

IMPACT OF DROUGHT STRESS ON DALBERGIA LATIFOLIA ROXB. AND PONGAMIA PINNATA L. PIERRE UNDER NURSERY CONDITION: THE MORPHOLOGICAL, PHYSIOLOGICAL AND BIOCHEMICAL OVERVIEW

Shephali Sachan¹, Sandeep Kumar², Pooja Kattiparambil³, Anil Kumar^{4*}, Neeraj Kumar Kushwaha⁵, and Avinash Jain⁶,

^{1,2,6} Tropical Forest Research Institute, Jabalpur (M.P.), India

³ Arid Forest Research Institute, Jodhpur (Rajasthan), India

⁴ Subject Matter Specialist (Agroforestry) Krishi Vigyan Kendra, (Amihit, Jaunpur-2), Acharya Narendra Dev University of Agriculture and Technology, Ayodhya, (U.P.), India

⁵ Senior Technical Officer, Krishi Vigyan Kendra (Shivpuri), Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior (M.P.), India

Corresponding Author E-mail:* badalanil253@gmail.com

Abstract

Water is a very useful non-tangible tool for survival. However, the impact of rising drought conditions is unimaginable. Plant, on which every other living organism is dependent for livelihood, becomes major victim under this condition. Two forest plant species of tropical nature viz: *Dalbergia latifolia* and *Pongamia pinnata* were selected for pot culture experiments on CRBD style. Moderate drought (MD) and severe drought (SD) stress created artificially under nursery conditions for one-year duration. Height, Collar diameter, Leaf size, Total biomass and Carbon stock were determined as growth traits and calculated mortality percentage. Photosynthetic rate, transpiration rate and stomatal conductance were considered for physiological study. Total Chlorophyll, Carotenoids, Proline, Ascorbic acid and Protein content were analyzed to know biochemical status. Statistical data study was carried out using ANOVA, CD and correlation studies.

The experiment results showed that the drought condition becomes more pronounced in *D. latifolia* seedlings as 100% mortality rate obtained under MD. Further, decreasing biomass, carbon stock, physiological parameters, chlorophyll and protein content, and increasing carotenoid, proline and ascorbic acid content confirmed the morphological results. Hence, this article proves the plantation of *Pongamia pinnata* species is better in moderate drought-affected areas.

Keyword: Drought, Nursery, Morphological, Physiological and Biochemical.

Introduction

Amongst different global environmental problems, the increasing drought area is the most common. The rapidly growing population and urbanization make drought situations more

challenging (Ludlow and Muchow, 1990). The uneven distribution, too much wastage, pollution and unsustainable management of water bodies are the major cause of arising and increasing the drought condition (UN-Water, FAO, 2007).

Drought stress is responsible for plant's low water availability, which causes a significant reduction in growth, productivity and ultimately their yields (Ludlow and Muchow, 1990). Drought stress condition arises due to a shortage of water in the soil than its optimal prerequisite. In addition, the atmospheric conditions cause constant water loss by transpiration or evaporation phenomena. The presence of inadequate water quantity in the soil makes them incapable to receive oxygen by plants, that is, creating an anaerobic environment for them which in turn give rise to stressful condition (Jaleel *et al.*, 2009). The drought condition is the result of many factors such as evaporative demands, occurrence and distribution of rainfall, and soil moisture storing capacity (Wery *et al.*, 1994). The variety of modifications begins due to drought in the biological and chemical processes of plants, affecting plant growth and soil properties (Bigler *et al.*, 2006).

The people suffering from substantial scarcity of water are around 1.2 billion with more 500 million people approaching this situation. Alternatively, economically water shortage (lack of required infrastructures in order to take water from various water bodies) affected approximately 1.6 billion people. This way the demand for water has become double the speed of the increasing population in the preceding century (FAO, 2007).

Trees death in the European southern region due to elevated temperature (Bigler *et al.*, 2006), the increasing mortality rates of western North American temperate and boreal forests trees (van Mantgem *et al.*, 2009) and expanding trees death since 1997 which is responsible for damage of 10 mha of forest land (Raffa *et al.*, 2008) are the examples of loss in forestry sector because of drought stress. In India, About 32.55% of land of the country is affected by drought stress which is spread over administrative districts including Bihar, Haryana, Andhra Pradesh, Jammu and Kashmir, West Bengal, Madhya Pradesh, Karnataka, Gujarat, Odisha, Tamil Nadu, Rajasthan, Uttar Pradesh and Maharashtra (Nagarajan, 2003). The events like global warming, changes in the rainfall pattern, and powerful El Niño Southern Oscillation (ENSO) episodes, may increase the occurrence and harshness of drought in the tropical region (Nepstad, 2004).

Drought condition interferes with the plant's morphological, physiochemical and molecular processes consequent to photosynthesis inhibition, chlorophyll content reduction, protein changes, stomata closure, consecutive reduction of transpiration, and finally growth inhibition. There also occur differential responses of antioxidative enzyme (Xiao *et al.*, 2008, Chave *et al.*, 2009). The defensive processes inbuilt in plants like higher antioxidative enzymes [Peroxidase (POD), Superoxide dismutase (SOD), Monodehydroascorbatereductase (MDHAR), Catalase (CAT), Glutathione reductase (GR) and Ascorbate peroxidase (APX)], stress metabolites accumulation [Glycine betaine, Glutathione, Malate dehydrogenase (MDHA), alpha-tocopherol, Polyamines and Proline], and reduction in reactive oxygen species (ROS) accumulation. However, on the severity

of stress the inbuilt defence mechanism failed to protect the plant against the lethal effects (Reddy *et al.*, 2004; Amarjit *et al.*, 2005; Shao *et al.*, 2008).

The two species *viz.* *Dalbergia latifolia* and *Pongamia pinnata* selected for experimental purpose, belong to Fabaceae family, that is, with the nitrogen-fixing specificity and with the good medicinal and timber value (Orwa *et al.*, 2009; Arpiwi, 2013). *Pongamia pinnata* species is also known for its valuable diesel like seed oil, known as biodiesel plant (Arpiwi, 2013). The current research article focus on identifying the performance of *Dalbergia latifolia* and *Pongamia pinnata* seedlings under different levels of drought stress conditions. The study also reflects several morphological, physiological and biochemical changes, the selected species undergo in a nursery environment. The species screening is necessary to raise the plantations in drought affected areas and in nurseries for commercial purposes.

Material and Methods

1. Experiment Layout

The research was carried out in Tropical Forest Research Institute (TFRI) nursery, Jabalpur (M.P.) following the pot culture method in Complete Randomized Block Design (CRBD) manner for one-year duration. *Dalbergia latifolia* (Shisham) and *Pongamia pinnata* (Karanj), the 2 tropical forest tree species were chosen for experimentation. The seeds of the selected tree species were first planted in nursery beds (10m x 1m) in June before the first shower of rainfall using sand as a sowing medium. After germination, the plantlets with 2-3 leaves were shifted to soil, sand and farm yard manure (FYM) (2:1:1) containing transparent polythene bags (15 cm x 23 cm). For almost one month the polythene bags were firstly kept under shade and then openly in order to adjust to the surrounding environment.

2. Drought Treatment

The experiment was started in August 2015. Three treatments *viz.* Control (W1), Moderate drought (W2) and Severe drought (W3) were considered for experimentation. Each treatment consisted of nine seedlings with three replications. Each polybag was given water according to the calculated species-specific field capacity (Table 1), the formula adopted by Tyree *et al.*, (2002) method. Cumulative Pan Evaporation (CPE = Sum of evaporation values) values were calculated using the Open Pan Evaporimeter (Eliades, 1988; Savva and Frenken, 2002) which was used as the basis for interval period for providing irrigation. Permanent wilting point (PWP) was also determined in each species by the method adopted by Savva and Frenken, (2002), Table: 1. The CPE values are counted in fixed time intervals up to the particular permanent wilting point (PWP) in each species. This created Severe drought (SD) conditions. On the other hand, half of the CPE values counted for severe drought conditions, creating Moderate drought (MD) conditions. The frequency of watering polybags varied in different seasons, which was high during summer and low during

winter (Table: 2). The complete experiment was covered with transparent polythene shade during the rainy season.

3. Morphological parameters

Each seedling was measured for height, collar diameter and leaf size before the treatment stage and after treatment quarterly for one year. Scale/inch tape and vernier calliper were used for measuring height and collar diameter (CD) respectively (Husen, 2009). The leaf size of mature leaves was measured by the Systronics leaf area meter instrument.

Biomass was measured in the last period of an experiment by following the destructive method. Three seedlings from each treatment were uprooted and oven-dried at 50-60⁰C till constant weight is achieved. Carbon stock was calculated on the basis of IPCC (2006) Guidelines according to which 50% of the dried total biomass of plants is in the form of carbon.

The mortality percentage was calculated per treatment at the end of one year.

4. Physiological parameters

Photosynthetic rate, transpiration rate and stomatal conductance were analyzed by CID make Photosynthetic instrument between 8:00 AM to 10:00 AM, half-yearly (January and August 2016) for one year. PAR value was recorded as 803-2298 $\mu\text{mol}/\text{m}^2/\text{s}$. The instrument was calibrated before use. Mature leaves were considered for measurement per treatment and per replication by placing them in the leaf chamber of the instrument (Husen, 2009).

5. Biochemical parameters

Chlorophyll, Carotenoids, Proline, Protein and Ascorbic Acid were analyzed in the last part of the experiment.

Chlorophyll and Carotenoids were determined according to Arnon's procedure (1949). 0.1g fresh leaf sample was taken to crush in 5ml 80% acetone following centrifugation at 5000 rpm for 5min. The supernatant was separated and the procedure was repeated again with the pellet. Both the supernatant mixed and proceeded for absorbance at 645, 663 (Chlorophyll) and 470 nm (Carotenoids) against the 80% acetone (solvent) as blank. The extracted quantity was considered in the mg chlorophyll/g tissue and further calculated by the formula illustrated in Sadasivam and Manickam (2008).

Bates et al. (1973) technique was used for the quantification of proline content in the leaves. 0.1g dried leaf sample crushed in 5ml sulfosalicylic acid (3% aqueous). 3000 rpm rotation scale used for centrifugation for 25 min (especially for dried sample). The solution was then filtered by Whatman filter paper (No. 2). A test tube with 2ml of filtrate, glacial acetic acid and acid ninhydrin was taken to heat in the boiling water bath for 1 hour. The reaction was terminated by inserting

the test tube into the ice bath. Further, the reaction mixture stirred for 20-30 seconds containing 4ml toluene. The toluene layer with red colour was separated and measured at 520 nm. The standard curve was prepared by running a series of standard using pure proline in a parallel way. For both sample and standard toluene was taken as a blank. The proline quantity in the trial sample was expressed with the help of a standard curve as illustrated in Sadasivam and Manickam (2008).

Ascorbic acid (Vitamin C) was determined as per suggested by Malik and Singh (1980) using a spectrophotometer illustrated in Sadasivam and Manickam (2008).

Protein was measured following the Bradford method by UV-VIS Spectrophotometer with slight modification. A fresh leaf sample (50µg) was taken and grounded finely using liquid nitrogen in an already chilled pestle-mortar. 5 ml lysis buffer (1.0M Tris Base buffer of pH 8.0 containing 1% PVP, 25 mM EDTA10 and mM β-mercaptoethanol) was added to this crushed sample. The test sample was centrifuged for 10 min at 13000 rpm in 4⁰C and then 100 µl supernatant proceeded for analysis. A standard curve of 10 to 100 µl concentration of working standard (Bovine Serum Albumin) was prepared with distilled water (100 µl) used as a reagent blank (for both samples and standards). All the test tubes were filled with 300 µl distilled water. Each tube were further added with 3 ml dye (Coomassie brilliant blue dye) and manually shaken. The test tubes were kept for 15-30 minutes in dark and finally, the absorbance was calculated at 595 nm. The protein concentration in the test samples was quantified with the help of a standard curve.

6. Statistical Analysis

ANOVA (Analysis of Variance) table was used for analyzing the data statistically. The table was prepared with the help of CD (critical difference) present at 5% significance levels amongst the selected treatments. Further, the selected different parameters were also evaluated for correlation at 5% and 1% significant levels.

Results

1. Morphology

Height, CD and leaf size differ significantly ($P < 0.05$) amongst selected treatments (Fig.: 1-6). *D. latifolia* and *P. pinnata* seedlings under control conditions survived for one year *i.e.* August 2016. *D. latifolia* seedlings continued to exist till May 2016 under W2 and W3 treatments while *P. pinnata* seedlings survived till May 2016 under W3 treatment.

Height and CD of *D. latifolia* and *P. pinnata* seedlings continuously increased with age but followed decreasing trend on increasing severity of stress. In *D. latifolia*, height raised to 117.6% under W1 and 67.24% and 57.63% under W2 and W3 treatments while CD increased by 3.25 times under W1, 2 times under W2 and 1.27 times under W3 treatments till survival of the plants. In *P. pinnata*, 91.63%, 97.74% and 11.45% increase in height with age was observed in control, MD and SD treatments respectively. Though, 6.36 times increment under W1, 2.02 times under W2 and 1.79 times under W3 was observed in CD during the experimental period.

The average leaf size of *D. latifolia* decreased from August 2015 (3.85 cm²) to May 2016 (2.75 cm²) and then increased to a maximum (6.70 cm²) in August 2016 after rainfall in W1. Under W2 and W3 treatments decreased till leaf fall. *P. pinnata* leaves attained maximum size just before leaf fall under W1 and W2, but under W3 treatment leaf size continuously decreased, hence the effect was more pronounced in W3 than W2.

The biomass and carbon content parameters were analyzed at the last phase of experiment (Table: 3). They were observed significantly ($P < 0.05$) different under different treatments. The biomass and carbon content found to be declined under drought stress with the harshness of stress from W2 to W3. *P. pinnata* seedlings confirmed the highest reduction in biomass (69.37% & 89.29%) and lowest in *D. latifolia* (24.67% & 43.27%), under W2 and W3 treatments respectively. The root/shoot ratio was recorded in both the species and was found to be greater than 1 (< 1) and higher than *D. latifolia* in *P. pinnata* seedlings.

Maximum mortality was found in *D. latifolia*, followed by *P. pinnata* (Fig. 7). Mortality increased with an increase in drought stress *i.e.* minimum in control, followed by W2 and W3. Total mortality was recorded in *D. latifolia* at the end of the experiment (August 2016) under W2 and W3 treatment respectively whereas *P. pinnata* showed total mortality under only W3 treatment.

2. Physiology

The final half-yearly observations (July 16) in *D. latifolia* for W2 & W3 and in *P. pinnata* for W3 were unavailable due to the nonappearance of leaves. Like biomass content of the seedlings, Photosynthetic rate (PR), Transpiration rate (TR) and Stomatal conductance (SC) also showed an identical trends. The highest PR, TR and SC were found in *P. pinnata* followed by *D. latifolia* under W1 conditions. But under drought treatments, maximum PR, TR and SC were traced in *D. latifolia* followed by *P. pinnata* seedlings. PR, TR and SC dropped off from W2 to W3. The selected physiological parameters were recorded maximum during the rainy season (July 16) and minimum during winters (January 16) in both the species (Table: 4).

3. Biochemistry

The foliar biochemical showed a significant ($P < 0.05$) difference (Fig.: 8 & 9). Chlorophyll and ascorbic acid amounts were determined maximum in *D. latifolia*, followed by *P. pinnata* and vice versa for other selected biochemical parameters.

The chosen tree species seedlings showed a decrease in chlorophyll content from W2 to W3. Carotenoids were observed minimum in control and increased from W2 to W3. Proline and ascorbic acid also followed an identical trend to carotenoids and raised from W2 to W3. Protein content decreased under drought stress and was found maximum in W1 and minimum in W3 seedlings.

4. Correlation

The growth traits are evidenced for significant ($P < 0.05$ to $P < 0.01$) positive correlation with physiological characteristics and biochemical parameters like total chlorophyll and protein in the

selected species. However, growth traits, physiological characteristics and total chlorophyll in *D. latifolia* did not show a significant ($P>0.05$) correlation with protein. The carotenoid, proline and ascorbic acid were observed in significantly ($P<0.05$) negative correlation with growth and physiological parameters (Tables: 5 & 6).

Discussion

The drought condition in the soil creates water deficiency inside the plant body, hampering the biological processes as a result of which the growth/yield becomes highly negatively affected. The current work proved that the height and CD of the selected tree species seedlings significantly differed among the selected treatments. During the course of experiment, the height and CD of the seedlings found increased on maturity but reduced when drought condition increased. *Dalbergia latifolia* seedlings observed to be more sensitive than *Pongamia pinnata* under drought conditions. The above finding coincides with the results of Ashraf (2004), whose study on *Dalbergia sissoo* and *Dalbergia latifolia* plants under water deficit conditions recorded a significant reduction in height and stem diameter. Also, in another study performed by Husen (2009) and Sneha *et al.*, (2012) under drought conditions, the selected *Tectona grandis* plant showed a reduction in growth. Similarly, reduction in height with the increasing water deficit condition was found in *Tectona grandis* L., *Dalbergia sissoo* Roxb., *Shorea robusta* Gaertn.f., *Leucaena leucocephala* (Lam.) de Wit., and *Albizia lebbek* Benth., during the experiment conducted in Tarai region (Uttarakhand) from 2002 to 2004. *Leucaena leucocephala* reduced maximum while *Albizia lebbek* reduced minimum (Rao *et al.*, 2008).

In *D. latifolia* and *P. pinnata* tree species, November to February is the time period considered for leaf drop while April to May is for the appearance of new leaves (Deciduous nature). Leaf size was recorded as minimum in the first data collected after leaf fall *i.e.* in February and maximum after the rainy season in control except for *P. pinnata* leaves which maintained the maximum size at the time of leaf fall. This might be a type of strategy to maintain the photosynthesis process by increasing the surface area of leaves for absorption of more light during the leaf fall period (Osakabe *et al.*, 2014). Under drought treatments, on the maturity of seedlings and increasing severity of stress, the decreasing trend observed in leaf size. From May onwards lack of leaves was recorded under drought which could be due to the non-emergence of leaves after leaf fall as there occurred deficiency of water in the seedlings. The leaf also acquires a smaller size which might be an adaptive feature under drought treatments due to restricted photosynthesis. Water availability to the plants further decreased during summer with a higher deficit of vapour pressure in the air along with the increase of transpiration rate in the plant canopies. Therefore, there occurs either decrease in the size of leaves or leaf fall, especially under drought conditions (Kirschbaum, 2004). The reduced leaf expansion rate further reduces the area for transpiration. This feature may be helpful to plants under drought stress. (Mahajan and Tuteja, 2005). The reduction of total leaf area was observed during the dry season when the atmospheric vapor pressure is very low, thus water loss was prohibited more efficiently in *Melia azedarach* under drought stress (Jhou *et al.*, 2017).

The biomass and carbon stock showed a significant reduction under W2 and W3 treatments. The biomass was observed to be decreasing in *Albizzia lebbek*, *Dalbergia latifolia*, *Leucaena leucocephala*, *Shorea robusta*, *Tectona grandis* (Tarai region, Uttarakhand), and in *Pongamia pinnata* (Acharya N.G. Ranga Agricultural University Campus, Tirupati) species with the raising level of drought conditions in an experiment conducted by Rao *et al.*, (2008) and Swapna and Rajendrudu (2015) respectively. The drought condition disturbs the balance between roots water uptake and shoots photosynthesis which results in allocating more biomass in roots to increase additional water absorption, thus root/shoot ratio rises. In the current study results, the root/shoot ratio (<1) increased in the *P. pinnata* tree species under drought stress. Sneha *et al.*, (2012) also found a higher root/shoot ratio under the treatments IW/ET= 0 and 0.3 (0 and 30% irrigation water) in an experiment conducted on six months old seedlings of teak.

Minimum mortality in *P. pinnata* was found among the selected tree species under study, exhibiting resistance to drought stress. The seedlings of this species survived till the end of the experiment under moderate drought conditions. *D. latifolia* seedlings did not survive till the end of experiment and showed susceptibility toward drought stress. Mortality in plants due to drought conditions could be due to failure of phloem transport capacity between the source (leaves) and sink (roots) organs because of lower hydrostatic pressure (Dannoura *et al.*, 2018).

The maximum photosynthetic rate was recorded in *P. pinnata* under control, which sharply reduced to a minimum under drought treatments depicts the positive correlation of photosynthetic rate with biomass content, *i.e.*, more the photosynthetic rate takes place more biomass building up occurs in the seedlings. Both the species followed a reduction in the selected physiological traits like photosynthetic rate, transpiration rate and stomatal conductance under drought stress. The drought experiment performed on *Melia azedarach* and *Swietenia macrophylla* (Jhouet *al.*, 2017) and on teak clones of 4.5 years old (Husen, 2009) provided similar results, *i.e.*, reduction of physiological parameters. *Dalbergia sissoo* and *D. latifolia* seedlings also showed a significant reduction in stomatal conductance (*gs*), transpiration rate (*E*), and net photosynthetic rate (*PN*) due to drought stress (Ashraf *et al.*, 2004). The roots expose first under drought stress. This conveys the signal from stem to leaves and a reduction in stomatal conductance occur. Further, this entire circumstances becomes major reason of reduction in photosynthesis under drought stress (Kausar *et al.*, 2006; Schachtman and Goodger, 2008).

Silva *et al.* (2010) found a significantly higher net photosynthetic rate during the rainy season in the plants of *Byrsonima sericea*, which corroborate our study, where higher photosynthetic rate, transpiration rate and stomatal conductance were observed during the rainy season chased by winter.

The alterations in the selected foliar biochemicals were also found to differ significantly under drought stress. The chlorophyll content was recorded declining in both the tree species. Water is very necessary for the photosynthesis process. The unavailability of water causes photoinhibition

which results in damage to chlorophyll pigment (Chaudhry, 2006). The chlorophyll loss, inspite of being a negative modification under drought stress, might be proven as an adjustive feature. The low chlorophyll pigment minimizes the light-harvesting and protect the photosynthetic machinery from any further damage (Munne-Bosch and Alegre, 2000; Kranner *et al.*, 2002). The leaves showed a reduction in an experiment conducted by Nautiyal *et al.*, (1996) performed a drought experiment on *Pongamia pinnata* (L) Pierre under controlled laboratory conditions. They analyzed minimum chlorophyll a, b and total chlorophyll quantity in the leaves which caused less photosynthesis and finally low growth. The comparative chlorophyll content also decreased in the Teak seedlings under IW/ET= 0 and 0.3 (0 and 30% irrigation water) drought treatments (Sneha *et al.*, 2012).

Total carotenoids, proline and ascorbic acid content showed increasing with rising drought levels. The abiotic stress causes the release of various defensive compounds. These compounds protect the photosynthetic apparatus against the destructive effects of light and Reactive Oxygen Species (ROS). The ascorbic acid and carotenoids are also the part of the defensive compound family (Apel and Hirt, 2004). The accumulation of proline for osmotic adjustment is a familiar way of protecting function in plants (Xiao *et al.*, 2008). Toscano *et al.*, (2016) showed that proline was higher in two ornamental shrubs (*Eugenia* and *Photinia*) when subjected to drought stress. *Eugenia* showed a higher response for proline and then greater tolerance as compared to *Photinia* under drought stress. The ascorbic acid is produced when over-accumulation of stress-induced oxidative damages occurs to provide protection to cells and organelles (Latif *et al.*, 2016; Mukhtar *et al.*, 2016; Naz *et al.*, 2016).

The current study illustrates the decreasing trend of protein content in both tree species with the raising drought levels. Similarly, in *Pongamia pinnata* seedlings, a significant decline in protein content was observed, when subjected to different drought conditions. The lower protein content in water stressed seedlings may be attributed to the damage to membranes (Swapna and Rajendrudu, 2015).

The positive correlation between growth traits and physiological parameters evidently explains the fact that the better photosynthetic rate and osmoregulation process are the reason for optimal growth and biomass. These physiological parameters reduce under drought effect to avoid water loss, thus disturbing the production and growth of plants (Larson and Funk, 2015). The result coincides with the *T. grandis* (Husen, 2009) and *Salix* species (Singh *et al.*, 2012) under drought and normal conditions respectively. A significant negative correlation between proline content, antioxidants (Carotenoid and Ascorbic acid) and physiological parameters were observed in the current studies. The two ornamental shrubs *i.e.* *Photinia X fraseri* Dress and *Eugenia uniflora* L. also showed identical consequences under drought stress (Toscano *et al.*, 2016). Both tree species illustrated the significant positive correlation among physiological parameters and chlorophyll content, and growth traits. The above fact was also observed in other species like *Acacia*

auriculiformis, *Alstonia macrophylla*, *Terminalia arjuna*, *Azadiracta indica* and *Artocarpus heterophyllus* (De Costa and Rozana, 2000).

Conclusion

Drought condition affects the plant negatively in terms of growth, yield and productivity, thus, causing huge damage to the agriculture and forestry sectors. In the present study, drought stress negatively impacted physiological parameters, total chlorophyll and protein content of *Dalbergia latifolia* and *Pongamia pinnata* seedlings. These alterations in physiology and biochemistry have greatly hampered the growth traits of seedlings. The ascorbic acid, carotenoids, and proline content were increased under the current study. The significant difference confirmed the above results. Further, the correlation studies showed clearly the relationship of species morphology with its physiology and biochemicals. The role of the defensive mechanism was evidently well-built in plants to provide protection for survival. The survivability of *P. pinnata* (W2 and W3) specifies the ability and level of tolerating the drought stress. The information about the drought tolerance level of any plant species is very important to have a clear plantation aim in the affected sites. The forest tree species provides plentiful substantial and insubstantial benefits. Hence, the plantations of various economically benefitted tree species local to such affected sites are suitable for accomplishing the resource demands of the increasing population.

References

- Amarjit, K.N., Kumari, S. and Sharma, D.R. 2005. In vitro selection and characterization of water stress tolerant cultures of bell pepper. *Indian Journal of Plant Physiology*, 10 (1): 14-19.
- Apel, K., and Hirt, H. (2004). Reactive oxygen species: metabolism, oxidative stress and signal transduction. *Annual Review of Plant Biology*, 55:373–399.
- Arnon, D.I. 1949. *Plant Physiology*, 24, p. 1.
- Arpiwi, N.L., Yan G., Barbour E.L. and Plummer J.A. 2013. Genetic diversity, seed traits and salinity tolerance of *Millettia pinnata* (L.) Panigrahi, a biodiesel tree. *Genetic Resources and Crop Evolution*, 60: 677–692.
- Ashraf, M., Ashraf, M.Y., Khaliq, A. and Rha, E.S., Growth and leaf gas exchange characteristics in *Dalbergia sissoo* Roxb. and *D. Latifolia* Roxb. under water deficit. *Photosynthetica*, 2004, 42 (1), 157-160.
- Bates, L.S., Waldeen, R.P. and Teare, I.D. 1973. *Plant Soil*, 39, p. 205.
- Bigler, C., Braker, O.U., Bugmann, H., Dobbertin, M. and Rigling, A. 2006. Drought as an inciting mortality factor in Scots pine stands of the Valais, Switzerland. *Ecosystem*, 9: 330–343.
- Bradford, M.M. 1976. *Analytical Biochemistry*, 72.
- Chaudhry, A.K. 2006. Abiotic and biotic stresses on shisham trees. National Agricultural Research Centre, Islamabad.
- Chave, J., Coomes, D., Jansen, S., Lewis, S.L., Swenson, N.G. and Zanne, A.E., Towards a worldwide wood economics spectrum. *Ecology Letters*, 2009, 12, 351–366.

- FAO. 2007. Coping with water scarcity - Challenge of the twenty-first century. UN-Water.
- Dannoura, M., Epron, D., Desalme, D., Massonnet, C., Tsuji, S., Plain, C., Priault, P and Gérant, D. 2018. The impact of prolonged drought on phloem anatomy and phloem transport in young beech trees. *Tree Physiology*, tpy070.
- De Costa, W.A.J.M. and Rozana, M.F., Effects of shade and water stress on growth and related physiological parameters of the seedlings of five forest tree species. *Journal of the National Science Foundation of Sri Lanka*, 2000, 28 (1), 43-62.
- Eliades, G. 1988. Irrigation of greenhouse-grown cucumbers. *Journal of Horticultural Science*, 63(2): 235-239.
- Husen, A. 2009. Growth Characteristics, Physiological and Metabolic Responses of Teak (*Tectona Grandis* Linn. F.) Clones Differing in Rejuvenation Capacity Subjected to Drought Stress. *Silvae Genetica*, 59, 2.
- Jaleel, C.A., Manivannan, P., Wahid, A., Farooq, M., Somasundaram, R. and Panneerselvam, R., Drought stress in plants: A review on morphological characteristics and pigments composition. *International Journal of Agriculture and Biology*, 2009, 11, 100–105.
- Jhou, H.C., Wang, Y.N., Wu, C.S., Yu, J.C. and Chen, C.I., Photosynthetic gas exchange responses of *Swietenia macrophylla* King and *Melia azedarach* L. plantations under drought conditions. *Botanical Studies*, 2017, 58(1), 57.
- Kirschbaum, M. U. F. 2004. Direct and indirect climate change effects on photosynthesis and transpiration. *Plant Biology*, 6: 242– 253.
- Kranner, I., Beckett, R.P., Wornik, S., Zorn, M. and Pfeifhofer, H.W. 2002. Revival of a resurrection plant correlates with its antioxidant status. *The Plant Journal*, 31: 13–24.
- Larson, J.E. and Funk, J.L., Seedling root responses to soil moisture and the identification of a belowground trait spectrum across three growth forms. *New Phytologist*, 2015, 210, 827–838.
- Latif, M., Akram, N. A. and Ashraf, M. 2016. Regulation of some biochemical attributes in drought-stressed cauliflower (*Brassica oleracea* L.) by seed pre-treatment with ascorbic acid. *Journal of Horticultural Science and Biotechnology*, 91: 129–137.
- Ludlow, M. M. and Muchow, R. C., A critical evaluation of traits for improving crop yields in water- limited environments. *Advances in Agronomy*, 1990, 43, 107–153.
- Mahajan, S. and Tuteja, N., Cold, salinity and drought stresses: An overview. *Archives of Biochemistry and Biophysics*, 2005, 444(2), 139-158.
- Malik, E.P. and Singh, M.B. 1980. *Plant Enzymology and Histochemistry* (1st Edn.) Kalyani Publishers: New Delhi, 286.
- Mukhtar, A., Akram, N. A., Aisha, R., Shafiq, S. and Ashraf, M. 2016. Foliar-applied ascorbic acid enhances antioxidative potential and drought tolerance in cauliflower (*Brassica oleracea* L. var. *Botrytis*). *Agrochimica*, 60: 100–113.
- Munné-Bosch, S. and Alegre, L., Changes in carotenoids, tocopherols and diterpenes during drought and recovery, and the biological significance of chlorophyll loss in *Rosmarinus officinalis* plants. *Planta*, 2000, 210, 925–931.

- Nagarajan, R., Drought: Assessment, Monitoring, Management and Resource Conservation. Capital Publishing Company, New Delhi, 2003, pp.312.
- Nautiyal, S., Negi, D.S. and Kumar, S., Effect of water stress and antitranspirants on the chlorophyll contents of the leaves of *Pongamiapinnata* (L.) Pierre. *The Indian Forester*, 1996, 122 (11).
- Naz, H., Akram, N. A. and Ashraf, M. 2016. Impact of ascorbic acid on growth and some physiological attributes of cucumber (*Cucumissativus*) plants under water-deficit conditions. *Pakistan Journal of Botany*, 48: 877–883.
- Nepstad, D., et al. 2004. Amazon drought and its implications for forest flammability and tree growth: a basinwide analysis. *Global Change Biology*, 10(5): 704–717.
- Orwa, C., Mutua, A., Kindt, R., Jamnadass, R. and Anthony, S. 2009. Agroforestry Database: a tree reference and selection guide version 4.0. World Agroforestry Centre, Kenya.
- Osakabe, Y., Osakabe, K., Shinozaki, K., and Lam-Son, T. 2014. Response of plants to water stress. *Frontier of Plant Science*, 5(86):1–8.
- Raffa, K.F. et al., Cross scale drivers of natural disturbances prone to anthropogenic amplification: the dynamics of bark beetle eruptions. *Bioscience*, 2008, 58, 501–517.
- Rao, P.B., Kaur, A. and Tewari, A., Drought resistance in seedlings of five important tree species in Tarai region of Uttarakhand. *Tropical Ecology*, 2008, 49, 43-52.
- Reddy, A.R., Chiatanya, K.V. and Vivekanandan, M., Drought induced responses of photosynthesis and antioxidant metabolism in higher plants. *Journal of Plant Physiology*, 2004, 161(11), 1189–1202.
- Savva, A.P. and Frenken, K. 2002. Crop Water Requirements and Irrigation Scheduling. Irrigation Manual Module 4, FAO, Harare.
- Schachtman, D.P. and Goodger, J.Q.D. 2008. Chemical root to shoot signaling under drought. *Trends Plant Science*, 13: 281–287.
- Shao, H.B., Chu, L.Y., Jaleel, C.A. and Zhao, C.X., Water-deficit stress-induced anatomical changes in higher plants. *Comptes Rendus Biologies*, 2008, 54(3), 215–225.
- Silva, E.N., Ribeiro, R.V., Ferreira-Silva, S.L., Viégas, R.A. and Silveira, J.A.G. 2010. Comparative effects of salinity and water stress on photosynthesis, water relations and growth of *Jatropha curcas* plants. *Journal of Arid Environments*, 74(10): 1-8.
- Singh, N.B., Sharma, J.P., Huse, S.K., Thakur, I.K., Gupta, R.K. and Sankhyan, H.P., Heritability, genetic gain, correlation and principal component analysis in introduced willow (*Salix* species) clones. *Indian Forester*, 2012, 138 (12), 1100.
- Sneha, C., Santhoshkumar, A.V. and Sunil, K.M., Effect of controlled irrigation on physiological and biometric characteristics in teak. *Journal of Stress Physiology & Biochemistry*, 2012, 8(3), 196-202.
- Swapna, B. and Rajendrudu, G., Seed Germination of *Pongamia Pinnata* (L.) Pierre under Water Stress. *Research Journal of Recent Sciences*, 2015, 4(6), 62-66.

- Toscano, S., Farieri, E., Ferrante, A. and Romano, D. 2016. Physiological and Biochemical Responses in Two Ornamental Shrubs to Drought Stress. *Frontiers in Plant Science*, 7: 645.
- Tyree, M.T., Vargas, G., Engelbrecht, B.M.J. and Kursar, T.A. 2002. Drought until death do us part: a case study of the desiccation-tolerance of a tropical moist forest seedling-tree, *Licania platypus* (Hemsl.) Fritsch. *Journal of Experimental Botany*, 53(378): 2239-2247.
- Van Mantgem, P.J., Stephenson, N.L., Byrne, J.C., Daniels, L.D., Franklin, J.F., Fulé, P.Z., et al., Widespread Increase of Tree Mortality Rates in the Western United States. *Science*, 2009, 323, 521–524.
- Wery, J., Silim, S.N., Knights, E.J., Malhotra, R.S. and Cousin, R. 1994. Screening techniques and sources and tolerance to extremes of moisture and air temperature in cool season food legumes. *Euphytica*, 73: 73–83.
- Xiao, X., Xu, X. and Yang, F. 2008. Adaptive responses to progressive drought stress in two *Populus cathayana* populations. *Silva Fennica*, 42 (5): 705-719.

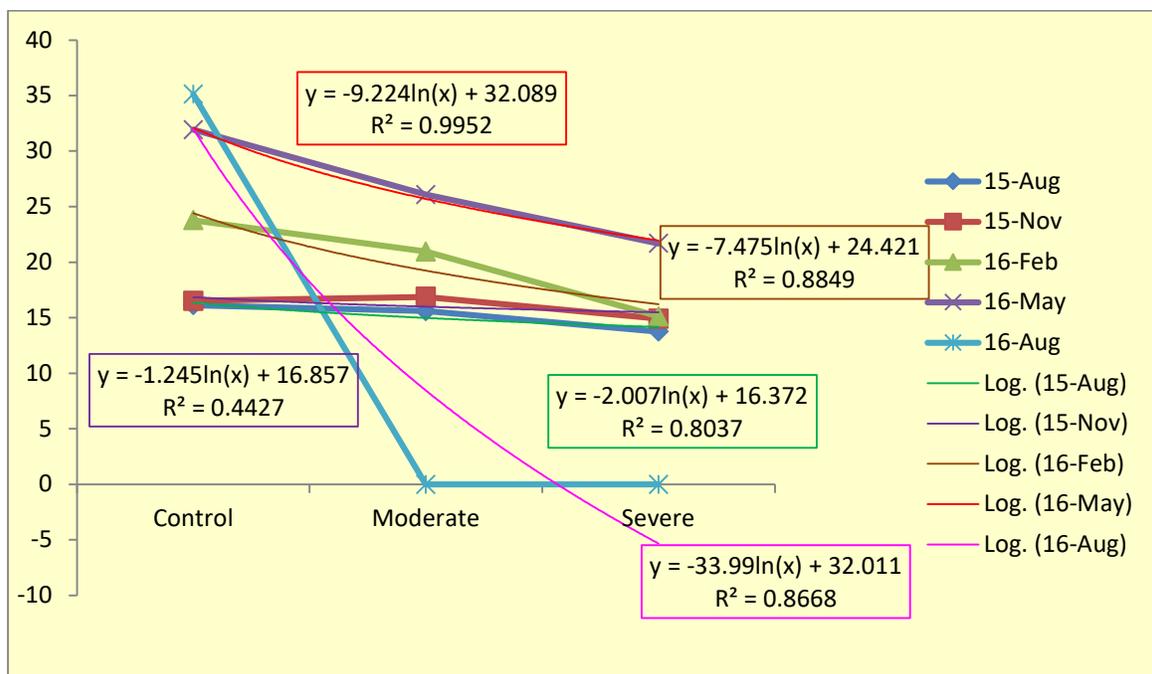


Fig. 1: Effect of drought on height of *Dalbergia latifolia* showing trendline and regression graph with R² value

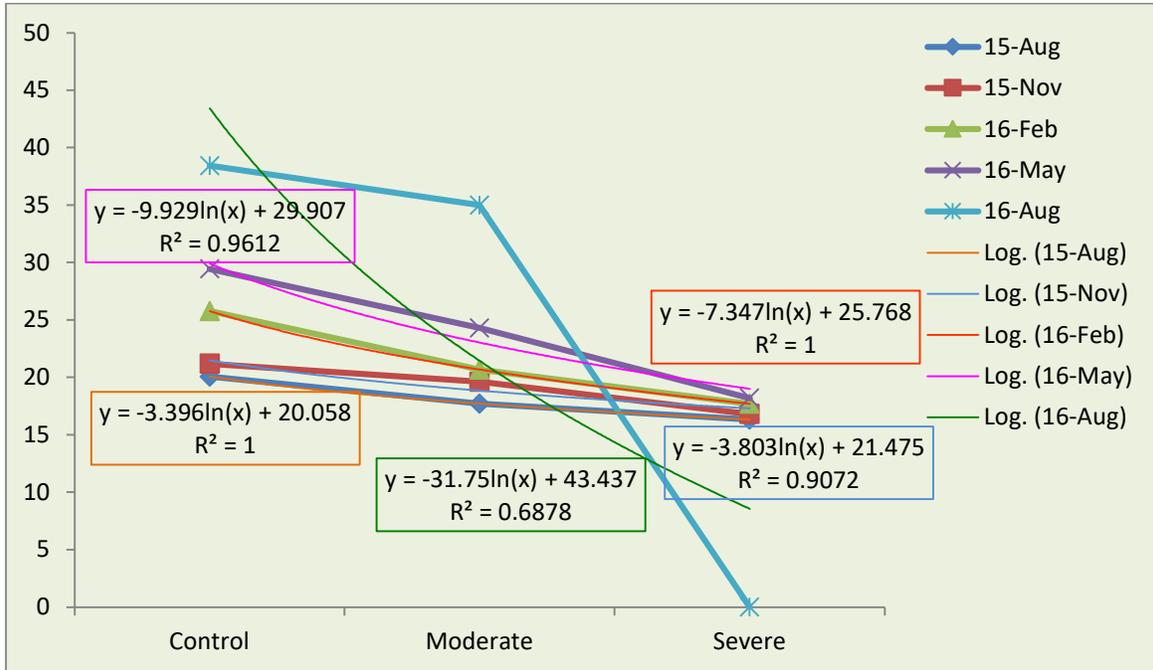


Fig. 2: Effect of drought on height of *Pongamia pinnata* showing trendline and regression graph with R²value

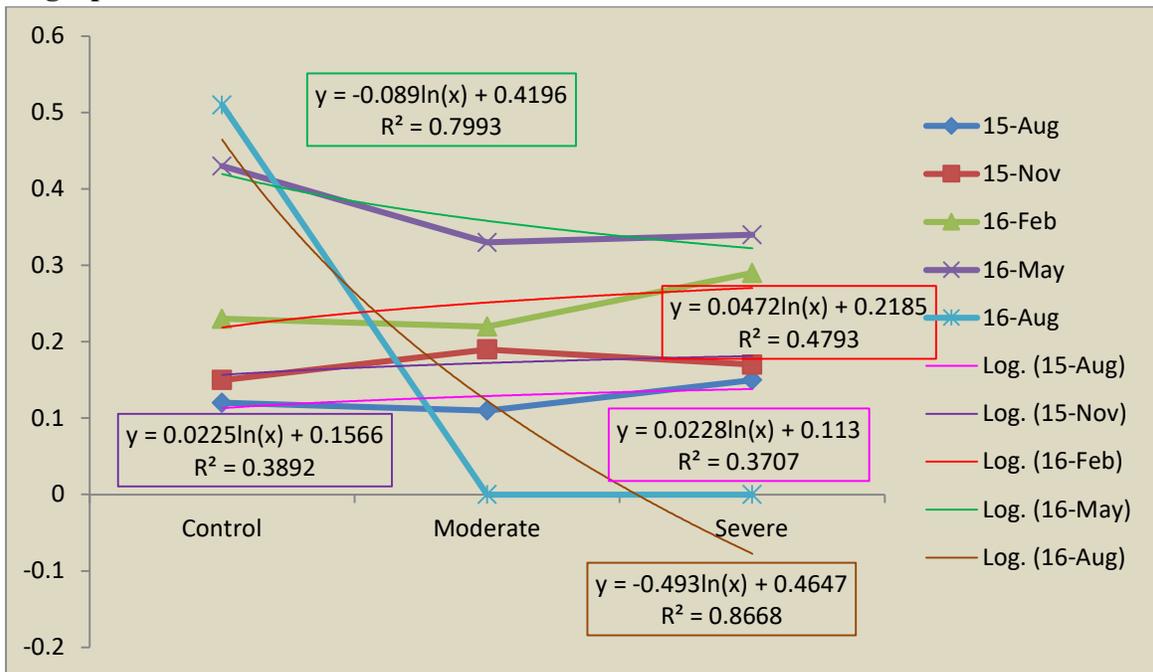


Fig. 3: Effect of drought on Collar diameter of *Dalbergia latifolia* showing trendline and regression graph with R²value

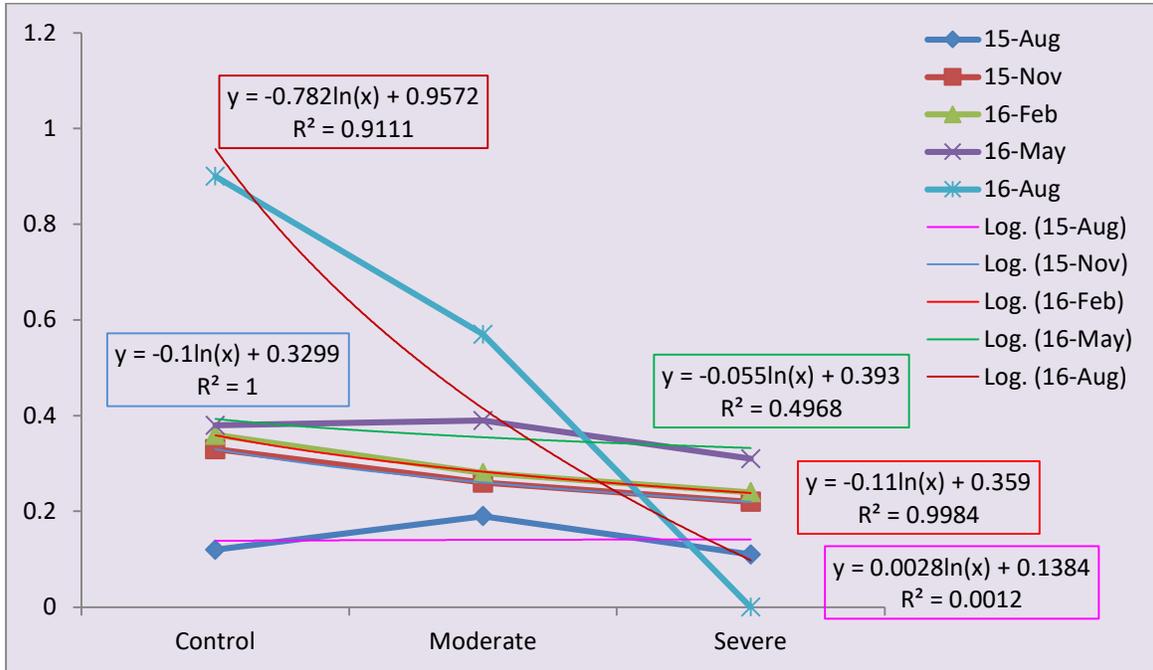


Fig. 4: Effect of drought on Collar diameter of *Pongamiapinnata* showing trendline and regression graph with R²value

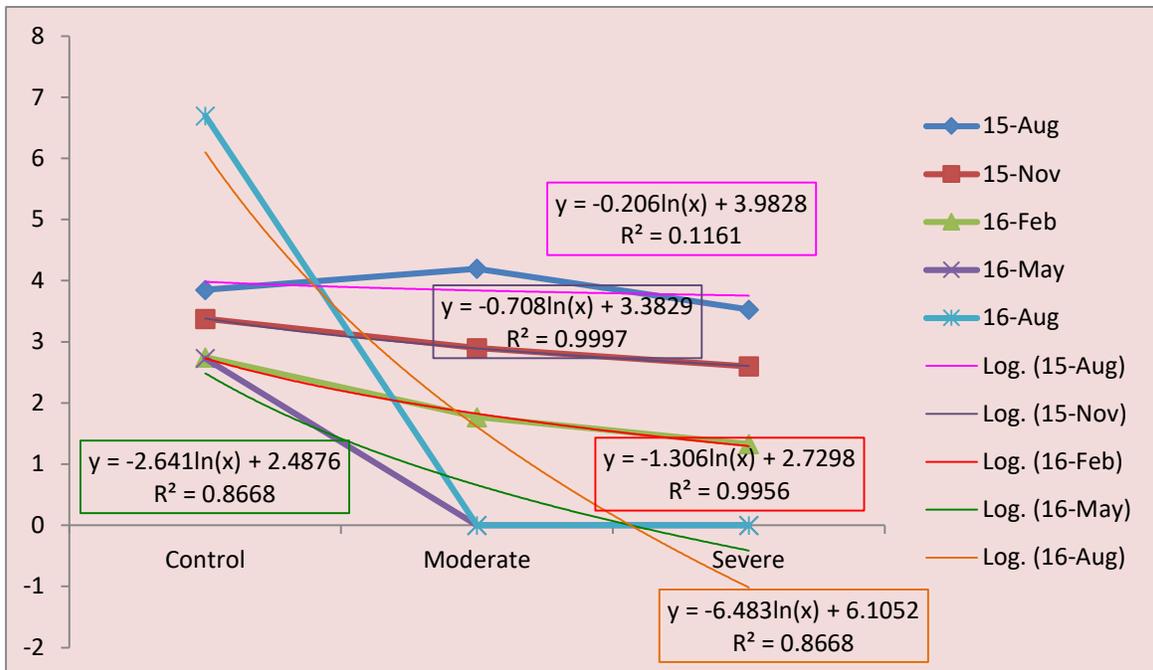


Fig. 5: Effect of drought on leaf size of *Dalbergialatifolia* showing trendline and regression graph with R²value

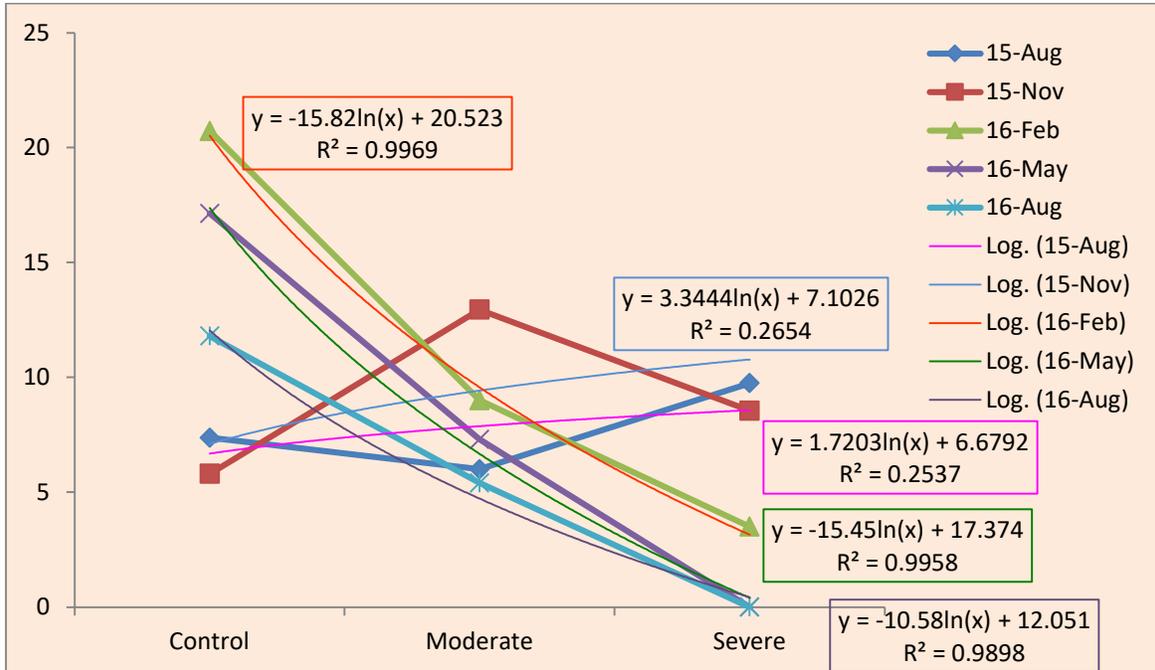


Fig. 6: Effect of drought on leaf size of *Pongamiapinnata* showing trendline and regression graph with R²value

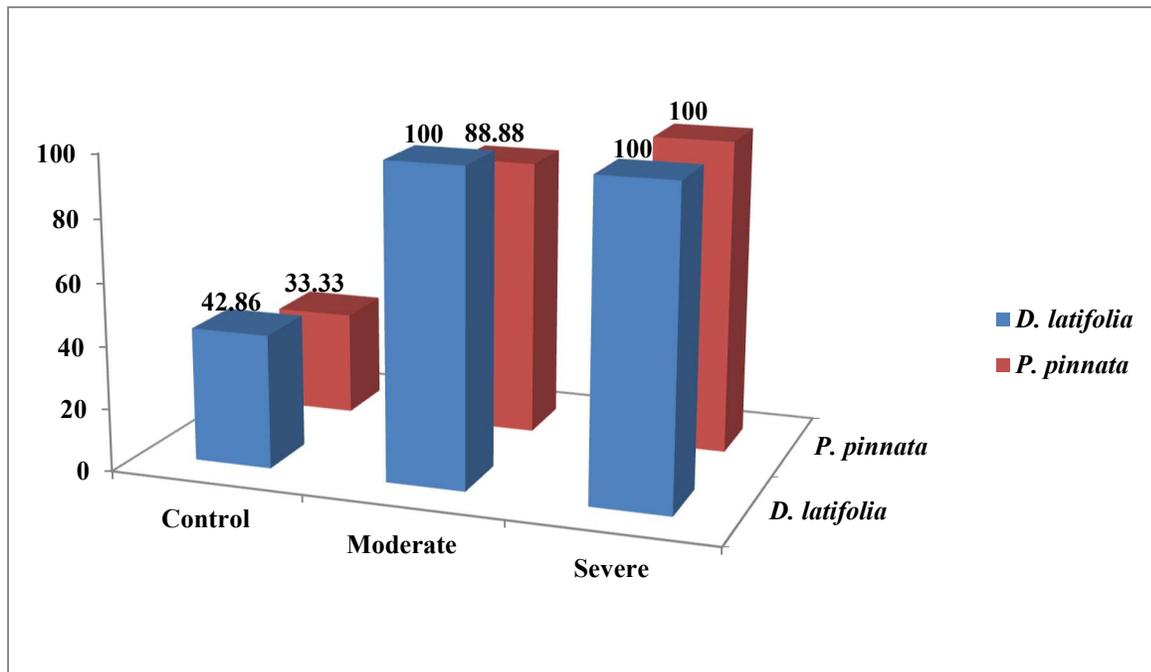


Fig. 7: Percent mortality of tree seedlings of *Dalbergialatifolia* and *Pongamiapinnata* under drought conditions

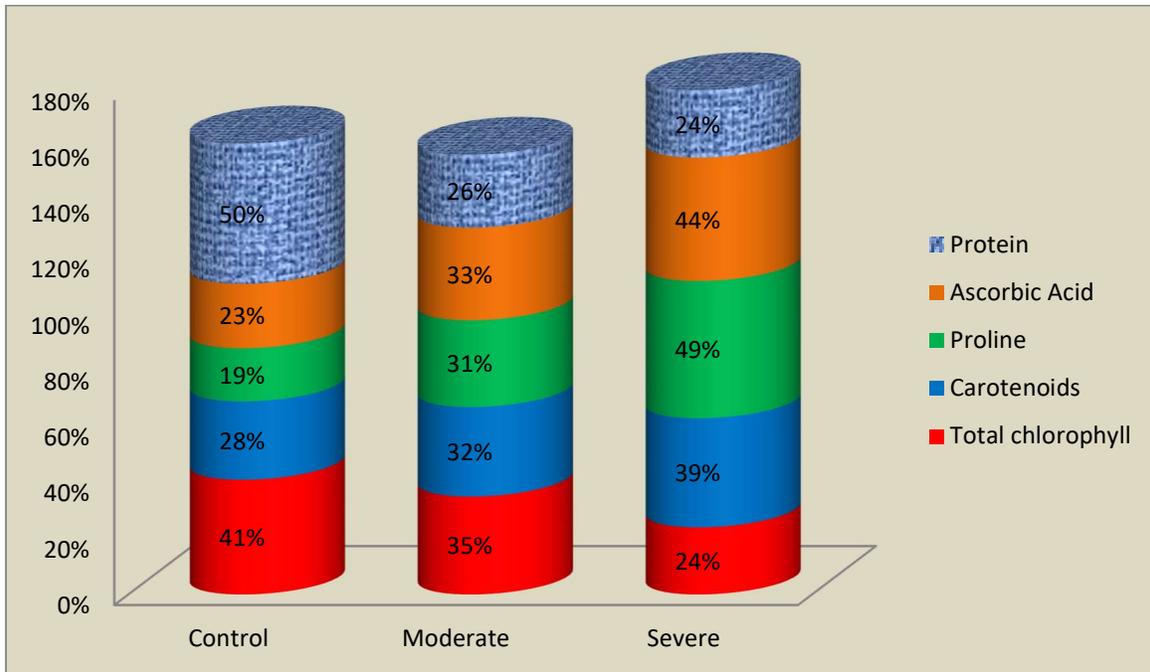


Fig. 8: Effect of drought on foliar biochemicals of *Dalbergialatifolia*

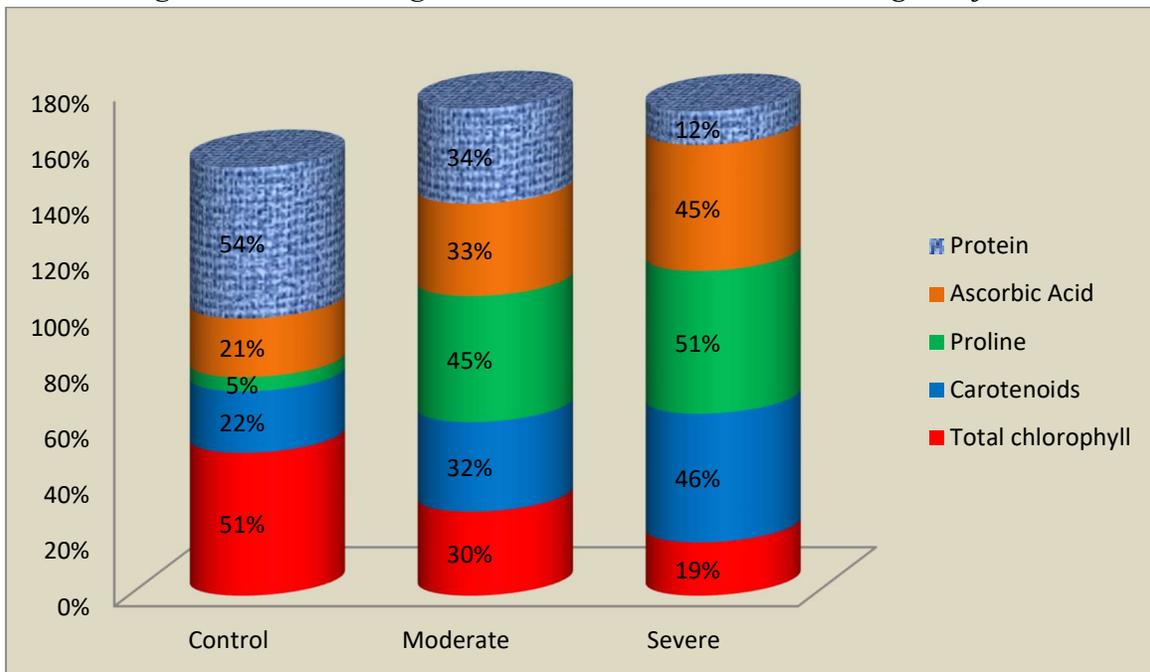


Fig. 9: Effect of drought on foliar biochemicals of *Pongamiapinnata*

Table 1: PWP, FC and Volume of Irrigation water

Species	PWP (%)	FC (%)	Quantity of calculated irrigation water (ml)
<i>D.latifolia</i>	10.89	43.23	864.60

<i>P.pinnata</i>	3.41	24.86	497.20
------------------	------	-------	--------

Table 2: CPE values and time period attaining species specific CPE.

Species	Drought Level	CPE (mm)	CPE (in days)		Irrigation according to CPE values (in days)	
			Summer	Winter	Summer	Winter
<i>Dalbergialatifolia</i>	Severe	60	8	20	7	19
	Moderate	30	4	11	3	10
<i>Pongamiapinnata</i>	Severe	80	15	31	14	30
	Moderate	40	7	15	6	14

Table 3: Biomass, carbon content and root-shoot ratio of seedlings

Treatment	Total Biomass (TB) (g)	Carbon Content (g)	Root/Shoot ratio
<i>Dalbergialatifolia</i>			
Control	8.39	4.20	0.45
Moderate	6.32	3.16	0.46
Severe	4.76	2.38	0.32
CD _{0.05}	1.213		
SE _±	0.301		
<i>Pongamiapinnata</i>			
Control	12.70	6.35	1.26
Moderate	3.89	1.94	1.47
Severe	1.36	0.68	1.27
CD _{0.05}	2.170		
SE _±	0.538		

Table 4: Photosynthetic Rate (Pn), Stomatal Conductance (C) and Transpiration Rate (E) of seedlings

Treatment	Pn (μmol/m ² /s)		C (mmol/m ² /s)		E (mmol/m ² /s)	
<i>Dalbergialatifolia</i>						
	January 16	July 16	January 16	July 16	January 16	July 16
Control	1.90	2.25	7.98	8.20	0.83	1.02
Moderate	1.78	-	7.56	-	0.32	-

Severe	1.07	-	5.10	-	0.20	-
<i>Pongamiapinnata</i>						
Control	2.54	3.05	11.90	12.10	0.92	1.10
Moderate	0.45	0.56	7.14	7.28	0.49	0.65
Severe	0.31	-	3.52	-	0.15	-

Table 5: Correlation study of *D. latifolia* species among the selected parameters.

		Morphological parameters				Physiological parameters			Biochemical parameters				
		H	CD	LS	CS	Pn	C	E	TC	CAR	P	AA	PT
Morphological parameters	H	1											
	CD	0.706*	1										
	LS	0.642	0.693*	1									
	CS	0.159	-0.078	0.089	1								
Physiological parameters	Pn	0.672*	0.306	0.782*	0.414	1							
	C	0.685*	0.300	0.714*	0.512	0.959**	1						
	E	0.780*	0.813**	0.920**	0.052	0.735*	0.700*	1					
Biochemical parameters	TC	0.602	0.374	0.736*	0.350	0.853**	0.906**	0.795*	1				
	CAR	-0.706*	0.493	0.652	0.586	0.812**	0.884**	0.652	0.735*	1			
	P	-0.784*	0.443	0.672*	0.615	0.900**	0.924**	0.731*	0.835**	0.898**	1		

	A	-	-	-	-	-	-	-	-	0.87	0.83	1	
	A	0.65	0.55	0.75	0.4	0.76	0.87	0.79	0.91	0**	5**		
		8	7	6*	57	6*	7**	4*	3**				
	P	0.60	0.59	0.73	0.2	0.72	0.59	0.76	0.58	-	-	-	1
	T	8	4	2*	57	6*	1	9*	2	0.62	0.75	0.5	
										3	5*	24	

Table 6: Correlation study of *P. pinnata* species among the selected parameters.

		Morphological parameters				Physiological parameters			Biochemical parameters				
		H	CD	LS	CS	Pn	C	E	TC	CAR	P	AA	PT
Morphological parameters	H	1											
	C	0.70	1										
	D	4*											
	L	0.85	0.79	1									
	S	5**	7*										
	C	0.61	0.89	0.75	1								
	S	7	2**	0*									
Physiological parameters	P	0.65	0.80	0.84	0.947	1							
	n	5	0**	8**	**								
	C	0.76	0.93	0.88	0.930	0.91	1						
	E	0.82	0.90	0.91	0.869	0.86	0.97	1					
	n	9**	7**	5**	**	8**	9**						
Biochemical parameters	T	0.68	0.73	0.91	0.626	0.73	0.72	0.74	1				
	C	-	-	-	-	-	-	-	-				
	A	0.59	0.77	0.75	0.734	0.74	0.90	0.89	0.5	1			
	R	8	3*	5*	*	2*	0**	7**	54				
	P	-	-	-	-	-	-	-	-	0.78	1		
		0.66	0.84	0.85	0.961	0.99	0.94	0.89	0.7	4*			
		8*	0**	2**	**	1**	2**	2**	21*				

	A	-	-	-	-	-	-	-	-	-	0.66	0.80	1	
	A	0.80 7**	0.84 6**	0.80 8**	0.802 **	0.77 7*	0.85 3**	0.82 2**	0.7 09*	0.66 9*	0.80 2**	1		
	P	0.74	0.86	0.89	0.757	0.80	0.90	0.90	0.8	-	-	-	-	1
	T	9*	9**	4**	*	5**	8**	4**	33* *	0.85 5**	0.83 3**	0.7 65*	1	

H - Height, CD - Collar Diameter, LS - leaves Size, CS - Carbon Stock, Pn - Photosynthetic rate, C - Stomatal Conductance, E - Transpiration rate, TC - Total Chlorophyll, CAR - Carotenoids, P – Proline, AA - Ascorbic Acid and PT – Protein.

** . Correlation is significant at the 0.01 level (2-tailed).

* . Correlation is significant at the 0.05 level (2-tailed).