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Physiological evaluation of drought tolerance in Poplar (Populus deltoids L.) for different drought levels

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ABSTRACT

Experiments were conducted to evaluate drought tolerance under different levels of water stress in poplar trees. The cuttings of Populus deltoides L. (clone Kranti) were exposed to four different watering regimes (100, 75, 50 and 25% of the field capacity) and changes in physiological parameters related with drought tolerance were recorded. Drought treatments (75%, 50% and 25% FC) decreased net photosynthetic rate (Pn), transpiration rate (E), chlorophyll fluorescence (Fv/Fmax), plant height, number of leaves, specific leaf area (SLA), leaf area index (LAI) and total biomass content in all the three watering regimes compared to control (100% FC). Cuttings were showed poor performance with increasing levels of drought stress. Severity were observed in Pn, E, Fv/Fmax, plant height, stem diameter, leaf area and number of leaves, SLA, LAI and total biomass content with increasing levels of water stress. Decreased CO_2 assimilation and transpiration rate due to instantaneous closure of stomata to protect the plants against hazardous effects of water stress leads to overall decrease in biomass of cuttings with 60 days water stress treatments. By visualizing the results, we can say that Scarcity of water is a severe environmental constraint to plant productivity. Drought-induced loss in plant productivity, since both the severity and duration of the stress are critical. Secondly, we can emphasise with our experiment that poplar plants can maintain their better growth and biomass only up to 75-50% of FC after that stress shows its severity so much that the aim of plants is only to survive and biomass maintenance become vague.

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INTRODUCTION

Poplar (*Populus deltoides* L.) (clone Kranti) is one of the most important tree species of agroforestry and grown either in blocks or on the boundaries of fields in India. It is also known as American cotton wood is an economically important fast growing tree species, which belongs to family Salicaceae. It is soft wood tree, raised through stem cuttings and suitable for commercial use like manufacture of matches, furniture, packing cases, plywood, sport goods, pulp and paper, etc. It is most preferred by the farmers because of its fast growing, short duration and more compatibility with intercrops (Gupta *et al.*, 2014). Drought is one of the most important abiotic stress factors that limit plant growth and ecosystem production around the world (Xiao *et al.*, 2009). Water stress is an important environmental factor that affects photosynthesis, affecting plant growth and biomass (Akcay *et al.*, 2010). Drought stress reduces leaf size, stems extension and root proliferation, disturbs plant water relations and reduces water-use efficiency. Plants display a variety of physiological and biochemical responses at cellular and whole-organism levels towards prevailing drought stress, thus making it a complex phenomenon. CO_2 assimilation by leaves is reduced mainly by stomatal closure, membrane damage and disturbed activity of various enzymes, especially those of CO_2 (Farooq *et al.*, 2009).

Low production of wood per unit area and heavy deforestation resulted in increasing gap between demand and supply of wood. Fast growing and short rotation tree species are considered best for increasing wood productivity. The Eucalyptus and Poplar have the potential for narrowing down the gap between demand and supply of soft woods. These trees are highly suitable for agroforestry (either on bunds or for intercropping). There is currently considerable interest worldwide in the potential for trees on farms to increase income in a manner that maintains or enhances the diversity of species. Resource-poor farmers find poplar useful if it enhances the yield or quality of the understorey crops and or supplies products such as fodder, firewood and materials for house construction. Poplar is usually known as one of the most drought-sensitive woody plant groups, but its drought tolerance varies greatly among species, populations and clones due to their great genetic diversity (Monclus *et al.*, 2006).

For establishment of poplar tree the seedling stage (upto 60 days after transplanting of cuttings) is very important and is susceptible for water stress. Therefore, objective of this

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research paper is to study the effect of different levels of water stress (i.e. simulation of declining water availability due to changing rainfall pattern) on various growth and physiological characteristics of poplar plants and how they adjust their physiology to progressive drought stress conditions.

MATERIAL AND METHODS

Plant and growth conditions

The experiment was conducted at Department of Plant Physiology, G.B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand. Cuttings of Poplar clone Kranti, were raised by using uniform diameter and length (size 18-20cm) purchased from WIMCO seedlings (Pvt.) Ltd., Rudrapur, Udham Singh Nagar (Uttarakhand). The Poplar cuttings were planted in polybags for a period of 15 days. Sprouted cuttings were transplanted in earthen (diameter-30 cm and depth-30 cm) pots, filled with fertile soil, i.e. soil texture was silty clay loam, pH-7.1, organic carbon 0.86% and N, P, K- 245, 35.5 and 172 kg ha^{-1}. Twenty-five pots were kept under control conditions (100% of field capacity) and 25 pots each under different levels of drought stress (75, 50 and 25% of field capacity (FC)). Transplanted sprouted cuttings were left for another 15 days for initial establishment in earthen pots as a normal condition. Treatments were imposed after establishing sprouted cuttings. Drought levels were maintained in different group of plants by keeping soil moisture status at 75, 50 and 25% of field capacity as compare to control.

Thus, control seedlings were also similarly grown in the earthen pots, except regular irrigation was allowed up to field capacity. Drought levels were maintained on the basis of CPE (Cumulative Pan Evaporimeter) reading taken from agro meteorological section of Crop Research Centre (CRC), GB Pant University of Agriculture & Technology, Pantnagar (Uttarakhand). The cumulative pan evaporation (CPE) dependent drought induction method is standard method and regularly used in farm and pots for drought induction. To maintain different drought levels, the treated plants were irrigated with 2.5 L of water when the day for specific drought stage came, while alternate day to control plants. Water stress was imposed for a period of 60 days and physiological, growth and biomass responses of the plants were measured at specific time intervals.

Photosynthetic, chlorophyll status and chlorophyll florescence measurement

Photosynthetic measurements were done with a directly using CIRAS-1, IRGA, portable photosynthesis system (PP system, England), using the youngest completely expanded leaf of each plant under saturating natural sunlight between 8:00-10:00 h at photosynthetic photon flux density (PPFD) >1400-1800 $\mu mol\ m^{-2}s^{-1}$ to avoid high temperature and low humidity. A chlorophyll meter was used to estimate the relative chlorophyll status of crops. The instrument measures transmission of red light at 650 nm, at which chlorophyll absorbs light, and transmission of infrared light at 940 nm, at which no absorption occurs. On the basis of these two transmission values the instrument calculates a Soil Plant Analysis Development (SPAD) value that is quite well correlated with chlorophyll content. SPAD readings were recorded by a portable SPAD meter (Opti Science, CMM-200, USA) in sunlight for fully mature leaves of each plant to get a mean value. Fluorescence measurements were done with a Plant Efficiency Analyzer (Handy, PEA, Hansatech King Lynn, UK). Prior to each measurement, a clip was placed on the leaf for 30 min for dark adaptation. Photochemical efficiency of photosystem II (Fv/Fm) was calculated as: Fv/Fm = 1-Fo/ Fm, where Fv is the variable fluorescence (Singh *et al.*, 2013).

Growth Parameters

At the end of the experiment (60 days), plant height, radial stem diameter and number of leaves of the seedling were determined and then a destructive harvest was carried out. Ten seedlings of Kranti for control and treatments were randomly sampled. Leaf area was measured using a CI-203 Area Meter (CID, USA). Specific leaf area of the whole plant (SLA) (cm2 g-1) was calculated as per Beadle (1993). Leaf area index (LAI) (m^2/m^2) was calculated as (leaf area)/(land area). Dry weights were obtained by weighing the plant material after drying at 75 °C±3 unit, a constant mass reached. The harvest index was calculated by harvesting at least 10 seedlings of each treatment (n=10) as per Michael et al. (1988). Data management and statistical analysis were performed using SPSS software (SPSS, Chicago, IL, USA). Means were expressed and compared by ANOVA. All statistical tests were considered significant at $p \leqslant 0.05$.

RESULTS AND DISCUSSION

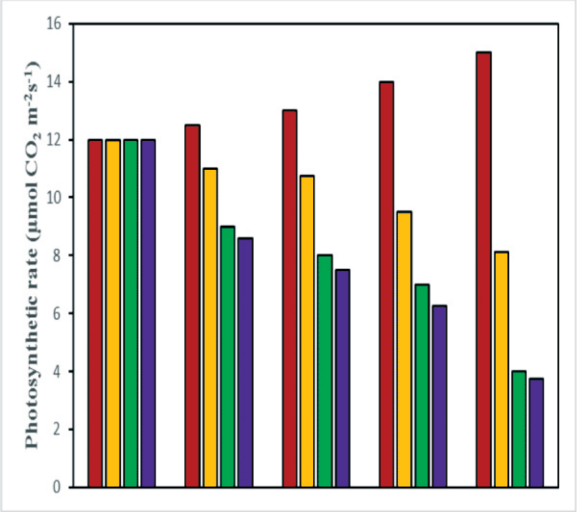
The percent increase in height was higher in case of control plants as compared to treated plants at harvest stage, i.e. 60 days after stress. We observed decline in percent increase of height ca. 24 to 69% compared to control as we move from 75, 50 and 25% of Field capacity (FC) of water stress. Upto 75% FC of water stress decline was very less after that level of water stress decline in height was severe. The trend found for plant stem diameter, leaf number and total leaf area on per plant basis were similar to that of plant height in drought induced poplar plants (Table 1). Percent increase in stem diameter was higher in case of control plants as compared to that of water stress treated plants and decline with severity of water stress. Decline can be neglected upto 75% FC of water stress.

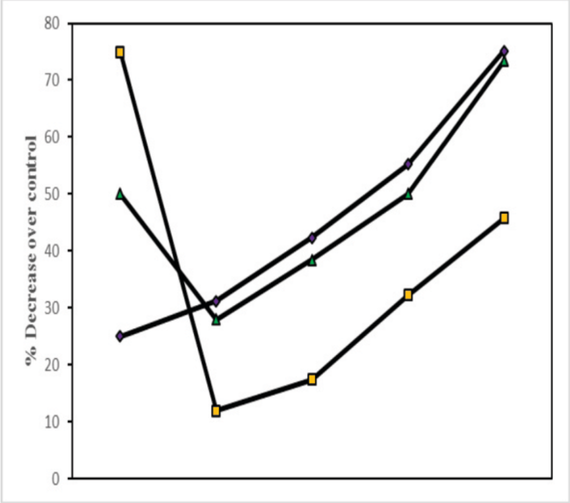
Specific leaf area (SLA) is the measure of leaf density or area occupied by unit weight of leaf. It was found that with increasing level of drought stress there was reduction in specific leaf area (Table 1) which might be adaptation against drought stress. Both the economical and biological, yields decreased with increasing levels of water stress. It was very less at the 75% of FC level of drought stress and after 50% of FC decrease was more than 50% at all the stage of sampling. Hence, harvest index (HI) also decrease with increasing levels of drought. We found significant decrease in HI with increasing levels of drought stress. The reduction was found to be 62% for Kranti clone of poplar when severe drought was imposed (25% of FC irrigation water). Decline was very little in case of 75% FC of drought stress.

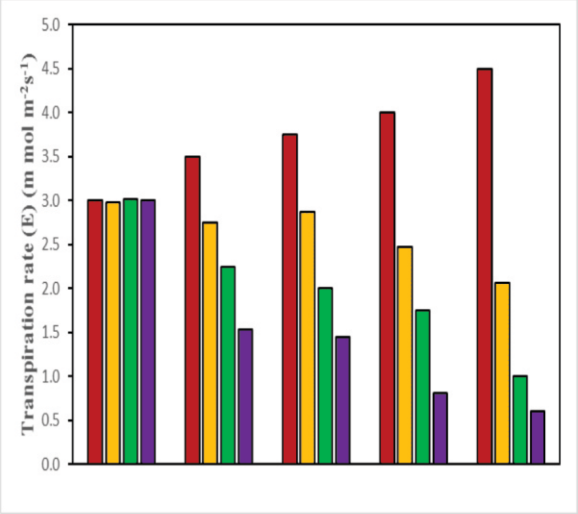
Table 1: Growth parameters of Populus deltoides L. clone Kranti seedling grown under four progressive water stress, i.e., control (100% FC) and stressed at 25, 50 and 75% of field capacity at final harvest (60 days after stress). Water stress levels maintained by withdrawing water levels i.e., control (100%of FC) and stressed (25, 50 and 75% of FC).

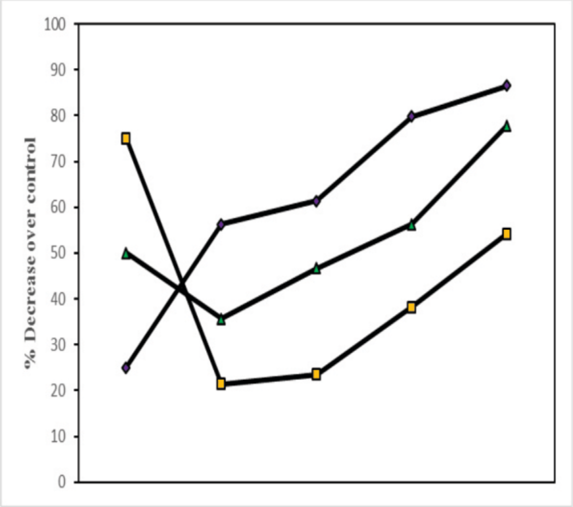
Characteristics	Control	Stressed (% of FC)			Decline over control (%)		
	(% of FC) 100	75	50	25	75	50	25
Height (cm)	78.80	60.20	36.00	24.60	23.6	54.3	68.8
Radial stem diameter (cm)	9.1	7.2	6.6	5.1	21.1	27.9	44.0
Number of leaves	21	14	6	4.2	33.3	71.4	80.0
Leaf area expansion (cm^2)	71.8	51.8	34.09	22.9	27.9	52.5	68.1
SLA ($cm^2 g^{-1}$)	98.30	93.33	91.00	90.33	27.86	52.52	68.11
Total biomass (g)	34.29	27.8	15.76	10.9	18.9	54.0	68.2
Harvest index	0.74	0.58	0.34	0.28	21.6	54.1	62.2

The effect of sustained drought upto 60 days resulted loss in photosynthetic CO^2 assimilation efficiency (Fig. 1). It was 15, 8.1, 4 and 3.7 μmol CO^2 m^{-2}s^{-1} at 60 days after imposing stress for 75, 50 and 25% of FC of water stress respectively. Drought treated plants showed gradual decrease in rate of photosynthesis with increased levels of severity and duration of drought. The drought affected seedlings followed a continuous loss in transpiration efficiency. The loss in









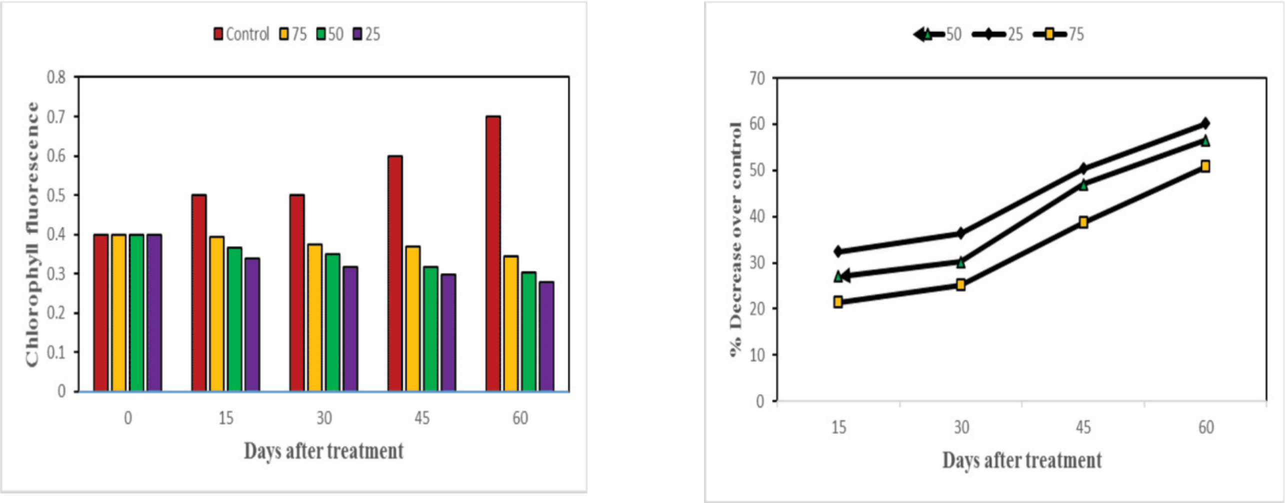


Fig. 1: (A) Leaf net photosynthetic rate (B) % decrease in photosynthetic rate over control (C) transpiration rate (D) % decrease in transpiration rate over control (E) PS II maximum quantum yield (Fv/ Fm) i.e., chlorophyll fluorescence (F) % decrease in chlorophyll fluorescence over control in Kranti clones of poplar seedlings grown under four progressive drought stress at different time after imposing drought stress. Drought stress levels maintained by withdrawing water levels i.e., control (100% of FC) and stressed (75, 50 and 25% of FC).

transpiration rate (E) is almost parallel to the loss in photosynthetic CO^2 assimilation (Fig. 1). The transpiration rate (E) decreased with gradual increase in drought levels and duration of imposing drought. Reductions were 54, 78 and 87% over control for 75, 50 and 25% of FC of water stress respectively at 60 days after stress. Decline in rate of photosynthesis and transpiration can be neglected upto 75% FC of water stress, but beyond that effect was very severe.

It was found that there was more reduction in chlorophyll fluorescence with severity of drought. The reduction in chlorophyll fluorescence increased as we increased the severity of stress. Reductions were 51, 57 and 60% over control for 75, 50 and 25% of FC of water stress respectively at 60 days after stress (Fig. 1).

Whole plants respond to drought through morphological, physiological, and metabolic modifications occurring in all plant organs. Xiao et al. (2009) reported that drought stress greatly inhibited the plant growth in two populations of Poplar tree, leading to a pronounced reduction in shoot height and basal stem diameter. At the whole-plant level, the effect of drought stress is usually perceived as a decrease in photosynthesis and growth, which is associated with alterations in carbon and nitrogen metabolisms (Cornic and Massacci, 1996). Long-term effect of water or drought stress results essentially changes in carbon allocation between plant organs, mainly in roots at the expense of leaves and shoots. The major mechanisms include curtailed water loss by increased diffusive resistance, enhanced water uptake with prolific and deep root systems and its efficient use, and smaller and succulent leaves to reduce the transpiration loss (Farooq *et al.,* 2009). Consequences of this phenomenon are an overall leaf area expansion reduction, linked to decreases in both number and individual size of the leaf and finally biomass (Tschaplinski *et al.,* 1998; Xiao *et al.,* 2005). A decrease of the specific leaf area (SLA: indicator of density and or thickness of the leaves) has often been observed with a subsequent increase of water retention due to an accentuation of the resistance to water transfer in the leaf (Nautiyal *et al.,* 2002, Singh *et al.,* 2013). The low SLA at high drought levels was due to the decreased translocation of nutrients to the leaves (Singh et al., 2013). Water shortage or water stress primarily affects net photosynthetic CO2 assimilation rate and therefore sets a limit to plant growth, productivity and yield (Haldimann *et al.,* 2008).

The reduction in the photosynthetic activity is due to several coordinated events, such as stomatal closure and the reduced activity of photosynthetic enzymes (Chaves *et al.,* 2003). Stomatal closure is probably the most important factor controlling carbon metabolism, but the relative role of stomatal limitation on photosynthesis depends on the severity of water deficit. Plants display a range of mechanisms to withstand drought stress. Chlorophyll fluorescence is the measure of maximum photochemical efficiency of photosystem II. Hence, increased levels of drought stress and exposure time of water stress results in decreased photochemical efficiency of photosystem II. Parallel decline in rate of photosynthesis and transpiration indicates that maintenance of transpiration rate is an essential factor for enabling CO^2 entrance through stomata into the chloroplast around Rubisco enzyme to sustain carboxylation efficiency adequately.

CONCLUSION

Our results clarified that acclimation to drought stress is not only related with the environmental factors of plant's natural habitats and genetics of plants but also related with the severity and duration of the drought event and their interaction. Different responses to different field capacity in poplar improved our understanding about the mechanisms that enable plants to survive under gradual water stress.

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