



PHYSIOLOGICAL RESPONSES OF MULTIPURPOSE TREE SEEDLINGS TO INDUCED WATER STRESS

Rex Immanuel, R., M. Ganapathy, M. Thiruppathi, G.B. Sudhagar Rao and J. Nambi

Department of Agronomy, Faculty of Agriculture, Annamalai University, Annamalai nagar,

Tamilnadu, India – 608002

e-mail: rrximmanuel@gmail.com

Abstract

Adjustment in leaf water status parameter and chlorophyll stability can be important elements of a tree's response to continued water stress. Water stress affects plant growth in the vast semi-arid regions of North Eastern Coastal Agro-ecosystem of Tamil Nadu. The present study was aimed at investigating the effect of drought stress on the physiology of multipurpose tree seedlings. Six months old seedlings were grown in poly pots under controlled condition at Annamalai University Experimental Farm, Annamalinagar, Tamilnadu and were subjected to water stress for two weeks. Treatments were in a randomized block design replicated four times. The physiological parameters determined were chlorophyll stability index and relative water content of leaves by using standard methods. The results revealed that the water stress reduced the physiological parameters of the seedlings. The stronger stress tolerant capacity of *Acacia nilotica*, *Anacardium occidentale*, *Pongamia pinnata* and *Tamarindus indica* seedlings in terms of higher CSI and RWC showed that the seedlings can considerably improved the survival under water stress environments.

Key words: chlorophyll stability index, relative water content, seedling drought tolerance, stress tolerance.

Introduction

Climate change in recent decades leads to decrease in the annual rainfall and distribution in semi arid regions. In these regions drought is one of the most significant severe abiotic stress, which affects almost every phase of plant growth. Seedling drought resistance is imperative for the early establishment of plants under stress conditions. Seedlings subjected to severe water stress can show leaf injuries and even wilting without leaf abscission. Plants exposed to water stress adapt overall physiology to minimize the impact of water stress and maximize the efficiency of use of available water. Water stress affects multiple physiological parameters of plants including a reduction in chlorophyll content and leaf water content (Khayatnezhad *et al.*, 2011; Wang *et al.*, 2012).

The selection of drought tolerant trees is essential to improve survival during afforestation of drought stressed environments. Researchers have considerable capacity to evaluate trees for drought tolerance under field conditions at drought prone sites, but the evaluation of drought tolerance in the field is expensive, prolonged and often produces variable results due to site specific variations (Salvatori *et al.*, 2016). For selected drought tolerant tree species that show better performance under induced stress environments generally possess some unidentified physiological attributes. It is imperative for screening drought resistant species to identify the specific physiological

traits that improve adaptation to water limited environments.

Therefore, understanding the physiological basis of drought tolerance in tree seedlings are important to identify tolerant species, which can be used as selection criteria. The drought-resistant capacity is an integrated characteristic of plants under soil drought stress; it would be very hard to evaluate accurately the drought-resistant capacity of a plant species by the individual indices. Hence, chlorophyll stability index (CSI) and relative water content (RWC) were used to evaluate the drought resistance of MPT's. Dorcus and Vivekandan (1997) indicated that CSI is a significant drought resistance parameter for the stress tolerance capacity of plants.

The RWC of the leaves has been proposed as a better indicator of water stress of the plant (Rachmilevitch *et al.* 2006) and has been used to estimate plant water status in terms of cellular hydration under the possible effect of leaf water potential and osmotic adjustments (Ozkur *et al.* 2009). In the present study we aimed to study the effects of water stress, experimentally produced by water exclusion on the chlorophyll stability index and relative water content of sixteen multipurpose tree species (MPT's).

Materials and Methods

The experiment was set up under controlled conditions at Annamalai University Experimental Farm,

Annamalainagar, Tamil Nadu, India which is situated of a latitude 11°23' N and longitude 79°41'E. The location received an annual mean precipitation of 1350 mm of which 80 per cent received during North East monsoon (Oct - Dec). The mean temperature is 29.6°C with a relative humidity of 85 per cent.

The seeds of 16 tree species (T₁ - *Acacia auriculiformis*, T₂ - *Acacia ferruginea*, T₃ - *Acacia nilotica*, T₄ - *Ailanthus excelsa*, T₅ - *Albizia lebbbeck*, T₆ - *Albizia saman*, T₇ - *Anacardium occidentale*, T₈ - *Azardirachta indica*, T₉ - *Calophyllum inophyllum*, T₁₀ - *Casuarina equisetifolia*, T₁₁ - *Ceiba pentandra*, T₁₂ - *Eucalyptus tereticornis*, T₁₃ - *Leucaena leucocephala*, T₁₄ - *Pongamia pinnata*, T₁₅ - *Senna siamea*, T₁₆ - *Tamarindus indica*) were collected from forest department and planted in a nursery under a shade. Three months after germination, the uniformly grown seedlings were transplanted into 10 kg polythene pots. The composition of the soil mixture used as the rooting medium in the pots was in the ratio of 1: 1: 1 with sand, red earth, FYM, respectively. The polypots were perforated at the bottom to allow for proper drainage of water in order to avoid water logging.

Treatments commenced 12 weeks after transferring the seedlings to the polypots. The drought tolerance studies were conducted as per the method followed by Michelsen and Rosendahl (1990). During the first 12 weeks of the experiment, all the seedlings were grown under normal condition. The seedlings were watered in to saturation and then watering was withheld from half of the plants in 13th and 14th week to induce moisture stress for two weeks. Another half of the plants were maintained as control. After seventh day of drought treatment leaf samples were collected and subjected to estimating CSI based on the method suggested by Kaloyereas, (1958). Based on this criterion, species were classified in to highly tolerant (CSI % > 80), moderately tolerant (CSI % 70-80) and less tolerant (CSI % < 70).

In the same period Relative Water Content (RWC) of leaves were determined by following the method described by Barrs and Weatherley (1962). Fresh weight, turgid weight and dry weight data of leaf/needle discs were used for the determination of RWC. The experimental set up was a completely randomized design consisting of 16 tree species and three replications.

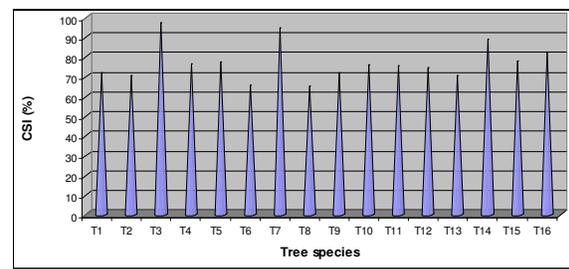
Results

Effect of water stress on the Chlorophyll Stability Index (CSI) of MPT's

The rate of CSI was higher in the well watered plants compared to the water stressed plants. After one

week of drought treatment, CSI of the leaves in all treatments were greatly reduced in response to water stress (Fig. 1). The CSI was higher in *Acacia nilotica* (97.2%), *Anacardium occidentale* (94.6%), *Pongamia pinnata* (88.5 %) and *Tamarindus indica* (82.1%). This was followed by *Senna siamea* (77.8 %), *Albizia lebbbeck* (77.0%), *Ailanthus excelsa* (76.2%), *Casuarina equisetifolia* (75.7 %), *Ceiba pentandra* (75.3%), *Eucalyptus tereticornis* (74.3 %), *Acacia auriculiformis* (72.0%), *Calophyllum inophyllum* (71.8%), *Leucaena leucocephala* (70.5%) and *Acacia ferruginea* (70.5%). The tree species *Albizia saman* (65.3%) and *Azardirachta indica* (64.6%) exhibited low CSI values.

Fig. 1: Influence of water stress on the Chlorophyll Stability Index (CSI) of MPT's

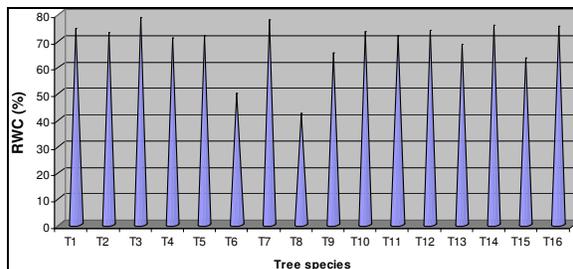


(T₁ - *Acacia auriculiformis*, T₂ - *Acacia ferruginea*, T₃ - *Acacia nilotica*, T₄ - *Ailanthus excelsa*, T₅ - *Albizia lebbbeck*, T₆ - *Albizia saman*, T₇ - *Anacardium occidentale*, T₈ - *Azardirachta indica*, T₉ - *Calophyllum inophyllum*, T₁₀ - *Casuarina equisetifolia*, T₁₁ - *Ceiba pentandra*, T₁₂ - *Eucalyptus tereticornis*, T₁₃ - *Leucaena leucocephala*, T₁₄ - *Pongamia pinnata*, T₁₅ - *Senna siamea*, T₁₆ - *Tamarindus indica*)

Effect of water stress on the Relative Water Content (RWC) of MPT's

The results for the effect of water stress on RWC of leaves are shown in Figure 2. The trend in RWC is almost similar to that of CSI. Significantly huge drop in RWC was observed in the stressed plants of all species due to the high water vapor pressure difference during the experiment. The results suggest that RWC of leaves was higher in *Acacia nilotica* (78.2 %), *Anacardium occidentale* (77.6 %), *Pongamia pinnata* (75.4 %) and *Tamarindus indica* (75.1 %). This was followed by *Acacia auriculiformis* (74.3 %), *Eucalyptus tereticornis* (73.6 %), *Casuarina equisetifolia* (73.2 %), *Acacia ferruginea* (72.8 %), *Ceiba pentandra* 71.6 %), *Albizia lebbbeck* (71.3 %), *Ailanthus excelsa* (70.6 %), *Leucaena leucocephala* (68.2 %), *Calophyllum inophyllum* (64.9 %) and *Senna siamea* (62.9 %). The lowest RWC value of 42.1 and 49.8 were recorded in *Azardirachta indica* and *Albizia saman*, respectively.

Fig. 2: Influence of water stress on the Relative Water Content (RWC) of MPT's



(T₁ - *Acacia auriculiformis*, T₂ - *Acacia ferruginea*, T₃ - *Acacia nilotica*, T₄ - *Ailanthus excelsa*, T₅ - *Albizia lebbbeck*, T₆ - *Albizia saman*, T₇ - *Anacardium occidentale*, T₈ - *Azardirachta indica*, T₉ - *Calophyllum inophyllum*, T₁₀ - *Casuarina equisetifolia*, T₁₁ - *Ceiba pentandra*, T₁₂ - *Eucalyptus tereticornis*, T₁₃ - *Leucaena leucocephala*, T₁₄ - *Pongamia pinnata*, T₁₅ - *Senna siamea*, T₁₆ - *Tamarindus indica*)

Discussion

Influence of water stress on the Chlorophyll Stability Index (CSI) of MPT's

The chlorophyll content affects the photosynthetic process of green plants. Higher CSI value of stressed *Acacia nilotica*, *Anacardium occidentale*, *Pongamia pinnata* and *Tamarindus indica* indicates that the drought stress did not have much effect on chlorophyll content of these plants. This might be the active adaptability to the drought environment in order to maintain the normal photosynthesis. The chlorophyll content of the tolerant plant has been observed to increase and remain unchanged under drought.

Under high temperature experienced in drought condition, the chlorophyll molecules of plants remained stable, thus its assimilate production remain undisturbed (Newcomb, 1999). This helps to withstand stress through better availability of chlorophyll, leads to increased photosynthetic rate and more dry matter production (Sivasubramanian, 1992). In general, the increase in CSI and/or chlorophyll accumulation is indicative of high efficiency of photosynthetic apparatus under extreme conditions (Farghali and Rayan, 2005)

In *Albizia saman* and *Azardirachta indica*, increased drought reduced the chlorophyll content and thus affects the photosynthetic rate leads to poor seedling growth and mortality of the seedlings in the early stages. The results of the study are in line with the reports of Dorcus and Vivekandan, (1997). Along with the increase of soil moisture stress the cell membrane started to be injured because of water loss, and the plasma would leak out, the normal physiological biochemistry pathways would be blocked, and the

chlorophyll contents would drop eventually (Tang and Zhao 2006).

Influence of water stress on the Relative Water Content (RWC) of MPT's

There higher RWC of *Acacia nilotica*, *Anacardium occidentale*, *Pongamia pinnata* and *Tamarindus indica* indicating that the growth of these species were not much influenced by soil drought stress, hence these species were classified as highly tolerant species. Following their high water loss rates from their leaves, the other species could have dehydrated earlier than the above said species. However, these four species persisted drought conditions much longer than the other species because of their rapid adaptive mechanisms. Under water stress, cells loose their turgidity causing low RWC in less tolerant plants. The RWC is usually higher in plants, which are adapted to dry conditions, and similar observations had been reported by other researchers. Jiregna *et al.* (2004) studied the response of two *Eucalyptus* species and three deciduous tree species to severe drought in summer and reported that *Eucalyptus* maintained higher RWC of leaf and is relatively drought resistant species.

Pandey *et al.* (1994) found that *Prosopis chilensis* maintains high RWC in leaves and grows successfully in dry regions. The drought tolerant experiments, suggest that MPT's growing in dry habitats has experienced selection favoring water conservative or drought surviving adaptations hence its importance in the rehabilitation of drought stressed environments. Wilting of some seedlings under water stress was observed after seventh day of drought stress treatment. This might be due to the plants were unable to absorb capillary water which was scarce and tightly held by the soil particles due to low soil water potential. In case of drought tolerant plants, the leaves maintained higher RWC and photosynthetic activities under water stress than that of drought sensitive genotype (Ananthi *et al.*, 2013).

Conclusion

The study has lined way to develop the most reliable parameter such as chlorophyll stability index and relative water content for screening drought tolerance in multipurpose tree species. Plants that were highly tolerant apparently had a higher photosynthetic rate than less tolerant, which received more water implying a possible resistance of the photosynthetic apparatus to water stress. The stronger stress tolerant capacity of *Acacia nilotica*, *Anacardium occidentale*, *Pongamia pinnata* and *Tamarindus indica* seedlings in terms of higher CSI and RWC showed that the seedlings can considerably improved the survival under water stress environments.

References

- Ananthi, K.; Vijayaraghavan, H.; Karuppaiya, M. and Anand, T. (2013). Drought-induced Changes in Chlorophyll Stability Index, Relative Water Content and Yield of Cotton Genotypes. *Insight Botany*, 3: 1-5.
- Barrs, H.D. and Weatherley, P.E. (1962). A re-examination of the relative turgidity technique for estimating water deficits in leaves. *Aust. J. Biol. Sci.* 15: 413-428.
- Dorcus, D. and Vivekanandan, M. (1997). Exploitation of mulberry genotypes for drought resistance potential. *J. Seric. Sci.*, 66(2): 71 – 80.
- Farghali, K.A. and Rayan, A.M. (2005). Chlorophyll stabilization in some succulent and non-succulent species inhabiting Kharga and Dakhla oasis in Egypt. *Assiut Uni. J. Botany*, 34(2): 391-409.
- Jiregna, G.; Andrey, R. and Negash, N. (2004). Response of seedlings of two *Eucalyptus* and three deciduous tree species from Ethiopia to severe water stress. *For. Eco. Manage.* 201: 119 – 129.
- Kaloyereas, S.A. (1958). A new method of determining drought resistance. *Plant Physiol.* **33**: 232 –233.
- Khayatnezhad, M.; Gholamin, R.; Jamaati-e-Somarin, S.H. and Zabihie M.R. (2011). The leaf chlorophyll content and stress resistance relationship considering in Corn cultivars (*Zea mays*). *Adv. Environ. Biol.*, 5(1): 118-122.
- Michelsen, A and Rosendahl, N. (1990). The effect of VA-Mycorrhizal fungi, phosphorus and drought stress on the growth of *Acacia nilotica* and *Leucaena leucocephala* seedlings. *Plant and Soil.*, 124: 7 – 13.
- Newcomb, W. (1999). Plant structure and development. In Dennis, D.T.; Layzell, D.B. and Turpin D.H. (eds) *Plant Metabolism* Longman, UK. pp. 257-261
- Ozkur, O.; Ozdemir, F.; Bor, M. and Turkan, I. (2009). Physiochemical and antioxidant responses of the perennial xerophyte *Capparis ovata* Desf. to drought. *Environ. Exp. Bot.* 66(3): 487-492.
- Pandey, A.N.; Rokad, M.V. and Thakarar, N.K. (1994). Root penetration and survival of *Prosopis chilensis* and *Dalbergia sissoo* in dry regions. *Proc. Indian Natn. Sci. Acad.* 60: 137-142.
- Rachmilevitch, S.; DaCosta, M. and Huang, B. (2006). Physiological and biochemical indicators for stress tolerance. In *Plant-environment interactions*, 3rd ed. Edited by B. Huang. CRC Press, Boca Raton, FL., 321–356.
- Salvatori, E.; Fusaro, L. and Manes, F. (2016). Chlorophyll fluorescence for phenotyping drought-stressed trees in a mixed deciduous forest. *Ann. Bot.*, 6: 39–49.
- Sivasubramaniawn, K. (1992). Chlorophyll stability index: methods for determining drought hardness of *Acacia* species. *Nitrogen Fix. Tree Res. Rep.*, 10: 111-112.
- Tang, T. and Zhao, L. (2006). Characteristics of water relations in seedling of *Machilus yunnanensis* and *Cinnamomum camphora* under soil drought condition. *J. For. Res.*, 17(4): 281-284
- Wang, W.F.; Peng, C.H.; Kneeshawdaniel, D.; Larocqueguy, R. and Luo Z. (2012). Drought-induced tree mortality: ecological consequences, causes, and modeling. *Environ. Rev.*, 20(2): 109–121.