate with final product loss around the globe, different types of genetic and molecular techniques has been introduces to obtain the salinity tolerant species (Harfouche *et al.*, 2014; Osakabe *et al.*, 2012). Many studies has been reported that different degree of salinity tolerance in many forest plants including the poplar (Beritognolo *et al.*, 2011; Sixto *et al.*, 2005) provide the possible way for future cultivation of marginal lands which affected by salinity. Plant survival and efficiency are mostly affected by salinity through survival methods, mostly linked to osmotic stress (Munns and Tester 2008), nutritional disorder (Munns 2002) and ion toxicity (Tester and Davenport 2003). The mostly observed initial stage of salinity stress on plant is osmotic effect and it is reduce the water potential in the medium, reduce the capacity of water potential by the roots (Munns 2002). When this effect will be remove the high concentration of salts in the soil mostly affect the absorption rate of the important nutrients, affecting the nutritional disorders (Munns and Tester 2008). At the end high salt concentration in the plant its causes toxic, resulting in the restriction of many physiological and other biochemical processes (Chinnusamy *et al.*, 2005). Amongst these, the photosynthetic activity is alarm in various cases, mostly due to the largely changes at stomatal level. This details has been highlight by (Rajput *et al.*, 2015). The reductions on growth are generally attributed to low photosynthetic rates due both stomatal and non-stomatal limitations. *Populus* is a fast growing tress that containing number of species, it is widely distributed throughout the different parts of Asia and Europe and play very important role in economic and environment in different parts of the world (Zheng 1983). Some biomass production and nutrients cycling studies are present for fast growing tree species like eucalyptus, pine and poplars which belongs to acidic and neutrals soils (Baker and Attiwill, 1985; Wittwer and Stringer, 1985). Studies on this plant has been shown over many years reading their physiological, ecological with photosynthesis and respiration activities (Chen *et al.*, 2006; Wang *et al.*, 2007). *P. deltoides* is a model plant for research purpose because they have lot of features for example they have capability to grow very fast and it is possible to produce plant material for research purpose in a very short time as compare to the other tree species, *P. deltoides* have wide distribution of genus and they consists of at least 35 species (Rajagopal *et al.*, 2000) they have ability to adopted different degree of stress and adaptation to different habitats. *P.deltoides* can be easily grown on the land which is not suitable for agricultural purpose and increase carbon sequestrations, thus reduce the competition between the food and fuel purpose and also decreasing the carbon debt suffered through land use changes (Sannigrahi *et al.*, 2010). *P.deltoides* also provide a valuable raw material for plywood, paper industry, furniture industry, fuel wood, sports goods, fine paper, fiber board, match industry and packing paper they make extra income for the local people. The aim of this study was to identify the *P. deltoides* species performance under different level of salinity stress. We also analyzed the growth, biomass production and partitioning of *P. deltoides* under different saline environment condition.

Materials and method

Experiment Design

Experiment was conducted to check the influence of salt stress on growth parameters of forest species (*Populous deltoids*) at Department of Forestry and Range Management, (31°26N, 73°06E; 184.4m), University of Agriculture Faisalabad (UAF) Pakistan. Earthen pots were filled with soil mixture composed of (peat/sand, 2v/1v).

Salinity stress

Artificial salinity was developed by using commercial salt (NaCl). Three level of salinity were developed for this experiment (2, 6, 12 dSm^1). Each pot have 5kg soil and salinity developed by soil weight basis. Soil salinity was measured by suing the EC meter. In control condition the EC level is $2dSm^1$ and no addition salt was added in this pots. For obtained medium $6dSm^1$ and high saline stress $12dSm^1$

respectively adding suitable amount of commercial sodium chloride on soil weight basis. By adding 4.095g of NaCl in 5kg of soil to developed 6EC and 10.235g of NaCl to developed 12 EC.

Growth measurements

Six months old cutting were selected for the salinity stress. The plant were harvested after 50 days of salinity (NaCl) stress. The following growth parameters were measured Plant height (cm), stem diameter (mm) by suing vernier caliper, number of leaved and branches twice a week by manually. At the end of experiment plants was harvest and divided into different parts (leaf, stem and root) for calculating the leaf, stem and root fresh weight all the plants parts were weighted immediately after harvesting by using digital weight balance after calculating the fresh weight the leaf, stem and root different parts of plants were kept in incubators at 70°C for 24h and cooled it in natural condition after cooling dry weight were measured by suing the digital weight balance. Chlorophyll meter was used for measuring the chlorophyll contents when the experiment was completed.

Biomass partitioning

Biomass partitioning was calculated different part of plant e.g. leaves biomass allocation percentage, stem biomass allocation percentage and root biomass allocation percentage by using this formula.

$$Biomass\ allocation\ partitioning\ (BAP\ \%) = \frac{Dry\ mass\ (DM)}{Total\ biomass\ (TB)} X\ 100$$

Data Analysis

The data collected during the trial was analyzed using analysis of variance technique. (ANOVA) by using the statistical software SPSS. All tests, co relations was taken as significant at ($P < 0.05$) (Steel *et al.*, 1997).

Results and discussion

The Plant height of *P. deltoides* show that it was decreased significantly under different level of salt stress (Table 1). The heights percentage reduction in plant height was observed in plant which were grown in salinity stress environment 12 dSm^1 NaCl followed by 2 and $6dSm^1$. The minimum reduction that was observed in control condition (22.13), under medium stress (16.44) and high stress was (11.88). The maximum reduction was obtained (46.31%) under high saline stress as comparison to control treatment and the minimum was observed (25.71%) under medium saline stress as compare to control. Stem diameter was remains same under control (15.22) and medium saline stress (15.11) but also decrease in high saline stress (9.67). Chlorophyll contents, number of branches and number of leaves were also decreased when salinity stress increase. The maximum reduction in all growth parameters was observed under high saline stress $12dSm^1$ as compare to other salinity levels. Salinity have negative effect on plant growth. They decrease the growth parameters under medium and high saline stress. Chlorophyll contents was decreased in different level of salinity stress. A decrease in chlorophyll concentration in glycophytes due to salt stress has been reported earlier by several workers (Chavan and Karadge, 1986).

Salinity reduce plant growth for two reasons: first, deficit of water and second due to salt-specific or ion-excess effects (Munns *et al.*, 2006). Various types of mechanism developed by different plant species to cope with these effects (Munns, 2002). In this experiment, decreased in plant height, number of leaves and number of branches were observed (Table 1). Similar results was obtained the previous studies of Neves *et al.*, (2004) umbu plant stem diameter was less affected by salt stress as compare to the plant height and number of leaves. These results are reliable with other studies show that osmotically stressful environments reduce plant growth (Stanton *et al.*, 2000). Decreasing the growth of plant height of woody species due to increasing the level of salinity was extensively reported (Game *et al.*, 2007; Ramoliya and Pandey 2003; Mehari *et al.*, 2005). In our 4-week stress experiment stem diameter of *P.deltoides* was not decreased significantly under salt stress until 6 $dS\ m^1$ NaCl and number of branches was also less affected under 6 dSm^1 however cutting was not well perform in $12dSm^1$ NaCl (Table 1). Fung *et al.*, (1998) stated that mortality of cuttings of *P. popularis*, *P. robusta*, and *P. berolinensis* was evident in 1% (170mm) sodium chloride two weeks after applying of salinity stress.

Table 1. Growth Parameters under various level of Salinity.

Growth Parameters	*P. Deltoids* Control	*P. Deltoid* M. Stress	*P. Deltoid* H. Stress	P-Values
Plant height (cm)	22.13 (0.33) a	16.44 (0.29) b	11.88 (0.18) c	P<0.001
Stem Diameter (mm)	15.22 (1.13) a	15.11 (1.01) a	9.67 (0.32) b	P<0.001
No. of leaves	55.74 (0.98) a	49.01 (0.78) b	34.70 (0.45) c	P<0.001
Chlorophyll contents	27.56 (0.33)a	22.55 (0.23) b	19.31 (0.15) c	P<0.001
No. of Branches	7.23 (0.27) a	6.33 (0.20) b	4.86 (0.19) c	P<0.001

Significant level p<0.05 High Significant p<0.001.

Stem fresh weight of *P.deltoides* was remains same under control and medium saline stress environment (14.98g), (14.45g) but under high saline stress minimum values was obtained (11.78g) (Table 2). Leaf fresh weight was minimum under high saline. Leaf fresh weight was minimum under high salinity stress (14.22g) and maximum under control condition (18.89g) and medium saline condition was (15.37g). Minimum reduction was observed under medium saline condition but the root fresh weight was increased under medium saline condition (12.46g) and high saline condition (12.98g) as compare to the control condition (11.67g).

So that the root fresh weight was increased under different level of saline environment. Leaf and stem moisture contents percentage was decreased under both condition of salinity stress but the root moisture contents percentage was increased under medium (52.76%) and high saline stress (56.73%) as compare to the control condition (50.22%) but the response of root shoot ratio was also decreased under medium saline stress 6dSm^{1} and under high saline stress 12dSm^{1} when compare with control condition 2dSm^{1} (Table 2).

In commonly, salt gathering in the root area that reasons the development of osmotic stress and disrupts cell ion homeostasis by restriction of the uptake of essential nutrients such as K^+, Ca^{2+}, and NO_3^-, with accumulation of Na^+ and Cl^- to potentially toxic levels within cells (Chartzoulakis 2005). All of these initial stresses cause hormonal imbalance (Munns 2002). In our experiment the Biomass production was increased under different level of salinity stress especially at 6 dSm^{1}. These results are similar to the findings of Suwalka and Qureshi (1995), on the biomass growth of *Eucalyptus* and *D. sissoo* on sodic soils. Kurban *et al.*, (1999) stated that total plant dry weight increase at low salt stress (50mm) but decrease at high concentration of salt stress (100 and 200mm) in *Alhagi pseudalhagi* young seedlings. Fresh and dry weights of *Salicornia rubra*, a halophyte, increase with the increasing in salt stress, with optimum growth at 200mm sodium chloride, after which decrease in growth with additional of more salt was reported (Khan 2001). Our present study show that stem and root fresh weight were increased under medium 6dSm^{1} NaCl (Table 2) and root moisture contents was also increased in 12dSm^{1} NaCl.

Table 2. Biomass production under different levels of salt stress.

Biomass production	*P. Deltoids* Control	*P. Deltoid* M. Stress	*P. Deltoid* H. Stress	P- Values
Leaf fresh weight (g)	18.89 (1.23) a	15.37 (1.11) b	14.22 (0.67) c	P<0.001
Stem fresh weight (g)	14.98 (0.78) a	14. 45 (0.56) a	11.78 (0.34) b	P=0.064
Root fresh weight (g)	11.67 (1.45) a	12.46 (1.22) b	12.98 (1.13) b	P=0.081
Leaf moisture contents %	43.75 (0.89) a	41.56 (0.67) b	39.87 (0.22) c	P<0.001
Stem moisture contents %	45.77 (1.23) a	44.56 (0.91) b	40.13 (0.56) c	P<0.001
Root moisture contents %	50.22 (1.11) a	52.76 (0.88) ab	56.73 (0.44) abc	P=0.016
Root Shoot ratio	2.24 (0.45) a	2.19 (0.23) a	2.09 (0.22) ab	P<0.001

Salinity significantly influenced the leaf and stem dry weight of *P. deltoides* forest species (Table 3). The largest leaf dry weight was obtained in plants grow in 2dSm^{1} and minimum was noticed under high saline condition 12dSm^{1} however the leaf dry weight continued to decrease with increase the salt stress environment. Stem dry weight was maximum under control saline condition (8.67g) and decrease in medium saline stress (6.56g) and also same in high saline stress (6.22g).

The Root dry weight was not significantly effect by salinity at highest salt level 12dSm^{1} NaCl root dry weight was increase under medium saline 6dSm^{1} and high saline stress 12dSm^{1}. Total dry biomass of *P.deltoides* decreased significantly at the higher salinity 12dSm^{1} NaCl progressively decreased with increase salt concentration. The leaf biomass allocation percentage decrease in the presence of salinity stress. Maximum decrease was observed in high saline stress (32.67%) and minimum was noticed under medium saline environment (35.23%) and the highest was obtained under control condition (38.34%). Stem biomass percentage was also decrease when salinity stress increase but the root biomass allocation percentage was shown same results under control and medium saline environment. The roots are less effected by salinity stress so they maintain their biomass allocation percentage in different level of salt stress. The result of present study also shown the negative effect of salinity on plant biomass production leaf and stem dry weight also decrease with increasing the salt stress (Table 3) but the root dry weight was increase in medium 6dSm^{1} and high saline environment condition 12dSm^{1}. Our funding was similar to the previous studies of Mehari *et al.*, (2005) who found that dry weight of leaf and stem did not decrease two forest species *Acacia nilotica* and *A. tortilis* response to salinity stress and did not different between species. Willliams in (1960) studies that nearly three-fold increase in dry weight of Haloxylon glomeratus on added of 1mm sodium chloride to Hoagland and Arnon solution. For *Suadea maritima* addition of 10 and 17mm sodium chloride to the culture solution increase the dry weight to 126% and 140% respectively of the growth in culture solution alone (Flowers *et al.*, 1977). Our finding was similar with the previous study of Takemura *et al.*, (2000), where maximum growth of the plant was show at 125mm of sodium chloride and they also increase their dry weight in *Bruguiera gymnorrhiza* mahgrove species. The decrease in leaf, stem and total dry weight of *Eucalyptus* seedlings with increasing salt stress level are in consistence with many other previous results on the effect of salinity in dry matter production. (Ramoliya and Pandey 2002, Gebauer *et al.*, 2004; El-Juhany and Aref 2005). Funding of Mass and Hoffman (1977) show that plant biomass is not decrease until a threshold level of salt stress is reached. Du to this threshold, biomass reduce linearly with increasing level of salt stress.

Table 3. Biomass production and Allocation percentage.

Growth Parameters	*P. Deltoids* Control	*P. Deltoid* M. Stress	*P. Deltoid* H. Stress	P-Values
Leaf dry weight (g)	9.24 (0.67) a	8.45 (0.56) b	6.87 (0.34) c	P<0.001
Stem dry weight (g)	8.67 (1.22) a	6.56 (1.11) b	6.22 (1.01) b	P<0.001
Root Dry weight (g)	5.78 (0.89) a	6.78 (0.45) b	6.22 (0.34) b	P<0.001
Total Biomass (g)	14.33 (0.34) a	13.21 (0.23) b	11.34 (0.19) c	P<0.001
Leaf Biomass allocation %	38.34 (0.34)	35. 23 (0.24)	32.67 (0.19)	P= 0.191
Stem Biomass allocation %	33.46 (0.21)	32.29 (0.18)	30.33 (0.16)	P= 0.306
Root Biomass allocation %	26.87 (0.76) a	25. 96 (0.56) a	21.49 (0.44) b	P=0.014

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Conclusion

The present study was to identify the growth behavior and biomass production of *P.deltoides* under different level of salinity stress. In conclusion the ability of *P.deltoides* to survival under medium saline stress 6dSm^{1} of NaCl was better as compare to high saline condition 12dSm^{1}. They improve root growth in medium saline stress environment. In short term the high saline stress strongly influenced the different growth parameters. They decrease the plant height, number of leaves, number of branches and also decrease the chlorophyll contents. However under medium saline condition the root fresh and dry weight was increase as compare to the high saline stress. According to the results of this study we concluded that *P.deltoides* perform well under the medium saline soils and they also improve their biomass.

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