

**DROUGHT RESPONNS IN PLUS TREES OF TEAK (*Tectona grandis* Linn. f.)
PROVENANCES OF KERALA**

By

MOHAMMED ALI ELTOUM HASSAN

(2017-17-014)

THESIS

**Submitted in partial fulfilment of the
requirement for the degree of**

MASTER OF SCIENCE IN FORESTRY

Faculty of Forestry

Kerala Agricultural University



DEPARTMENT OF FOREST BIOLOGY AND TREE IMPROVEMENT

COLLEGE OF FORESTRY

VELLANIKKARA, THRISSUR- 680 656

KERALA, INDIA

2020

DECLARATION

I hereby declare that the thesis entitled “**Drought response in plus trees of teak (*Tectona grandis* Linn. f.) provenances of Kerala**” is a bonafide record of research done by me during the course of research and that this thesis has not previously formed the basis for the award of any degree, diploma, fellowship or other similar title, of any other University or Society.

Vellanikkara

Date: 15/02/2020



Mohammed Ali Eltoum Hassan

(2017-17-014)

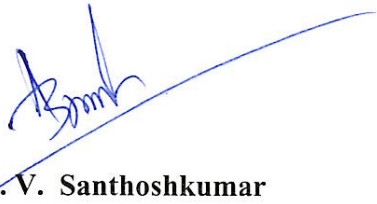
Dr. A. V. Santhoshkumar
Professor and Head
Department of Forest Biology &
Tree Improvement
College of Forestry
Kerala Agricultural University
Vellanikkara, Thrissur, Kerala

Date :

CERTIFICATE

Certified that the thesis, entitled “**Drought response in plus trees of teak (*Tectona grandis* Linn. f.) provenances of Kerala**” is a record of research work done independently by **Mohammed Ali Eltoum Hassan (2017-17-014)** under my guidance and supervision and that it is not previously formed the basis for the award of any degree, diploma, fellowship or associateship to him.

Vellanikkara


Dr. A. V. Santhoshkumar
Chairperson
Advisory Committee

CERTIFICATE

We, the undersigned members of the advisory committee of **Mr. Mohammed Ali Eltoum Hassan (2017-17-014)** a candidate for the degree of **Master of Science in Forestry** with major in Forest Biology and Tree Improvement, agree that this thesis entitled **“Drought response in plus trees of teak (*Tectona grandis* Linn. f.) provenances of Kerala”** may be submitted by him in partial fulfillment of the requirement for the degree.



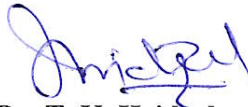
Dr. A. V. Santhoshkumar
Professor and Head
Department of Forest Biology &
Tree Improvement
College of Forestry
Kerala Agricultural University
Vellanikkara, Thrissur, Kerala
(Chairman)



Dr. E. V. Anoop
Professor & Head
Department of Forest products &
Utilization
Kerala Agricultural University
College of Forestry, Vellanikkara



Dr. C. M. Jijeesh
Assistant Professor
Department of Silviculture &
Agroforestry
Kerala Agricultural University
College of Forestry, Vellanikkara



Dr. T. K. Hrideek
Scientist, B
Department of Forest Genetics &
Tree Breeding
Kerala Forest Research Institute, Pechi

ACKNOWLEDGEMENT

*At the outset I thank **Almighty Allah** for his kind blessings bestowed on me in completion of this task,*

This thesis has been kept on track and been seen through to completion with the support and encouragement of numerous people including my well-wishers, my friends, colleagues and various institutions. At the end of my thesis I would like to thank all those people who made this thesis possible and an unforgettable experience for me. At the end of my thesis, it is a pleasant task to express my thanks to all those who contributed in many ways to the success of this study and made it an unforgettable experience for me.

*At this moment of accomplishment, first of all I pay homage to my major advisor, **Dr. A. V. Santhoshkumar**, Professor and head, Department of Forest Biology and Tree Improvement, College of Forestry, KAU, Vellanikkara for his valuable advice, constructive criticism, encouragement and his extensive discussions around my work. He has been a constant source of inspiration for me and has motivated me to take up the challenges of the research work. His flexibility in willing to accept any suggestions during the course of the research work was very helpful in coming up with interesting results during my work.*

*I extend my cordial thanks to **Dr. K. Vidyasagaran**, Dean, College of Forestry for extending the facilities available in the college for conducting the present study. I express my deep sense of gratitude to Kerala Agricultural University for the financial and technical support for pursuance of my research.*

*I feel privileged to express my gracious thanks to the respectable members of my advisory committee **Dr. T. K. Hrideek**, Scientist, KFRJ, Department of Forest Genetics and Tree Breeding, Peechi, **Dr. E. V. Anoop**, Professor and head, Department of Forest products & Utilization, College of Forestry, Vellanikkara, and **Dr. C. M. Jijeesh**, Assistant Professor, Department of Silviculture & Agroforestry, College of Forestry, Vellanikkara for their cooperation, intellectual advice and invaluable suggestions for the completion of my thesis.*

*With deep admiration I evince my heartfelt gratitude and unforgettable owe to my teachers **Dr. T. K. Kunhamu**, Professor and Head, Dept. of Silviculture and Agroforestry, **Dr. P. O. Nameer**, Professor and Head, Department of Wildlife Sciences and **Dr. V. Jamaludheen**, Professor, Department of Silviculture and Agroforestry, College of Forestry for their valuable support, inspiration, advise for my research works. Words are not enough to express my gratitude and respect for them. My deep sense of gratitude goes to **Dr. N. K Binu**, Assistant professors at College of Forestry for kindly providing timely advice and constant aid in a way of extending the facilities available in the department for conducting the present study.*

*I would like to convey my heartily thanks to **Ms. B. JUBY**, without her I may not complete the biochemical analysis part of my research works. I consider myself to be fortunate to work with her.*

I owe my sincere thanks to Kerala Forest Research Institute (KFRI), Department of Forest Genetics and Tree Breeding, Peechi for providing me accessions of teak plus trees.

*I owe my sincere thanks to **Dr. T. K. Kunhamu** Professor and head, Department of Silviculture & Agroforestry for providing greenhouse nursery to conduct my research experiment.*

*My sincere thanks to **Dr. J. S. Minimol**, Associate Professor, Cocoa Research Centre for giving me access to the laboratory and research facilities.*

*Special thanks to **Mr. B. R. Vishnu** who had been with me throughout my statistical analysis.*

*The help rendered by **Dr. Adarch** and **Ms. Raveena** during my research work is also remembered with immense gratitude. At this juncture, I would like to convey my special thanks, **my classmates, seniors and juniors** for their heartfelt moral support.*

I would like to state my full thanks to Indian Council for Cultural Relation (ICCR), African project for offering me this fellowship to carry out this postgraduate in Kerala Agricultural University.

*Whatever I am today, both in my professional and personal life, it's just because of the unconditional love and support of my family, My father **Dr. Ali Eltoum Hassan** and my*

mother Smt. Fatima berier Elhag gave me strength to face the challeges in life and gave advice to treat the life the way it comes almost modesty.

For all those whom I may forgetten I offer sincere apologies

Mohammed Ali Eltoum Hassan



Dedicated to my beloved
mother, father, brothers and sisters



CONTENTS

CHAPTER No.	TITLE	PAGE No.
1.	INTRODUCTION	1-2
2.	REVIEW OF LITERATURE	3-19
3.	MATERIALS AND METHODS	20-31
4.	RESULTS	32-82
5.	DISCUSSION	83-99
6.	SUMMARY	100-102
7.	REFERENCES	103-137
8.	ABSTRACT	138-139

LIST OF TABLES

Table No.	Title	Page No.
1.	Details of the plus tree accessions used in the study	20
2.	Biometric observations on plantlets of teak plus tree accessions in nursery condition at start of experiment.	34
3.	Growth parameters of plantlets of teak plus tree accessions at pre and post drought induction.	35
4.	Physiological parameters of plantlets of teak plus tree accessions under normal condition	38
5.	Physiological parameters of plantlets of teak plus tree accessions exposed to drought stress	41
6.	Physiological parameters of plantlets of teak plus tree accessions after rehydration	44
7.	Biochemical parameters of plantlets of teak plus tree accessions under normal condition	47
8.	Biochemical parameters of plantlets of teak plus tree accessions after drought induction	50
9.	Biochemical parameters of plantlets of teak plus tree accessions after re-watering	53
10.	Effect drought stress and their combination on various growth, physiological and biochemical parameters of teak plus tree accessions.	54
11.	Correlation and inter correlation of physiological and biochemical characters of plantlets of teak at drought condition	68
12.	Correlation and inter correlation of physiological and biochemical characters of plantlets of teak after rehydration	71
13.	Clustering of teak plus tree accessions based on physiological and biochemical characters under drought stress condition.	72
14.	Inter and intra cluster distances of the teak accessions under drought stress condition	73
15.	Clustering of teak accessions based on physiological and biochemical characters after rehydration	74
16.	Inter and intra cluster distances of the cluster diagram based on physiological and biochemical characters of teak accessions after rehydration	75
17.	Principal component analysis of physiological and biochemical characters of teak plus tree accessions at stress condition	77
18.	Principal component analysis of physiological and biochemical characters of teak plus tree accessions at after rehydration.	80
19.	Total score of biochemical responses of teak accessions during stress and recovery	82

LIST OF FIGURES

Fig. No.	Title	Page No.
1.	Change in relative water content and canopy air temperature differences among <i>Tectona grandis</i> plus tree accessions under water stress and rehydration	59
2.	Change in photosynthetic rate, stomata conductance and transpiration among <i>Tectona grandis</i> plus tree accessions under water stress and rehydration	60
3.	Change in photosynthetic pigments and chlorophyll stability index among <i>Tectona grandis</i> plus tree accessions under water stress and rehydration	61
4.	Change in total soluble protein and nitrate reductase among <i>Tectona grandis</i> plus tree accessions under water stress and rehydration	62
5.	Change in free amino acid, proline, glycine betaine and total soluble sugar among <i>Tectona grandis</i> plus tree accessions under water stress and rehydration	63
6.	Change in activity of super oxide dismutase and peoxidase among <i>Tectona grandis</i> plus tree accessions under water stress and rehydration	64
7.	Change in content of malondialdehyde and cell membrane stability index among <i>Tectona grandis</i> plus tree accessions under water stress and rehydration	65
8.	Dendrogram based on on physiological and biochemical attributes in <i>Tectona grandis</i> plus tree accessions under moisture stress condition	73
9.	Dendrogram of teak plus tree accessions based on physiological and biochemical characters after rehydration	75
10.	Loading plot of physiological and biochemical characters of <i>Tectona grandis</i> plus tree accessions at drought condition	78
11.	Loading plot of physiological and biochemical characters of <i>Tectona grandis</i> plus tree accessions after rehydration	81

LIST OF PLATES

Plate No.	Title	Between Pages
1.	One year old teak seedlings in nursery	21-22
2.	Teak seedlings in nursery after three months	21-22
3.	Teak seedlings in nursery after six months	21-22
4.	Views of plantlets of teak plus tree accessions during drought stress and after rehydration	21-22
5.	Recording observation on biometric parameters	22
6.	Recording the physiological parameters	25-26
7.	Extraction of biochemical components	30
8.	Equipments used for the biochemical analysis	30-31
9.	Estimation of biochemical components	30-31
10.	Estimation of antioxidant enzymes and malondialdehyde	30-31
11.	Preparation of standard curves for analysis of biochemical parameter	30-31



INTRODUCTION



INTRODUCTION

Teak (*Tectona grandis* Linn. f.), a member of family Lamiaceae, is a deciduous forest tree species which is considered as one of the noblest timber trees in the world due to its golden hue, wonderful texture and durability (Keogh 1979; 2009). Teak forests distributed naturally in Asia-Pacific region cover an area of 29,035 million ha in India, Lao, Myanmar and Thailand (Kollert and Cherubini, 2012). Due to its economic importance, teak has been introduced widely in the tropical regions since 19th century especially in Asia, Africa, Central America, and South America (Alcantara, 2013).

The area of planted teak forests is estimated to be 4.3 - 6.8 million ha in a total of 70 countries, of which 83 per cent are in Asia, 11 per cent in Africa, and 6 per cent in tropical America (Kollert and Kleine, 2017; Midgley *et al.*, 2017). India is one of the major teak producing countries and contains large genetic variability. Presently, 1.5 million ha of teak plantations exist in India and around 50,000 ha of teak plantations are raised annually (Subramanian, 2000). The plantations mainly exist in states of Kerala, Karnataka, Tamil Nadu, Andhra Pradesh, Maharashtra, Madhya Pradesh, Uttar Pradesh, West Bengal, Andaman and Assam (Sreekanth and Balasundaran, 2013). Teak plantations cover over 70,000 hectares of the State's total plantation area in Kerala, mainly in Nilambur, Wayanad, Ranni, Konni, Chalakkudy, Thrissur and Palakkad (Surendran, 2014).

Teak establishes over a wide range of climatic conditions, from very dry localities with an annual rainfall as low as 500 mm to very moist areas with annual rainfall up to 5000 mm per year (Seth and Khan, 1958). However, teak appears to avoid extreme dry and moist conditions (Saxe *et al.*, 2001). The natural distribution of teak in wide range of climatic conditions and edaphic zones has led to development of variance among ecotypes during the processes of evolution. The genetic components of this variation can be identified by testing and exploiting the superior populations of plant material to improve yield (Dupuy and Verhaegen, 1993).

Like many other plant species that grow in tropical environments, teak plantations are also exposed to long periods of drought stress. Consequently, a wide variation in the performance of different ecotypes has been previously recognized (Keogh, 1982; White, 1993). Water stress affects the most important determinants of yield-canopy architecture, photosynthesis and partitioning of assimilates.

The threat of global warming will increase the occurrence of drought in tropical regions and vulnerability of plants towards drought may lead to modification in growth, photosynthesis and anti-oxidative defense mechanism in various plant species. Drought affects morphological, physio-biochemical and molecular processes in plants which results in growth inhibition, stomata closure with consecutive reduction of transpiration, decrease in chlorophyll content and inhibition of photosynthesis, protein and nitrogen metabolism and differential responses in anti-oxidative enzymes to cope with the osmotic changes in their tissues (Chaves *et al.*, 2009). Efforts are to be made to find genotypes which are tolerant to drought stress. Evaluation of genotypes with high yield which can tolerate stress and understanding the associated characters are essential for improving crop productivity under drought stress conditions.

The present study attempts to identify the physiological and metabolic responses among plus trees of teak from various provenances of Kerala exposed to drought stress in order to identify the superior germplasm suitable for breeding programs for drought tolerance. The study was formulated with following objectives:

- 1) To evaluate the variability of drought stress and drought recovery on morphological, physiological and biochemical responses among plus trees of teak in different provenances of Kerala.

- 2) To study the responses of physiological and biochemical characters during drought stress and drought recovery in teak.



REVIEW OF LITERATURE



REVIEW OF LITERATURE

2.1 GENETIC IMPROVEMENT OF TEAK

Among the tropical woody trees, considerable work with regard to genetic improvement in teak has been done in India. Laurie (1938) stressed the significance of establishing tree plantation from good seed source. Sen Gupta (1939) clarified the superiority of seeds of local origin compared to the seeds from region outside the natural range of teak. He tested teak seeds from different origins and found that seeds of Nilambur origin grow better in dry zones compared to seeds of other origin. Similar observations were made by Kadambi in 1945. Egenti (1977) observed existence of variation in vigour and form of different teak seedlings among various provenances, where he noticed a clear difference in branching habit and foliage between provenances from India and other countries. Ferguson (1938) mentioned that the factors controlling stem form and branching in teak is constant for an individual tree. He pointed out that the use of seeds from selected trees is the only way to improve the quality of the stem.

An early attempt was made to improve the planting stock of teak when the first teak plus tree selection was done by Kedharnath and Mathew in 1961. Many authors reported that the plus trees selection and the regeneration of these selections through grafting could be the perfect methodology as the initial stage of tree improvement programme (Kedharnath and Matthews, 1962). Jones (1970) mentioned the common defects in the tree form that should be avoided during plus trees selection. Various tree characters like vigour, height, girth, fibril angle, fluting, and buttressing were reported to be heritable. Based on this knowledge, action programmes were started regarding plus trees selection, seed stands, clonal seed orchard establishment, plus trees evaluation and elite seed orchard establishment. As a consequence, selection of plus tree, seed stands and clonal seed orchard establishment progressed well in India. Kedharnath *et al.* (1969) have also clarified that considerable genetic gain can be obtained through selection since various tree characters are heritable. Keiding (1966) proposed the

selection of seed production areas for seed collection to provide a temporal source until seed orchards produce seeds sufficiently. He suggested a procedure for the establishment of seed production area where the selected seed stands should be superior to the surrounding stands and are marked, in which all the undesirable trees are removed while retaining only the good trees.

2.2 VARIATION IN TEAK PLUS TREES

Plus trees may be defined as outstanding individuals occurring in natural stands or in aged plantations having a number of desirable features. It is to be expected that such trees occur in low frequency and may be hard to find. But they do exist. These trees form the foundation for tree improvement by selection.

Progeny evaluation is one of the selection methods followed in tree improvement programs where superior genotypes are selected based on the performance of their respective progenies at an early age by providing similar environmental (growth) conditions (Wright, 1976). Individuals selected through this method are known to be superior with respect to their genetic characters. Hence, plus trees are generally graded as elite types based on progeny performance in progeny trials (Vasav *et al.*, 2011).

Kedharnath (1980) illustrated the important progress made in India for the selection of plus trees and breeding work in *Tectona grandis* with the mention of availability of overall 700 plus trees in various states since 1960. Till now more emphasis in teak plus tree selection has been given to vigour, form and crown of the selected tree in comparison to its check tree. This contributes to the yield in quantity of wood production of future plantations. A new approach has been undertaken by the authors who have come across the trees of *Tectona grandis* for different purposes other than yield or quantity of wood like resistance to adverse condition and resistant to important diseases and insect pests (Kedharnath, 1982).

As per Mathew's guidelines in 1961, Coimbatore gene bank was established in Dehradun and was organized by planner in Forestry Research. In

later years, variability was observed between plus trees established in Dehradun. Teak is reported to be a highly frost susceptible species and was found to be difficult to introduce to the frost belt of Northern India. One frost resistant clone with good germination was identified. Along with this, a clone for better growth has also been isolated. These two strains were raised in the campus of Forest Research Institute and yearly observations on various aspects have been recorded. The work has been taken over to the level of establishment of small seed orchards for these two strains with the objective of combining frost resistant and good growth traits in the genetically improved variety for large scale plantation in frost prone regions of the country (Vakshasya *et al.*, 1988).

Observations were recorded over the years on the relative resistance/susceptibility of the various teak clones under natural conditions to two leaf infecting fungi in the clonal seed orchard at New Forest, Dehradun and the germ plasm bank. The infection exhibited steady reaction where some clones showed complete resistance, some clones showed high susceptibility while some clone showed moderate resistance. However, both fungi diseases were not economically important (Kedharnath and Pratap-Singh, 1975).

2.3 DROUGHT AND RECOVERY RESPONSES IN WOODY PLANT

Recent experiences of extreme climate conditions results in forest decline (Choat *et al.*, 2018). Globally, drought is one of the major reasons of increasing tree mortality (Allen *et al.*, 2015). Climate change scenarios may intensify frequency and severity of the drought in the near future (Wassmann *et al.*, 2009).

Plants evolves the following strategies to tolerate water deficit: (1) drought avoidance by reducing water loss via rapid stomatal closure and also leaf area or canopy cover reduction (Yu *et al.*, 2017); (2) drought tolerance through adjusting the osmoregulatory molecules and increasing the cell wall elasticity to maintain the cell turgor pressure (Yi *et al.*, 2016); (3) drought escape through completing their life cycle by avoiding severe water limitation during growth production (Izanloo *et al.*, 2008), and (4) drought recovery by resuming the growth and

yielding gain after water stress-induced which causes a complete loss of cell turgor pressure and leaf dehydration (luo, 2010). Plants are able to employ these strategies consecutively or simultaneously in response to drought stress.

Considerable damage in many plant functions might be induced by drought stress. Reduction in carbon assimilation is the main effect which in turn results in imbalance of excited and utilized electrons through photosynthesis, leading to generation of reactive oxygen species (Benhassaine-Kesri *et al.*, 2002). This cause oxidative stress by damaging cell membrane, protein and nucleic acids (Benson *et al.*, 2004). Intercellular concentration of malondialdehyde shows the degree of oxidative stress extension (Bernacchi *et al.*, 2002).

To overcome stress, plants possess enzymatic and non-enzymatic activities mechanism to detoxify reactive oxygen species (ROS) like super oxide dismutase, peroxidase and catalase (Bernacchi, 2002). In order to maintain cellular function plants may alter their water relations (Bai *et al.*, 2008). Plants show osmoregulatory adjustment through synthesizing and accumulating convenient solutes like free amino acids, sugar, and proline (Ethier and Livingston, 2004). Adjusting the osmoregulation gives the plant an opportunity to maintain metabolic functions through maintaining turgor pressure and cell volume at low water potential (Bai *et al.*, 2008; Souza *et al.*, 2004). This may affect growth and production of the plant differentially relying on various variables like the stress length, the plant vegetative status, and the existence of other adverse environments such as high temperatures and high light irradiance (Kaiser, 1987).

Stress severity along with specific physiological changes like stomatal conductance, photosystem II down-regulation, conductance loss in xylem and outside-xylem tissues clarify whether stress-effects are permanent or speedily reversible (Miyashita *et al.*, 2005). This changes can be utilized an index for stress-recovery (Ruehr *et al.*, 2019). This, in turn, lead to three stress phases related to different recovery: (1) Mild stress-fast recovery where hydraulic tension increases which first affects stomatal conductance, but does not drop for

prolonged time beneath critical levels (near to stomatal closure) and loss of xylem conductance does not happen then responses should be fast and completely reversible (Ameye *et al.*, 2012) (2) Moderate stress-delayed recovery: Exposure to higher temperature ($>40^{\circ}\text{C}$) can cause PSII inhibition and results in tissue necrosis which typically delays recovery (Huve *et al.*, 2011; Curtis *et al.*, 2014). (3) Severe stress-impaired recovery: If the stress dosage is further increasing, non-reversible tissue damage develops. Continued high temperature stress will result in persisting leaf damages, while desiccation will ultimately result in massive hydraulic conductance in both outside-xylem and xylem tissues (Brodribb *et al.*, 2010; Li *et al.*, 2015). In addition, osmotic adjustment facilitates the recovery of metabolic activities after relief from stress (Souza *et al.*, 2004).

Moreover, if drought frequency increases and the time between stress periods decrease, recovery rate might determine survival (Schwalm *et al.*, 2017). Recovery of different physiological processes over time is typically measured relative to a control treatment or to pre-stress conditions and generally falls into three broad categories: (1) Complete recovery occur with and without repair mechanisms involved (Xu *et al.*, 2010). (2) Partial recovery where damaged tissues may not be fully restored. (3) Compensatory recovery where stress-induced reductions are recompensed by investments into alternative processes and tissues (Cano *et al.*, 2014). In some cases, overcompensation occurs, as has been shown in previously drought-stressed trees growing taller than control trees (O'Brien *et al.*, 2017).

2.4 IMPACT OF DROUGHT STRESS

2.4.1 Effects of drought stress on morphological characteristics.

The success of plantation in the field is determined by seedling growth characters which are influenced by both the genotype and environment. When influences of genetic components are greater, it leads to higher characteristic variations, which is more effective and better for selection through provenance trial. It has been observed that maternal effects exert greatest influence on the

growth of seedlings which diminishes with age, and not likely to be correlated with the performance of the individuals at a mature stage (O'Brien *et al.*, 2017).

A study conducted by Rawat and Bakshi (2011) clarified that coniferous species had significant variation between provenances like many tree species. However, effect of provenance in the seedling traits need not always be reflected in the later stages of growth.

Dlamini (2010) observed high significant differences for seedling height and root collar diameter between provenances at five and eight months old seedlings of *Sclerocarya birrea*. However, at eight months non-significant variations were depicted in height and root collar diameter increment percentage indicating that the best seedlings at the age of eight months are not necessarily the one that were growing best at five months.

Jayasankar *et al.* (1999) observed high variations in performance of teak seedlings from seven teak provenances. The variations were appeared to be under genetic control. They also suggested grading of seedlings at an early age based on their height and collar diameter as the provenances performance in the field follow nursery growth patterns.

Xiao and Zhiling (2012) studied fifteen provenances of *Magnolia officinalis* trees and observed a considerable variation among the provenances in shoot length, collar diameter, number of leaves per plant, main root length and dry weights. They found that provenance effect contributed 93 per cent of the total variation in seedling height and collar diameter. This indicates that adequate genetic variability for seedling height exists for the species and can be used for early selection of provenance. Egenti (1977) reported the differences in branching habit between Indian teak provenances and others.

It has been established that drought stress is a very important limiting factor at the initial phase of plant growth and establishment. It affects both elongation and expansion growth (Anjum *et al.*, 2003; Bhatt and Rao, 2005; Kusaka *et al.*, 2005; Shao *et al.*, 2008).

Zou et al. (2007) suggested that genotypes with drought resistance can be identified by measuring yield potential, delay in flowering, and reduction in plant height under well-watered and drought stressed test environments. Genotypes may differ in their recovery growth after vegetative stage drought. This might be related to the amount of leaf that remained after drought (Mitchell *et al.*, 1998).

2.4.2 Effects of drought stress on physiological parameters

2.4.2.1 Relative water content

Relative water content (RWC) is probably the most appropriate measure of plant water status in terms of the physiological consequence of cellular water deficit. It expresses the relative amount of water present in the plant tissues. A decrease in the relative water content in response to drought stress has been noted in wide variety of plants when leaves were subjected to drought (Nayyar and Gupta, 2006).

When two poplar species cuttings were subjected to progressive drought stress, relative water content decreased to 23.3 per cent in *Populus cathayana*, while in *Populus kangdingensis* it was 16 per cent. Relative water content was affected by the interaction of severity, duration of the drought event and species (Yang and Miao, 2010).

According to Husen (2010) the relative water content (RWC) of two teak clones (FG1 and FG11) decreased significantly when subjected to progressive drought of 5, 10, 15 and 20 days of withdrawing irrigation. Clone FG1 showed RWC of 67.75 per cent, 51.17 per cent, 43.89 per cent and 35.46 per cent respectively, whereas it was 60.53 per cent, 48.65 per cent, 40.10 per cent and 33.49 per cent respectively in clone FG11. After five days of re-watering FG1 showed values comparable to FG11.

2.4.2.2 Canopy air temperature differences

Measurements of canopy air temperature differences (CATD) have been widely used in recent years to study genotypic response to drought. CATD has been used to assess plant water status because it represents an overall, integrated physiological response to drought and high temperature (Amani *et al.*, 1996).

Leaf water potential, leaf rolling, leaf drying, canopy temperature and delay in flowering time can reflect the internal plant water status under water stress, and these traits can be considered as integrative traits to identify drought resistant genotypes (Pantuwan *et al.*, 2002; Jongdee *et al.*, 2002).

2.4.2.3 Photosynthetic rate

The ability of crop plants to acclimatize to different environments is directly or indirectly related with the plant's ability to acclimatize at the level of photosynthesis (Allen and Ort, 2001), which in turn affects biochemical and physiological processes and consequently, the growth and yield of the whole plant (Chandra, 2003).

Due to stomatal closure and reduced CO₂ assimilation, drought reduces the amount of available C within the plant (Allen and Ort, 2001; Gollan *et al.*, 1992). Also the allocation of assimilated C to different plant organs is affected, resulting in retardation of growth under drought (Ruehr *et al.*, 2009).

Husen (2010) found that inducing 20 days treatment over FGI and FG11 teak seedling clones led to decrease in net photosynthetic rate by 69.22 per cent and 74.25 per cent respectively. While during recovery both teak clones showed incomplete recovery compared to control (8.87 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ and 8.66 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ respectively).

2.4.2.4 Stomatal conductance

Stomatal conductance reduces transpiration and plays essential role in regulating plant water balance. Stomata closure also reduces cell expansion and

growth rate leading to a significant reduction in biomass and yield (Nemeskeri *et al.*, 2015; Rauf *et al.*, 2016). Many scientists believe that the first reaction of plants to severe drought is the closure of their stomata to prevent the water loss via transpiration (Mansfield and Atkinson, 1990; Berry *et al.*, 2010; Casson and Hetherington, 2010; Brodribb and Mc-Adam, 2011; Torres-Ruiz *et al.*, 2013; Nemeskeri *et al.*, 2015).

Stomatal conductance decreased by 82.56 per cent and 86.21 per cent respectively in FG1 and FG11 teak clones imposed with drought for 20 days. After five days of re-watering the clones did not recover fully. They were showing stomatal conductance as $0.145 \text{ mol m}^{-2} \text{ s}^{-1}$ and $0.123 \text{ mol m}^{-2} \text{ s}^{-1}$ respectively after re-watering compared to control $0.195 \text{ mol m}^{-2} \text{ s}^{-1}$ and $0.20 \text{ mol m}^{-2} \text{ s}^{-1}$ respectively (Husen 2010).

2.4.2.5 Transpiration

Transpiration rate plays a major role in estimating drought tolerance of plants. Water stress has been known to reduce the transpiration rate in plants as confirmed by many authors like Gartner *et al.* (2009), who found that birch and Norway spruce trees reduced their transpiration in response to drought. Similar findings were observed (Seiler and Johnson, 1985) in one year loblolly pine (*Pinus taeda* L.) seedling which reduced 30 per cent of transpiration rate when induced to drought.

Husen (2010) observed significant inhibition in transpiration rate of two clones of teak (FG1 and FG11) exposed to progressive drought treatment (5, 10, 15 and 20 days). Transpiration rate in FG1 was $1.93 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$, $1.09 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$, $0.97 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and $0.59 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ respectively compared to control $2.12 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ while in FG11 it was $1.91 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$, $1.05 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$, $0.88 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and $0.56 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ respectively. In control it was $2.10 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$. Complete recovery was shown by both the clones when irrigation was resumed

for 5 days with the rate transpiration of $2.03 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and $2.04 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ respectively.

2.4.2.6 Photosynthetic pigments

Photosynthetic pigments are important to plants mainly for harvesting light and production of reducing powers. Severe drought stress inhibits the photosynthesis of plants by causing changes in chlorophyll content, by affecting chlorophyll components and by damaging the photosynthetic apparatus (Iturbe-Ormaetxe *et al.*, 1998). Drought stress caused changes in chlorophyll 'a' and 'b' ratios and carotenoids (Farooq *et al.*, 2009). The chlorophyll decreases mainly because of damaging of chloroplasts by active oxygen species (Smirnov, 1993). Ommen *et al.* (1999) reported that leaf chlorophyll content decreases as a result of drought stress. Farooq *et al.* (2009) reported that both the chlorophyll 'a' and 'b' are sensitive to water deficit.

Excessive drought resulted in large decline in chlorophyll a, chlorophyll b, and total chlorophyll content in teak clones FG1 and FG11. While both clones showed complete recovery in chlorophyll b and total chlorophyll after rehydration for 5 days. FGI1 showed partial recovery in chlorophyll a compared to control (Husen, 2010).

The chlorophyll content decreased to a significant level at higher water stress conditions in young apple tree (Wang *et al.*, 2018)

2.4.2.7 Chlorophyll stability index

High chlorophyll stability index helps the plant to withstand stress conditions through better availability of chlorophyll. This results in increased photosynthetic rate and high dry matter production (Mohan *et al.*, 2000).

2.4.2.8 Membrane stability index

Cell membrane stability is a major physiological index used for the evaluation of drought tolerance (Premachandra *et al.*, 1992). Biological

membranes are the first target of many abiotic stresses and it is generally accepted that the maintenance of integrity and stability of membranes under water stress is a major component of drought tolerance in plants (Bajji *et al.*, 2001). Dhanda *et al.* (2004), in their work displayed that membrane stability of leaves was the most important trait to screen the germplasm for drought tolerance.

A study done by Husen (2010) in two teak clones showed that membrane stability increased by 72.29 per cent in FG1 and, while in FG11 it was 89.05 per cent when subjected to water stress for 20 days. However, after re-watering for 5 days, clones of FG11 did not show complete recovery as compared to FG1 and control.

In another study conducted on *Populus* species by Pelah *et al.* in 1997, it was found that water stress led to an increase of membrane stability. However, the membrane stability of *Populus popularis* leaves were significantly lower than those of *Populus tomentosa*.

2.4.3 Effects of drought stress on biochemical parameters

2.4.3.1 Total soluble protein content

Variation in total soluble protein content (TSP) is an essential part of plant response to environmental stress as well as for plants adaptation to changes in environmental conditions (Vierstra, 1993; Hieng *et al.*, 2004). Pierre and Savoure (1990) and Roy-Macauley *et al.* (1992) showed that TSP got decreased when plants were exposed to drought. However, studies by Todd and Basler (1965) showed that TSP had no influence on plants exposed to water stress. Husen (2010) noticed that after 20 days of inducing water stress there was increase in TSP content by 39.27 per cent and 38.81 per cent, respectively, in two teak clones (FG1 and FG11) Five days of rehydration did not resulted in complete recovery of the plants.

2.4.3.2 Nitrate reductase activity

Nitrate reductase activity (NRA) is one of the most sensitive enzyme to water stress (Ferrario-Mery *et al.*, 1998 ; Foyer, 1998) and gives a good estimate of the nitrogen status of the plant and is very often associated with growth and yield (Srivastava, 1980). It catalyzes the NO_3 to NO_2 - reduction and is, considered as a limiting step for conversion of nitrate-N to amino acids and so for protein synthesis (Ferrario-Mery *et al.*, 1998). In higher plants, cytosolic NAD(P)H-nitrate reductase (NRA) is rapidly modulated by environmental conditions such as light, CO_2 , or oxygen availability. Photosynthesis activates 60-80 per cent of NRA in leaves whereas after stomatal closure, leaf NRA is inactivated down to 20 or 40 per cent of its maximum activity (Kaiser *et al.*, 1999).

2.4.3.3 Accumulation of osmoregulators

Among the various mechanisms used by plants to reduce the negative effects of water stress, many plant species accumulate soluble organic compounds, such as osmoregulators. This process is known as osmotic adjustment and it is considered as an important tolerance-mechanism, which allows the maintenance of cellular turgor and favors the absorption of water (White *et al.*, 2000; Chaves *et al.*, 2003). In this process, the biosynthesis and accumulation of non-toxic molecules of low-molecular weight in the vacuole and cytosol, such as inorganic ions, free amino acids, proline, glycine-betaine and soluble sugars occurs. This along with other things contributes to keep the integrity of cellular membranes and proteins, which are necessary for metabolic activities. Therefore, studies on the accumulation of these molecules have been used as physiological indicators in the evaluation of drought tolerance in several species (Shao *et al.*, 2008; Farooq *et al.*, 2009).

2.4.3.3.1 Free amino acid

Several amino acids can act as precursors for the synthesis of secondary metabolites and signalling molecules. Polyamines are derived from Arginine (Alcazar *et al.*, 2006). Recent studies also suggested that autophagy and abscisic acid-induced protein turnover contribute to the increase in free amino acids (Barros *et al.*, 2017; Hildebrandt, 2018; Hirota *et al.*, 2018). In situations of insufficient carbohydrate supply plants can use amino acids as alternative substrates for mitochondrial respiration due to a decrease in photosynthesis rates that usually occur during stress conditions, (Araujo *et al.*, 2011; Hildebrandt, 2018; Hildebrandt *et al.*, 2015).

When two teak clones were subjected to progressive drought stress (FG1 and FG11), the increase of free amino acid was 60.18 per cent in FG1 whereas it was 59.34 per cent in FG11 after inducing drought stress for 20 days. After 5 days re-watering FG11 showed incomplete recovery (Husen, 2010).

Extensive amino acid accumulation in response to drought stress has been reported for leaves of six-month old jack-plants, two apple cultivars and sugar apple plants when subjected to eight days of water deficit (Rodrigues *et al.*, 2010) and (Sircelj *et al.*, 2005).

2.4.3.3.2 Proline

Apart from the role of proline as an osmolyte, it is also considered as a potent antioxidant and potential inhibitor of programmed cell death (PCD). Proline acts as nonenzymatic antioxidant that plant requires to mitigate the adverse effect of reactive oxidase species (ROS). Functional role of proline appears to be cytoplasmic osmoticant to lower cell water potential and make the plant to absorb water from soil (Dalvi, 2015).

According to Husen (2010), proline accumulated progressively in teak clones under moisture stress. Highest accumulation was 87.09 per cent and

82.09 per cent in FG1 and FG11, respectively. While complete recovery was shown by both clones after five days of re-watering.

Juby (2019) in her study on cocoa hybrids under moisture stress observed increase in proline accumulation ranging from 85.5 to 2817 $\mu\text{g g}^{-1}$. Two Mediterranean shrubs (*Halimium halimifolium* L. and *Pistacia lentiscus* L.) were reported to accumulate proline during stress as well (Ain-Lhout *et al.*, 2001).

Similar results were found in olive tree (Sofa *et al.*, 2004), leaves of grapevine (Schultz and Matthews, 1993) apple trees (Wang *et al.*, 1995) and cherry trees (Ranney *et al.*, 1991).

2.4.3.3.3 *Glycine betaine*

Quaternary ammonium compounds (QACs) viz., glycine betaine, choline and proline betaine are key osmolytes contributing towards osmotic adjustment (Huang *et al.*, 2000). QACs like glycine betaine in low concentration can improve salt and cold stress tolerance, possibly by protecting photosynthetic protein complexes (Holmstrom *et al.*, 2000), by reducing lipid peroxidation of the cell membranes (Chen *et al.*, 2000), or by stabilizing enzymes and membranes during stress conditions (Sakamoto and Murata, 2000). Levels of accumulated glycine betaine are generally correlated with the extent of stress tolerance (Rhodes and Hanson, 1993).

Glycine betaine accumulation has been found in many crops like sugar beet (*Beta vulgaris*), spinach (*Spinacia oleracea*), barley (*Hordeum vulgare*), wheat (*Triticum aestivum*) and sorghum (*Sorghum bicolor*) (Weimberg *et al.*, 1984; Fallon and Phillips, 1989; Mc-Cue and Hanson, 1990; Rhodes and Hanson, 1993 and Yang *et al.*, 2003) under stress conditions.

2.4.3.3.4 *Total soluble sugar*

Total soluble sugar (TSS) is substrate in the synthesis of various amino acids which act as antioxidant and osmoregulator to reduce osmotic potential of

cells and protect cells from a variety of free radical compounds. The quantity of TSS varies in the stress reaction of different plant species (Hudak *et al.*, 2010).

Many authors reported different influence of water deficit in TSS, as these solutes regulate plant structure and physiology in a complex manner (Lemoine, 2013), and their roles in plant metabolism are conflicting in various plant species under different stress conditions (Gorham *et al.*, 1981; Akinci and Losel, 2009). Drossopoulos *et al.* (1987) and Fazeli *et al.* (2006) showed that total soluble sugars were increased in wheat, sesame, maize, fenugreek, and alfalfa plants under stress. While, Steward (1971) and Pattanagul and Madore (1999) found that TSS had either stable or decreasing concentrations in the cotyledons of soybean, watermelon, variegated coleus, and in maize under moisture stress.

Husen (2010) noticed that after 20 days of inducing water stress there was increase in TSS in two teak clones FG1 and FG11 by 27.36 and 26.81 per cent respectively and after 5 days of rehydration they did not recovered fully. Another study conducted by Silva *et al.* (2009) showed reduction of TSS in leaves after 31 days of water stress in six-month-old seedlings of four umbu tree genotypes.

2.4.3.4 Antioxidant enzymes

In plants, there is a defensive system, to avoid injuries of active oxygen and thus guaranteeing normal cellular function (Horvath *et al.*, 2007). The balance between ROS production and activities of antioxidative enzyme determines whether oxidative signaling and/or damage will occur (Moller *et al.*, 2007). To minimize the effects of oxidative stress, plants have evolved a complex enzymatic and non-enzymatic antioxidant system, such as low molecular mass antioxidants (glutathione, ascorbate, carotenoids) and ROS scavenging enzymes like superoxide dismutase (SOD), peroxidase (PDX), catalase (CAT), ascorbate peroxidase (APX) (Apel and Hirt, 2004). Efficient destruction of O_2^- and H_2O_2 in plant cells requires the concerted action of antioxidants.

2.4.3.4.1 Superoxide dismutase

Superoxide dismutase (SOD) is most efficient intracellular enzymatic antioxidant and provides the first defense response against the toxic effects of high levels of ROS by converting it to H₂O₂ and O₂. Increased activity of SOD observed in many studies points towards its induction to quench higher levels of superoxide radical generated due to NaCl stress (Sheokand *et al.*, 2008).

A study was conducted in two teak clones (FG1 and FG11) by inducing progressive drought stress for different periods (5, 10, 15 and 20) and rehydration. The SOD activity increased as the drought stress progressed and reached the maximum after 20 days of stress imposition. On rehydration for five days, SOD values got decreased but complete recovery compared to control was not observed (Husen, 2010).

Ren *et al.* (2016) observed no variations in the activity of SOD in seedlings of two genotypes (HR and ND4) of *Cerasus humilis* at optimal growth conditions. But when subjected to water stress, HR showed increase in SOD activity by 67 per cent while ND4 showed no difference compared to normal condition.

Wang (2014) in his study on rubber trees found that the SOD activity increased at 3 days of inducing water stress, however from 5 to 9 days of inducing water stress, there was decrease in SOD activity. These result meant that rubber tree seedlings was not tolerant to water stress, and the mechanism of defensive system of biochemical responses by SOD activity lasted only for 3 to 5 days after inducing water stress.

2.4.3.4.2 Peroxidase

Peroxidase (PDX), an iron heme protein, which accelerates the reduction of H₂O₂ with a concurrent oxidation of a substrate, mostly located in cell wall; it is also involved in oxidation of phenol compounds as the key enzyme for polymerization towards the synthesis of lignin (Gaspar *et al.*, 2002; Ozdemir *et*

al., 2004). PDX is a major enzyme scavenging H₂O₂ in chloroplasts produced through dismutation of O₂⁻ catalyzed by superoxide dismutase.

Husen (2010) opined that after 20 days of withholding water from teak seedling clones resulted in elevated expression of PDX by 27.53 per cent and 25.44 per cent in FG1 and FG11 clone, respectively. After 5 days of re-watering, FG11 clone show a complete recovery, while FG1 was not fully recovered.

Wei *et al.* (2015) reported that there was intensive increase in PDX activity during drought stress in *Diospyros lotus*. There was only slight increase observed in PDX two genotypes of palm tree (Neto *et al.*, 2018) and one year old walnut tree (Lotfi *et al.*, 2010) after induction of stress. Wang (2014) reported slight increase of PDX activity in rubber tree after exposing water stress for one day and later it got decreased continuously from 3 to 9 days of exposing to water stress.

2.4.3.5 Malondialdehyde

Lipid peroxidation, one of the most important causes of cell deterioration during drought stress, generates changes in the composition of fatty acids which affect the structural and functional properties of cell membranes, such as the inactivation of membrane-bound proteins and the increase in membrane permeability (Smirnoff 1993; Asada 1999). Very often MDA levels have been utilized as a suitable marker for membrane lipid peroxidation (Masia, 2003). MDA is a widely used marker of oxidative lipid injury caused by environmental stress (Kong, 2016).

Husen (2010) found that two teak clones exposed to drought stress for 20 days led to accumulation in MDA content by 172.73 per cent in FG1 and by 182.78 per cent in FG11. Complete recovery was observed in both clones after five days of rehydration.



MATERIALS AND METHODS



MATERIALS AND METHODS

An experiment was carried out at the Department of Forest biology and Tree Improvement, College of Forestry, KAU from August 2018 to April 2019, to investigate on “**Drought response in plus trees of teak (*Tectona grandis* Linn. f.) provenances of Kerala**”. The experiment was planned and executed as described below.

3.1 EXPERIMENTAL MATERIAL

Experiments were conducted with eleven plus tree accessions of *Tectona grandis*. One year ramet of *Tectona grandis* were obtained from The Division of Plant Genetics and Tree Breeding, Kerala Forest Research Institute (KFRI), Peechi. The details of *Tectona grandis* plus trees accessions is given in Table 1

Table 1. Details of the plus tree accessions used in the study

No. allotted	Accession No.	Forest Division	Range	Locality
1	KFRI T1	Nilambur	Nilambur	Aravallikkavu
2	KFRI T4	Nilambur	Edavanna	Edacode
3	KFRI T5	Nilambur	Karulai	Karulai
4	KFRI T9	Nilambur	Karulai	Cherupuzha
5	KFRI T16	Konni	Naduvathmuzhy	Naduvathmuzhy
6	KFRI T24	Kannavam	Kannavam	Nedumpoil
7	KFRI T44	Wyanad	Begur	That Road**
8	KFRI T55	Nilambur	Karulai	Karulai
9	KFRI T116	Thrissur	Pattikkad	pattikkad
10	KFRI T142	Konni	Naduvathumuzhy	Vayakkara
11	KFRI T144	Aryankavu	Aryankavu	Anchumocku

The study comprised of two experiments:

3.1.1 Experiment-I: Screening of ramet performance

One year old ramets were evaluated for their initial vigour and growth performance for six months. The seedlings were kept in poly house covered by white polyethylene plastic sheet in all side (6 m x 4 m) for almost three month. After that, the seedlings were transplanted in polybags (12'' x 10''), filled with the mixture of loam soil, sand and farmyard manure in 2:1:1. These polybags were kept inside a shade house; the upper portion of shade house was covered with green plastic shade, while the other parts remained open. Complete protection was provided against diseases by foliar spray with fungicides when required. Further these plants were maintained carefully by regular watering and weeding.

The randomized block design was used for this experiment. Three replications, each of five plants ($3 \times 5 = 15$) were used for each treatment (Eleven accessions) and in total 165 teak seedlings were maintained. Initial reading, after three months and after six months were obtained by measuring the height, diameter, branch number and total leaves number of the individual ramet. The physiological and biochemical measurements were also taken at six months age.

3.1.2 Experiment-II: Screening of ramet at drought and recovery

The study materials used in the first experiment were used for this experiment too. All plants were irrigated to field capacity. Drought stress was imposed by withholding water till the majority of plants showed signs of wilting. These plants were then irrigated to relieve the stress through regular irrigation to field capacity. This cycle of drought was repeated again by withholding water till wilting signs appeared. The second cycle of drought was considered as 'maximum stress' where biometric, physiological and biochemical observations were taken. Immediately after this, the plants were irrigated to field capacity and maintained as stress free through continuous irrigation. The plants after recovery were denoted as 'recovered plants' and morphological, physiological and biochemical measurements were taken at this stage.



Plate 1. One year old teak seedlings in nursery



Plate 2. Teak seedlings in nursery after three months



Plate 3. Teak seedlings in nursery after six months



a. Seedling after inducing water stress



b. Seedling after re-watering following drought stress

Plate 4 a-b. Views of plantlets of teak plus tree accessions during drought stress and after rehydration.

3.2 OBSERVATIONS RECORDED

3.2.1 Biometric parameters

3.2.1.1 Height

The height of all seedlings were measured from base to the top of the plant with the help of meter scale and recorded as plant height (cm).

3.2.1.2 Diameter

The basal diameter (mm) was measured from the collar region of plants of each bag.

3.2.1.3 Leaves number

The total number of leaves per plant was counted in each bag and then the average was calculated.

3.2.1.4 Branch number

The total number of branches per plant was counted in each bag and then the average was calculated.



a. Diameter measurement using Digital calliper



b. Height measurement using Meter scale

Plate 5 a-b. Recording observation on biometric parameters

3.2.2 Physiological parameter

Mature and healthy leaf of the seedling was used to take the different physiological characters.

3.2.2.1 *Relative Water Content (RWC)*

Relative Water Content is a measure of plant water stress in terms of the physiological consequence of cellular water deficit. It estimates the current water content of the sampled leaf tissue relative to the maximal water content it can hold at full turgidity.

Fresh leaves were used to take twenty leaf discs of one centimeter diameter and fresh weight of each disk was recorded. Petri dishes containing the disks were filled with distilled water and kept for soaking at room temperature and ambient light for four hours. The wet discs were bloated using tissue paper and then the turgid weight was recorded. The leaf discs were then oven dried at 80°C for 6 hours and the dry weight was recorded (Barrs, 1962). RWC was estimated as follows

$$\text{RWC (\%)} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Turgid weight} - \text{Dry weight}} \times 100$$

3.2.2.2 *Canopy air temperature (CATD)*

The leaf temperature was measured using the Infrared Thermometer. Canopy temperature depression was calculated through the difference between air temperature (Ta) and canopy temperature (Tc). The reading was recorded during morning hours.

$$\text{CATD} = \text{Ta} - \text{Tc}$$

3.2.2.3 *Photosynthetic rate (Pn)*

The photosynthetic rate of leaves was measured using the Infrared Gas Analyser (IRGA). The reading was recorded during morning hours.

3.2.2.4 Stomatal conductance (Gs)

The stomatal conductance was measured using the infrared gas analyser (IRGA). The reading was recorded during morning hours.

3.2.2.5 Transpiration rate (E)

The transpiration rate of leaves was measured using the infrared gas analyser (IRGA). The reading was recorded during morning hours.

3.2.2.6 Photosynthetic pigment and Chlorophyll content

Fresh leaf sample (0.1 gm) was collected and cut into small pieces and put in a test tube. Seven ml dimethyl sulfoxide (DMSO) was added to the test tube. The mixture was kept in dark at room temperature overnight. Three ml of DMSO was added to make it up to 10 ml (V) after the period of incubation and were mixed well. Absorbance of the solution was read at 663 nm and 445 nm in a spectrophotometer. Chlorophyll a and b were determined using below equation.

$$\text{Chlorophyll a} = 12.7 (A \text{ at } 663) - 2.69 (A \text{ at } 445) \times \frac{V}{1000 \times W}$$

$$\text{Chlorophyll b} = 22.9 (A \text{ at } 645) - 4.69 (A \text{ at } 663) \times \frac{V}{1000 \times W}$$

$$\text{Total chlorophyll content} = \frac{A \text{ at } 652}{34.5} \times \frac{1000 \times V}{1000 \times W}$$

3.2.2.7 Chlorophyll Stability Index (CSI)

Two sets of fresh leaf samples of 0.1g were cut into small pieces. One was kept in distilled water and was subjected to a temperature of 55°C for 30 minutes on hot water bath (treated) and the other sample was kept at room temperature (control). The treated samples were cooled to room temperature after the treatment period. After draining the samples 7 ml dimethyl sulfoxide (DMSO) was added to each tube. The samples were incubated overnight at room temperature in dark. After the incubation period, three ml of DMSO was added to make it up to 10 ml (V). The absorbance at 652nm (A652) was recorded

(Kaloyereas, 1958). The chlorophyll content (mg g^{-1} of fresh tissue) of the samples (control and treated) were estimated as shown below:

$$\text{Total chlorophyll} = \frac{A \text{ at } 652}{34.5 \times \text{FW}} \times V$$

The Chlorophyll Stability Index was worked out using the following formulae:

$$\text{CSI (\%)} = \frac{\text{Total chlorophyll (heated)}}{\text{Total chlorophyll (control)}} \times 100$$

3.2.2.8 Cell Membrane Stability (CMSI)

The cell membrane stability was assayed by estimating the ions leaching from leaf into distilled water. Leaf discs of 0.1 g were taken in test tubes filled by 15 ml of distilled water. One set was kept in room temperature for 3 hrs and electrical conductivity of the solution was determined using a conductivity bridge after removing the leaf discs (C1). The second set was kept in a boiling water bath (80°C) for 10 min and its electrical conductivity was also determined after cooling to room temperature (C2). Cell membrane stability Index was estimated as

$$\text{Electrolyte leakage} = [1 - (C1/C2)] \times 100$$



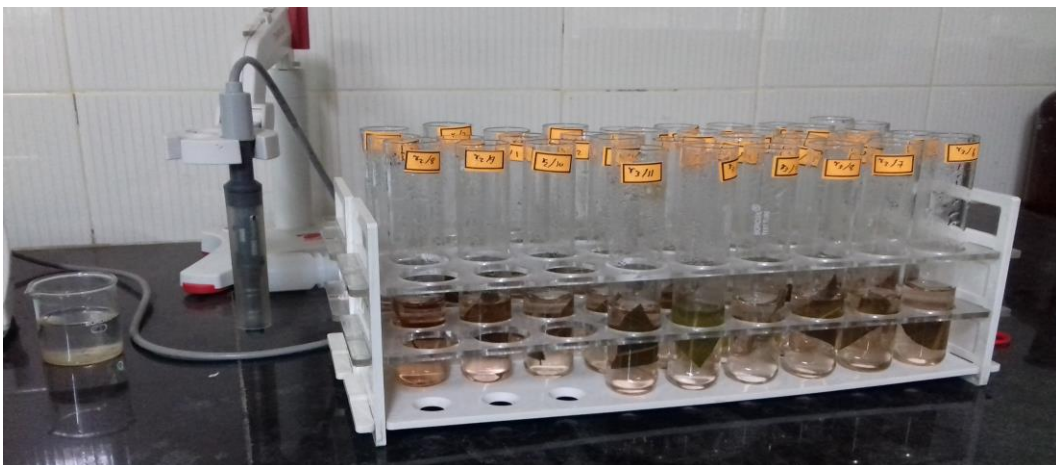
a. Measuring relative water content



b. Measuring gas exchange parameters using IRGA



c. Measuring canopy air temperature differences using IR thermometer



d. Measuring cell membrane stability using EC meter

Plate 6 a-d. Recording the physiological parameters

3.2.3 Biochemical parameters

Healthy third leaves from the top of the plants were collected for bioassay. The collected material was kept in a plastic zip bag. After the collection, each material was categorized and wrapped with an aluminum foil and was kept in refrigerator under -20°C till it was used.

3.2.3.1 Total soluble protein content (TSP)

Protein content was assayed following Lowry's method (Lowry *et al.*, 1951) with a standard curve prepared using bovine serum albumin and expressed in mg g^{-1} .

200 mg of leaves were homogenized in 2 ml of extraction buffer. The extracts were then centrifuged at 10000 rpm for 10 min. The supernatants were used for the estimation of total soluble protein content. A mixture of one ml of the supernatant along with five ml of reagent C (Alkaline copper solution) in a test tube was kept to stand for 10 minutes. After that, 0.5 ml of reagent D (Folin- Ciocalteu reagent) was added and mixed well and was incubated in dark for 30 minutes at room temperature. Blue color was developed and the absorbance was taken at 660 nm in a spectrophotometer.

3.2.3.2 Nitrate reductase activity (NRA)

Nitrate reductase activity in the leaves was determined using the method described by Nicholas *et al.* (1976) and the enzyme activity was expressed as $\text{mmol nitrate g}^{-1}\text{hr}^{-1}$.

0.2 g of leaf was ground to fine powder in a mortar with liquid nitrogen. Two ml of the extraction buffer containing 1mM EDTA, 25 mM cysteine, and 25 mM potassium phosphate (adjusted to pH 8.8) was added to the leaf tissue powder. The homogenate was centrifuged at 4°C for 15 min at 12,000 g. A reaction mixture consists of 0.5 phosphate buffer 0.1M (pH7.5), 0.2 ml potassium nitrate, 0.4 ml NADH and 0.7 ml water was prepared. The reaction was

initiated by adding 0.2 ml enzyme extract and then was terminated after incubating for 15 min by adding 1 ml of sulfanilamide followed by naphthylethylenediamine reagent. Absorbance was measured at 540 nm was observed in a spectrophotometer. Standard graph is prepared with potassium nitrate (0.01 M) with series of test tube and made the volume in each tube to 2 ml by adding distilled water.

3.2.3.3 Free amino acid (FAA)

Total free amino acid was estimated by following the method described by Moore and Stein (1948) and expressed in mg g^{-1} .

200 mg leaf sample was homogenized with 2 ml of 80% ethanol and centrifuged at 3000 rpm for 5 minutes. A 2 ml mixture containing extract supernatant, ninhydrin solution and distilled water (0.1ml, 1 ml and 0.9 ml respectively) was kept in boiling water bath for 20 minutes. Following that, 5 ml of diluent was added and then was mixed well. After 15 minutes the intensity of purple color was measured at 570 nm using a spectrophotometer. Standard solution of leucine ranged between 10 μg -100 μg is prepared and used for estimating the free amino acid.

3.2.3.4 Proline (Pro)

The proline content was determined using acid ninhydrin reagent by the method of Bates *et al.* (1973). The proline content was expressed in terms of $\mu\text{g g}^{-1}$.

A grounded leaf sample (0.2g) in liquid nitrogen was homogenized with 4 ml of 3 per cent sulphosalicylic acid and the homogenate was centrifuged at 3000 rpm for 10 minutes. Two ml of the filtrate was mixed with 2 ml acid ninhydrin and 2 ml of glacial acetic acid in the test tube. The test tube was kept in water bath (100 °C) for one hour and then was placed on ice to cool down. 4 ml of toluene was added to the reaction mixture and stirred vigorously using vortex for 15 to 20 seconds. Two separate layers were formed and the layer containing toluene was taken out and warmed to room temperature. The intensity of the colour was read at 520 nm using toluene as a blank. Content was calculated as

$$\mu\text{moles/g tissue of proline} = \frac{\mu\text{g of proline/ml} \times \text{ml toluene}}{115.5} \times \frac{5}{\text{g sample}}$$

3.2.3.5 *Glycine betaine (GB)*

Glycine betaine was assayed based on the method described by Grieve and Gratten (1983) and expressed in mg g^{-1} .

A five-day dried leaf sample (500 mg) was finely ground and put in 50 centrifuge tube containing 20 ml of distilled water. The sample was shaken for 24 hours using mechanical shaker. 0.5 ml acidified extract was prepared by diluting 1ml of sample extract in 1 ml of 2N H_2SO_4 . The acidified extract was placed in ice water for 1 hour. 0.2 ml of cold potassium tri iodide solution was added to it and was mixed gently in a vortex mixture and was stored at 4°C for 15 minutes. Following that, the sample was centrifuged at 10,000 rpm for 15 minutes at 0°C. The supernatant was pipetted out using micropipette leaving only the crystals. The crystals were dissolved in 9 ml of 1, 2-Dichloroethane with vigorous vortexing. The absorbance was measured at 365 nm in a spectrometer after 2.5 hours. Reference standard of glycine betaine is prepared in 1N H_2SO_4 and used for estimating the glycine betaine content and the results are expressed in mg g^{-1} .

3.2.3.6 *Total soluble sugar (TSS)*

Total water soluble sugar (TSS) was determined by anthrone method (Hedge *et al.*, 1962) and expressed in mg g^{-1} .

A 100 mg of dry sample was taken in a test tube and was boiled in water bath for three hours after adding 5 ml of 2.5 N HCl. The sample was cooled and then neutralized with sodium carbonate till the effervescence ceases and the volume was made up to 50ml after transferring to a 50ml centrifuge tube. One ml of the supernatant was taken for analysis and 4 ml of anthrone reagent was added to it and the mixture was heated in boiling water for 8 minutes. The sample was cooled rapidly and then the intensity of green color was measured at 630 nm using a spectrophotometer.

$$\text{Amount of TSS in 100 mg of the sample} = \frac{\text{mg of glucose}}{\text{Volume of test sample}} \times 100$$

3.2.3.7 *Super oxide dismutase (SOD)*

SOD activity was assayed by the method described by Dhindsa and Matowe (1981) and expressed as units protein⁻¹g⁻¹.

Frozen Leaf sample (0.2 g) was macerated in liquid nitrogen. Two ml of 250 mM potassium phosphate extraction buffer (pH 7.8) was added and homogenized and then centrifuged at 10,000 rpm for ten minutes at 4°C. The supernatant was used as an enzyme source within 12 hours of extraction. 50 µl of this supernatant was then added to a reaction mixture (3ml) containing 50 Mm phosphate buffer (pH 7.8) , 13 mM methionine, 75 µM NBT, 0.1mM EDTA, 0.8 ml of distilled water, in duplicate and then 2 µM riboflavin was added. As control, a reaction mixture was kept without adding any enzymes for obtaining maximum color. The tubes were placed under fluorescent lamps (15 W) for 15 minutes and transferred to dark to stop the reaction. A complete reaction mixture without enzyme extract kept in dark served as blank. The absorbance reading was taken at 560 nm in a spectrophotometer. One unit of enzyme activity is the amount of enzyme which reduced the absorbency reading to 50 per cent in comparison with tubes lacking the enzyme

$$\text{Unit (of enzyme)} = \frac{\text{Blank} - \text{Sample}}{\text{Blank}/2}$$

3.2.3.8 *Peroxidase (PDX)*

Peroxidase activity was based upon the method as described by Castillo *et al.* (1984) which measures the increase in absorbance at 470 nm.

Frozen Leaf sample (0.2 g) was macerated in liquid nitrogen. Three ml of 0.1 M phosphate buffer (pH 7) was added and homogenized and then centrifuged at 14,000 g for 15 minutes at 5°C and the supernatant was collected for assay.

3 ml of enzyme reaction mixture containing 50 mM phosphate buffer (pH 7.0), 0.05 ml guaiacol solution and 0.1 ml enzyme extract was mixed properly by using a micropipette for 3-5 seconds. The reaction was initiated by adding 0.5 ml of 12 mM H₂O₂. An increase in absorbance due to the formation of tetra-guaiacol was measured at 436 nm using a spectrophotometer for three min at an interval of 30 sec using stop watch. The enzyme activity was calculated as per micromole extinction coefficient of its dehydrogenation which is 6.39. The enzyme activity was expressed as min⁻¹ mg⁻¹ protein

3.2.3.9 Malondialdehyde (MDA)

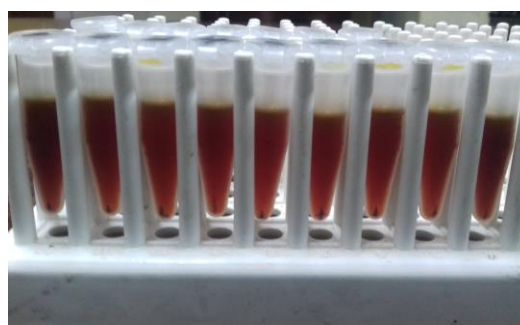
The level of lipid peroxidation was measured in terms of malondialdehyde (MDA) content which is a product of lipid peroxidation, following the method described by Rao and Sresty (2000), and expressed as n mol g⁻¹.

Leaf sample of 0.2 g was homogenized in 2 ml of 0.1 per cent trichloroacetic acid (TCA). The homogenate was centrifuged at 10000 g for 10 minutes. A mixture of 1 ml of the extract supernatant and 4.0 ml of 0.5 per cent thiobacbituric acid (TBA) in 20 per cent TCA was prepared and mixed well. The mixture was kept in a boiling water bath at 95° C for 15 min and then quickly placed in an ice bath for cooling. The sample was centrifuged at 10000 g for 5 min after that the absorbance was measured at 532 nm using a spectrophotometer.

MDA content was calculated by its extinction coefficient of 155 mM⁻¹ cm⁻¹



a. Free amino acid extraction



b. Glycine betaine extraction

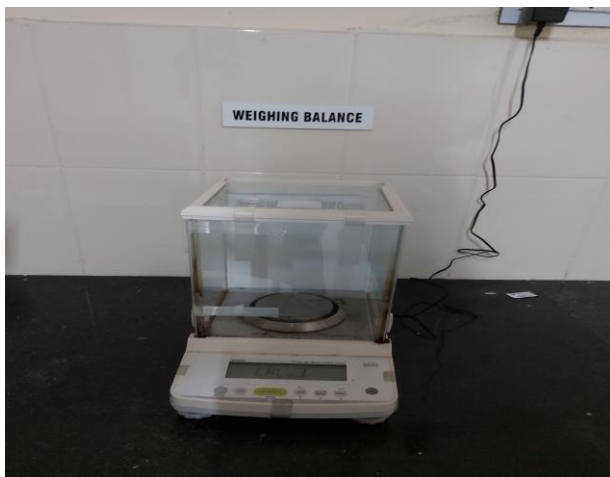
Plate 7 a-b. Extraction of biochemical components



a. Measuring the absorbance



b. Centrifuge to separate the supernatant



c. Weighting the leaves and chemicals



d. Maceration of leaves

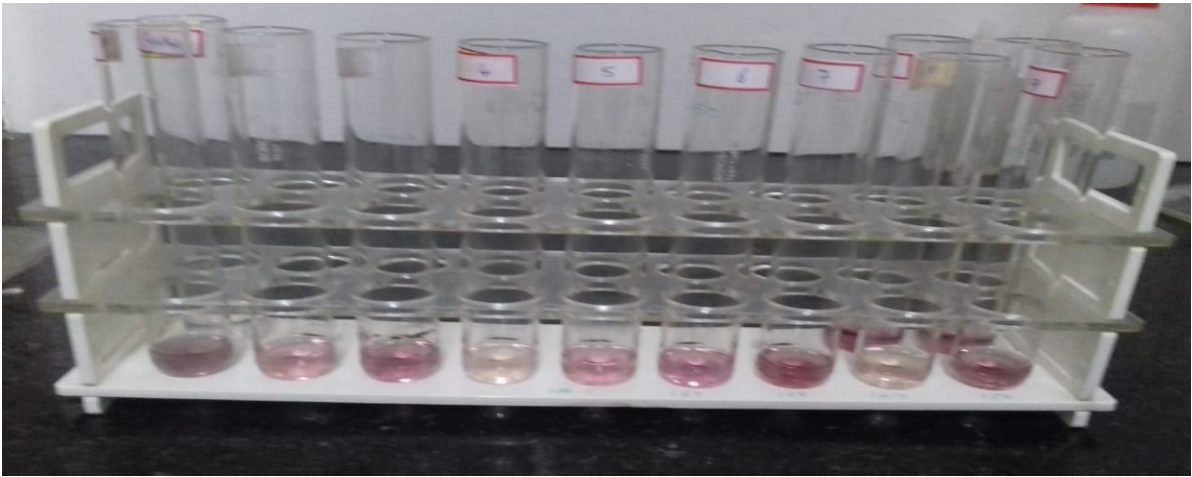


e. Micro pipette for taking solutions

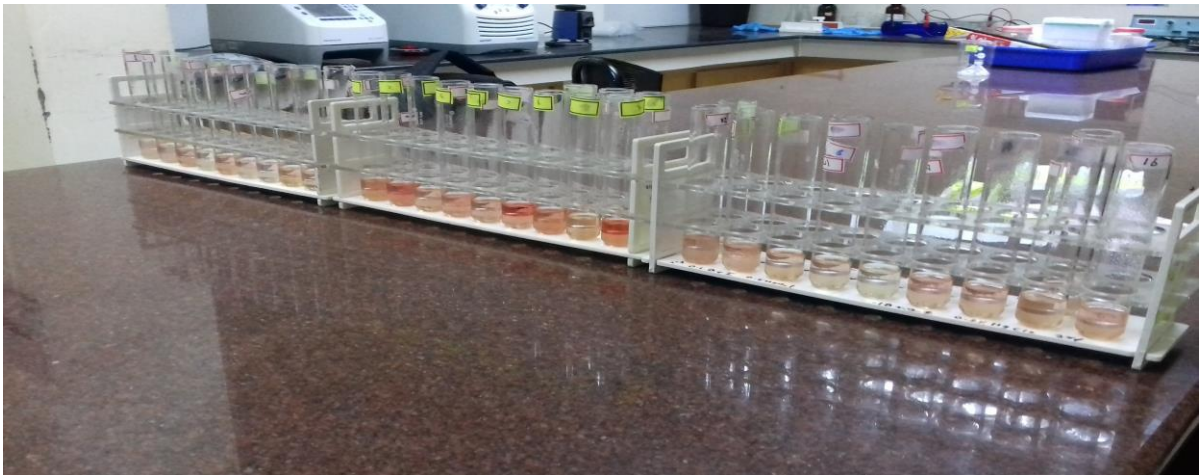
Plate 8 a-e. Equipments used for the biochemical analysis



a. Total soluble protein

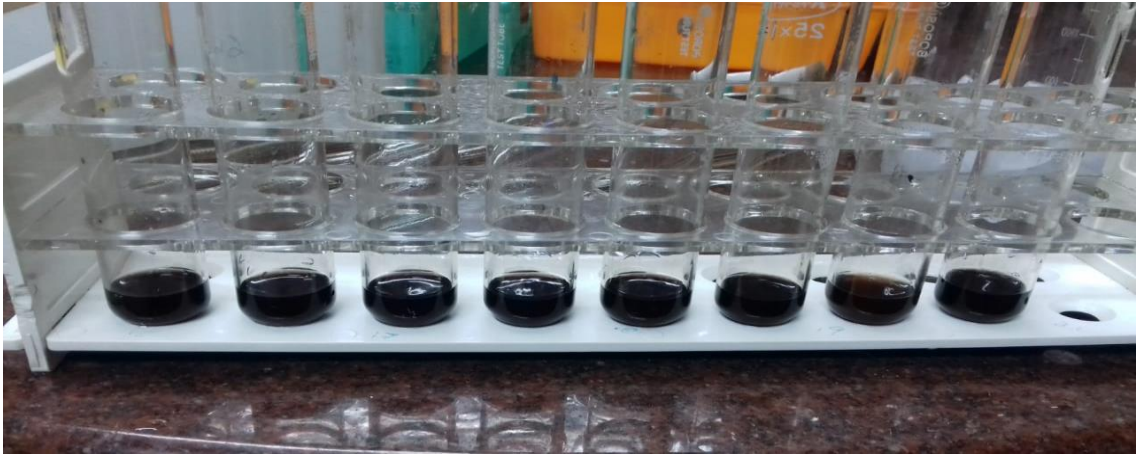


b. Nitrate reductase activity

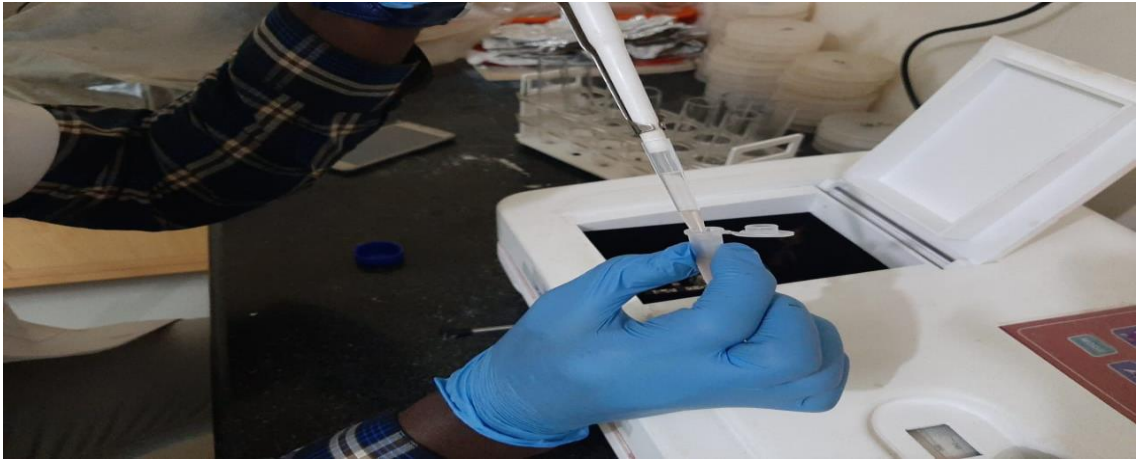


c. Proline

Plate 9 a-c. Estimation of biochemical components



a. Super oxide dismutase

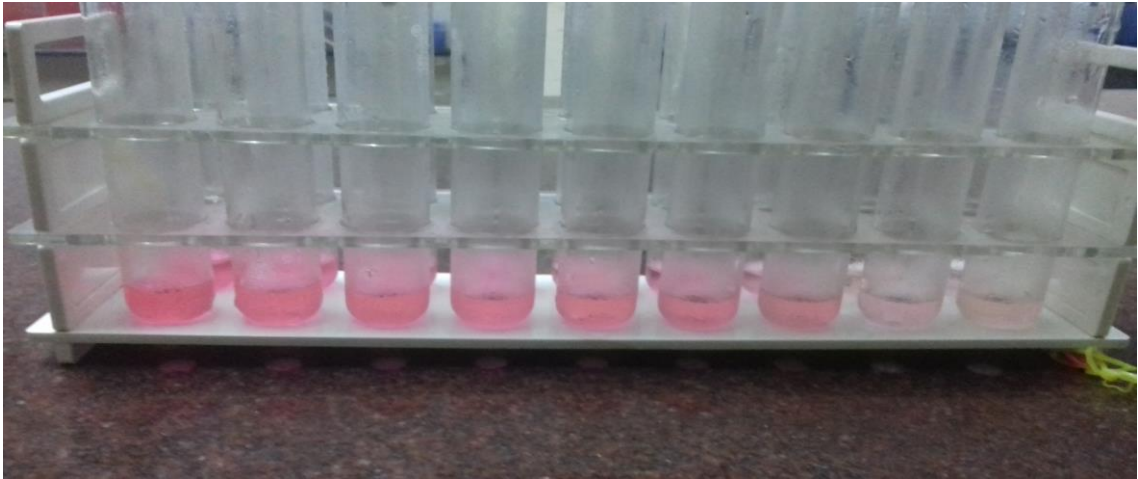


b. Peroxidase activity

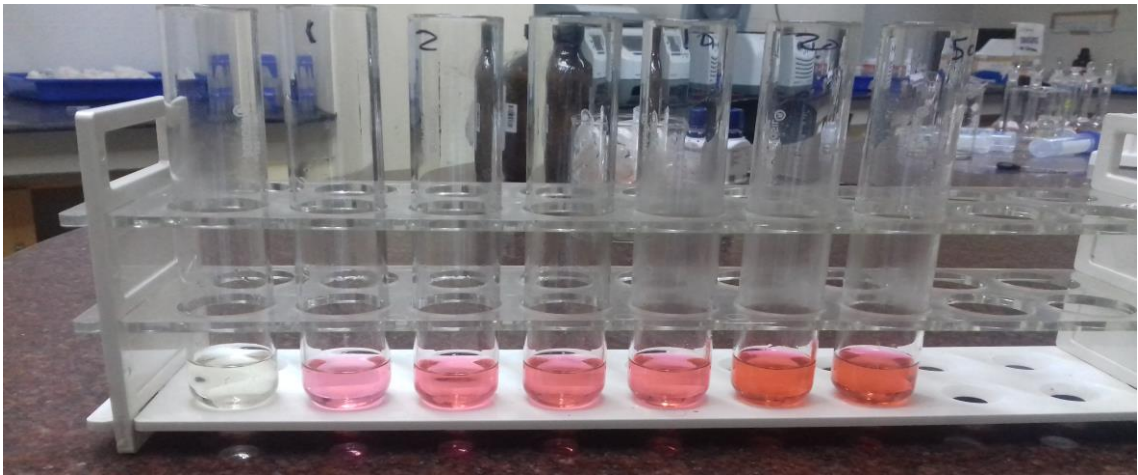


c. Malondialdehyde

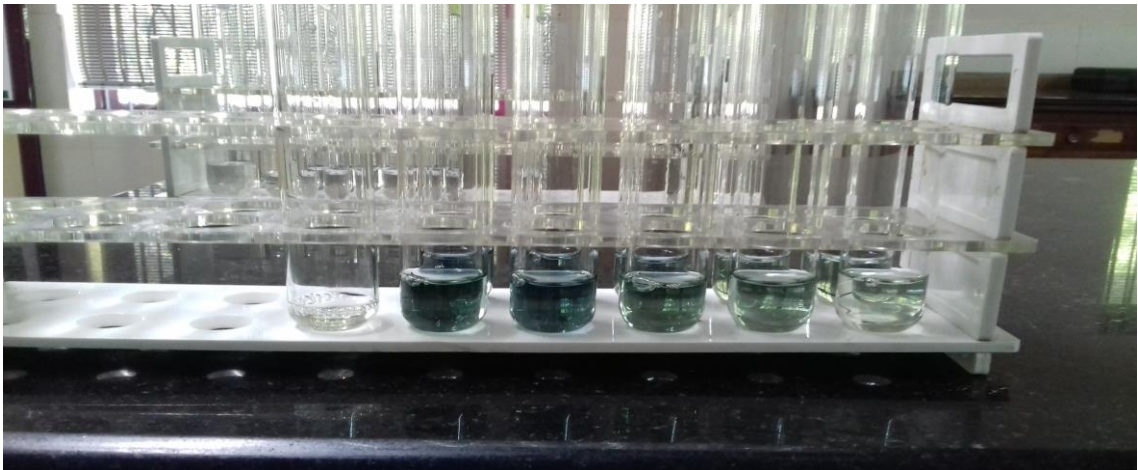
Plate 10 a-c. Analysis of antioxidant enzymes and malondialdehyde



a. Nitrate reductase activity



b. Glycine betaine



c. Total soluble sugar

Plate 11 a-c. Preparation of standard curves for analysis of biochemical parameter

3.2.4 Statistical analysis

3.2.4.1 Simple correlation analysis

The correlation coefficients were calculated to determine the degree of association of physiological and biochemical characters with number of leaves using SPSS software.

3.2.4.2 Cluster analysis

The data of physiological and biochemical variations of different accessions was subjected to hierarchal clustering analysis. The cluster analysis was carried out using between link linkage as the clustering method and euclidean distance as the interval using the R software.

3.2.4.3 Principal component analysis

The collected data on physiological and biochemical analysis was subjected to principle component analysis using R software.



RESULTS



RESULTS

The results obtained in the investigation on “**Drought response in plus trees of teak (*Tectona grandis* Linn. f.) provenances of Kerala**” are presented below. Some of the values were found to deviate from normality; hence, logarithmic transformation was done prior to statistical analysis.

4.1 BIOMETRIC OBSERVATIONS

4.1.1 Screening of plus tree accessions at normal condition

Biometric observations were recorded from one year old teak plants for six months under nursery condition and the results are summarized in Table 2.

Plant height recorded at first month showed significant difference among accessions. The overall mean height of the accessions was 26.5 cm. The height of the accessions ranged from 14.4 cm of KFRI T142 to 49.5 cm of KFRI T44. Accessions KFRI T44 and T4 had tall plants, which showed significant difference from KFRI T116 (37.9 cm), while, the accessions KFRI T9, T16 and T142 were having shorter plants.

Plant height observed at three and six months under nursery conditions nursery did not differ significantly ($P < 0.05$) among accessions. The overall mean of height after three month was 44.5 cm, while at six months the overall mean height was 67.9 cm.

The accessions did not show any significant difference for plant diameter at three stages of growth under nursery condition. During first month average diameter was observed to be 7.4 mm. At the third month the average diameter of the accessions was 10.6 mm and at six months the average diameter of the accessions was 15.7 mm.

Number of leaves also did not vary significantly among the accessions during three stages of growth under nursery condition in teak. The mean number

of leaves in initial, third and six month at nursery were 10.4, 17.5 and 22.0 respectively.

Accessions did not show variation in number of branches during the experiment with mean values of 0.45, 1.34 and 2.00 at first, third and six months respectively.

4.1.2 Performance of teak plus tree accessions under induced water stress

After evaluating the growth in nursery, teak accessions were subjected to two consecutive drought instances with an intermediate recovery, regular screening of biometric observation for three weeks after which they were analyzed for biochemical and physiological parameters.

After maximum drought induced, the main changes shown by drought stressed plants was the withering of leaves and change in branching habit (drying off). Data on the various biometric parameters are given in Table 3.

Analysis indicated that accessions did not differ ($P < 0.05$) in height, diameter, number of leaves and number of branches after stress treatment. The accessions showed slight increase in growth parameters like plant height and diameter. The mean increase in plant height and diameter was 7.5 and 5.5 per cent respectively. Drought resulted in reduction in the number of leaves in all the accessions. Accession KFRI T55 seemed to tolerate drought by losing 5.3 per cent of their leaves whereas KFRI T16 lost 57.3 per cent of their leaves. The average loss of leaves was 22.7 per cent.

Response of accessions to induced drought stress with respect to number of branches was varying. The number of branches was observed to increase in accessions KFRI T5, T9 and T144 by 6.3 per cent, 18.2 per cent and 37.5 per cent respectively, while other accessions showed decrease in number of branches and the decrease ranged from 0.71 per cent of KFRI T16 to 41.1 per cent of KFRI T24. The average reduction in number of branches under drought was 9.89 per cent.

Table 2. Biometric observations on plantlets of teak plus tree accessions in nursery condition at start of experiment.

SI No	Accession	Height (cm)			Diameter (mm)			Number of leaves			Number of branches		
		First	Third	Sixth	First	Third	Sixth	First	Third	Sixth	First	Third	Sixth
1	KFRI T1	21.1 ^{cd}	45.4	71.5	5.41	10.3	17.0	11.3	23.9	25.0	0.04	2.46	3.13
2	KFRI T4	48.5 ^a	58.7	69.2	9.65	12.6	16.2	11.1	20.9	24.8	0.76	2.11	2.39
3	KFRI T5	22.4 ^{cd}	40.9	64.2	6.62	10.0	15.6	9.76	13.3	19.3	0.17	0.48	1.27
4	KFRI T9	14.6 ^e	31.3	55.5	5.62	7.94	13.7	8.39	12.3	19.1	0.74	1.00	1.39
5	KFRI T16	18.3 ^{de}	34.5	66.9	7.12	9.72	17.4	10.3	15.7	20.5	0.97	0.88	2.00
6	KFRI T24	19.6 ^{cd}	45.9	69.6	6.05	10.4	15.6	12.1	15.1	18.0	0.26	0.78	1.39
7	KFRI T44	49.5 ^a	53.3	72.9	10.1	12.7	16.8	11.9	19.3	27.2	0.37	1.99	2.53
8	KFRI T55	22.0 ^{cd}	46.1	68.7	6.58	11.5	15.7	10.3	20.9	22.1	0.00	1.46	2.45
9	KFRI T116	37.6 ^b	55.4	74.6	8.69	12.0	15.0	9.87	20.4	24.8	0.31	1.27	2.04
10	KFRI T142	14.4 ^e	32.8	61.1	7.17	9.94	15.1	11.8	17.1	20.4	1.03	1.37	2.05
11	KFRI T144	23.8 ^c	44.9	69.7	8.19	10.3	16.5	7.65	13.9	20.9	0.33	0.92	1.30
S. E		0.000	0.129	0.206	0.181	0.200	0.320	0.359	0.142	0.602	0.268	0.074	0.448
F-value		49.998*	1.786	1.515	1.589	1.531	0.849	1.181	1.730	0.835	1.358	2.112	1.039

* Significant at 0.05 levels
 Values with same superscript within a column are homogenous

Table 3. Growth parameters of plantlets of teak plus tree accessions at pre and post drought induction.

SI No	Accession	Height (cm)			Diameter (mm)			Number of leaves			Number of branches		
		Stress inducement		Change (%)	Stress inducement		Change (%)	Stress inducement		Change (%)	Stress inducement		Change (%)
		Before	After		Before	After		Before	After		Before	After	
1	KFRI T1	71.52	75.59	5.69	17.04	17.09	0.29	25.03	22.12	-11.6	3.13	2.55	-18.6
2	KFRI T4	69.21	76.28	10.2	16.19	17.56	8.46	24.84	17.79	-28.4	2.39	1.44	-39.8
3	KFRI T5	64.21	65.87	2.59	15.6	16.05	2.88	19.32	16.2	-16.2	1.27	1.35	6.30
4	KFRI T9	55.52	60.75	9.42	13.69	14.80	8.11	19.10	14.39	-24.7	1.39	1.65	18.2
5	KFRI T16	66.85	70.36	5.25	15.89	16.43	3.40	20.52	8.773	-57.3	2.00	1.98	-0.71
6	KFRI T24	69.56	77.93	12.0	15.62	16.88	8.07	18.03	14.54	-19.4	1.39	0.82	-41.1
7	KFRI T44	72.95	74.27	1.81	16.78	17.77	5.90	27.15	22.27	-17.9	2.53	2.24	-11.6
8	KFRI T55	68.16	70.02	2.73	15.71	16.43	4.58	22.05	20.88	-5.31	2.45	1.73	-29.3
9	KFRI T116	74.6	78.98	5.87	15.04	16.39	8.98	24.84	22.68	-8.70	2.04	1.77	-13.4
10	KFRI T142	61.06	70.56	15.6	15.14	15.42	1.85	20.37	16.13	-20.8	2.05	1.72	-16.3
11	KFRI T144	69.74	77.88	11.8	16.49	17.71	7.40	20.89	12.57	-39.8	1.30	1.79	37.5
S. E		0.206	0.428		0.320	0.513		0.602	0.136		0.448	0.558	
F-value		1.515	1.069		0.849	0.948		0.835	1.758		1.358	0.89	

* Significant at 0.05 levels

Values with same superscript within a column are homogenous

4.2 PHYSIOLOGICAL OBSERVATIONS

4.2.1 Impact of optimal growth on physiological parameters of teak plus tree accessions

Observations of different physiological parameters among different plus tree accessions at normal condition is presented in Table 4. Statistical analysis showed that there was no significant ($P < 0.05$) difference in relative water content, canopy air temperature difference, photosynthesis, transpiration, chlorophyll a, chlorophyll b, total chlorophyll content, chlorophyll stability index and cell membrane stability index among the different teak plus tree accessions. Only stomatal conductance was differed between the accessions at this stage.

Relative water content of the accessions ranged from 98.7 per cent (KFRI T144) to 80.0 per cent (KFRI T116). The average was observed to be 85.2 per cent.

Canopy air temperature difference values were recorded between -2.16°C (KFRI T9) and -1.32°C (KFRI T44). The average value was observed to be -1.74°C and the differences were non-significant.

Photosynthetic rate of the accessions ranged from $6.36 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ in KFRI T116 to $7.58 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ in KFRI T144 with overall mean of $6.93 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$.

High stomata conductance was observed in KFRI T9 ($0.469 \text{ mol m}^{-2} \text{ s}^{-1}$) which was on par with KFRI T4, T5, T24, T142 and T144. Low stomatal conductance was exhibited by accessions KFRI T1, T16 and T116 which was on par with all other accessions except KFRI T9 indicating that the stomatal conductance of the accessions were also on par except KFRI T9.

Transpiration rate of the accessions ranged between $6.06 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ of KFRI T16 and $4.19 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ of KFRI T44. The average transpiration rate of the accessions was $5.34 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$.

Chlorophyll a content of the accessions ranged from 18.54 mg g⁻¹ (KFRI T24) to 19.75 mg g⁻¹ (KFRI T9) and the average chlorophyll content was 19.31 mg g⁻¹.

Chlorophyll b content did not differ among the accessions, and values ranged between 17.6 mg g⁻¹ (KFRI T24) and 30.36 mg g⁻¹ (KFRI T44). The average was 23.62 mg g⁻¹.

Chlorophyll stability index ranged from 88.82 per cent of KFRI T24 to 98.07 per cent of KFRI T9 and the mean value of accessions was 93.62 per cent.

Total chlorophyll content ranged from 36.14 mg g⁻¹ (KFRI T24) to 49.87 mg g⁻¹ (KFRI T44). The average of the accessions for total chlorophyll content was 42.93 mg g⁻¹.

Cell membrane stability index ranged from 0.26 in KFRI T16 to 1.64 in KFRI T24 and the average of the accessions was 0.99.

Table 4. Physiological parameters of plantlets of teak plus tree accessions under normal condition

SI No.	Accession	RWC	CATD	Pn	Gs	E	Chl. a	Chl. b	CSI	Total Chl.	CMSI
1	KFRI T1	91.2	-1.64	6.66	0.291 ^b	5.53	19.09	27.30	89.72	46.40	1.67
2	KFRI T4	84.8	-1.78	6.58	0.322 ^{ab}	5.47	19.59	27.96	94.38	47.54	0.61
3	KFRI T5	86.7	-1.73	6.91	0.318 ^{ab}	4.86	19.30	26.28	94.86	45.58	1.36
4	KFRI T9	87.9	-2.16	7.39	0.469 ^a	5.55	19.75	20.07	98.07	39.81	1.16
5	KFRI T16	81.0	-1.94	7.30	0.283 ^b	6.06	18.57	20.81	91.96	39.38	0.26
6	KFRI T24	81.5	-1.67	6.61	0.305 ^{ab}	5.75	18.54	17.60	88.82	36.14	1.64
7	KFRI T44	84.0	-1.32	6.61	0.230 ^b	4.19	19.50	30.36	95.08	49.87	1.37
8	KFRI T55	88.0	-1.55	7.07	0.289 ^b	4.53	19.44	29.85	92.29	49.29	0.80
9	KFRI T116	80.0	-1.50	6.36	0.269 ^b	5.09	19.44	19.80	95.59	39.24	0.42
10	KFRI T142	82.4	-1.86	7.13	0.388 ^{ab}	5.80	19.74	20.22	96.55	39.96	0.64
11	KFRI T144	98.7	-2.02	7.58	0.395 ^{ab}	5.93	19.40	19.58	92.47	38.98	0.97
S. E		0.510	0.416	0.370	0.004	0.665	0.490	0.044	0.194	0.060	0.068
F-value		0.954	1.081	1.154	3.885*	0.758	0.976	2.364	1.531	2.197	2.12

RWC = Relative water content (%), CATD = Canopy air temperature differences ($^{\circ}\text{C}$), Pn = Photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), Gs = Stomatal conductance ($\text{mol m}^{-2} \text{ s}^{-1}$), E = Transpiration rate ($\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$), Chl. a = Chlorophyll a (mg g^{-1}), Chl. b = Chlorophyll b (mg g^{-1}), CSI = Chlorophyll stability index (mg g^{-1}), Total Chl. = Total chlorophyll (mg g^{-1}), CMSI = Cell membrane stability index (rate)

* Significant at 0.05 levels

Values with same superscript within a column are homogenous

4.2.2 Impact of drought stress on physiological parameters of plantlets of teak plus tree accessions

Data on the physiological parameters of plantlets of different teak plus tree accessions exposed to drought stress are given in Table 5. Significant differences were observed between the accessions for stomatal conductance, chlorophyll a, chlorophyll b and total chlorophyll content while relative water content, canopy air temperature, photosynthesis, transpiration, chlorophyll stability index and cell membrane stability did not show significant difference among the accessions ($P < 0.05$).

Relative water content (RWC) of the accessions ranged between 47.3 per cent of KFRI T44 and 72.3 per cent of KFRI T9 and the mean value of RWC among the accessions was 58.8 per cent.

Canopy air temperature difference of the accessions was observed to be between 1.33 °C (KFRI T24) to 1.83 °C (KFRI T44). The average canopy air temperature difference among the accessions was 1.62 °C.

Photosynthetic rate of the accessions was between 3.49 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ (KFRI T9) and 4.03 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ (KFRI T142) with an overall mean value of 3.70 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$.

KFRI T116 showed the highest stomatal conductance (0.069 $\text{mol m}^{-2} \text{ s}^{-1}$) which was on par with KFRI T1, T4, T9, T16, T24, T55, T142 and T144. Lower values of stomatal conductance were showed by KFRI T5 and KFRI T44.

Transpiration rate of the accessions was in a range of 0.33 $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (KFRI T44) to 1.18 $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (KFRI T116) and the average was estimated to be 0.74 $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$.

Chlorophyll a content of the accessions showed significant difference. Accession KFRI T16 exhibited the lowest value of 10.87 mg g^{-1} . This accession

was on par with KFRI T1, T9, and T144. Accessions KFRI T1, T4, T24, T44, T55, T116 and T142 having high values of chlorophyll a content and were on par.

Highest chlorophyll b value was observed in accession KFRI T44 and KFRI T55 i.e., 20.41 mg g⁻¹ and 20.53 mg g⁻¹ respectively. These accessions differed from KFRI T16 having a value of 4.559 mg g⁻¹. The chlorophyll b content of accessions KFRI T1, T4, T5, T9, T24, T44, T55, T116, T142 and T144 were on par. Also the value of KFRI T16 was on par with values of KFRI T1, T4, T5, T9, T24, T116, T142 and T144.

Chlorophyll stability index values of the accessions ranged between 66.17 per cent (KFRI T44) and 79.33 per cent (KFRI T4) with a mean value of 73.48 per cent.

Among the accessions total chlorophyll content was lowest in KFRI T16 (15.43 mg g⁻¹) and it was on par with the accessions KFRI T1, T5, T9, T116, T142 and T144. While it differed from the total chlorophyll content of the accessions KFRI T4, T44 and T55. High chlorophyll content was observed in KFRI T4, T44 and T55 and their values were also on par with KFRI T1, T5, T9, T116, T142 and T144.

Cell membrane stability index of the accessions ranged from 3.36 of KFRI T142 to 5.92 of KFRI T24 and the mean CMSI was 4.79.

Table 5. Physiological parameters of plantlets of teak plus tree accessions exposed to drought stress

SI No.	Accession	RWC	CATD	Pn	Gs	E	Chl. a	Chl. B	CSI	Total Chl.	CMSI
1	KFRI T1	60.1	1.64	3.61	0.024 ^{ab}	0.58	16.98 ^a	13.37 ^{ab}	69.92	30.35 ^{ab}	3.86
2	KFRI T4	58.2	1.53	3.60	0.038 ^{ab}	0.90	17.76 ^a	17.41 ^{ab}	79.33	35.16 ^a	4.72
3	KFRI T5	48.0	1.77	3.59	0.013 ^b	0.35	14.57 ^{ab}	8.600 ^{ab}	73.38	23.17 ^{ab}	5.78
4	KFRI T9	72.3	1.65	3.49	0.039 ^{ab}	0.94	16.09 ^{ab}	12.69 ^{ab}	70.20	28.78 ^{ab}	4.77
5	KFRI T16	64.5	1.77	3.66	0.018 ^{ab}	0.46	10.87 ^b	4.559 ^b	70.45	15.43 ^b	4.16
6	KFRI T24	56.5	1.33	3.66	0.027 ^{ab}	0.86	16.60 ^a	14.02 ^{ab}	74.38	30.62 ^{ab}	5.92
7	KFRI T44	47.3	1.83	3.73	0.013 ^b	0.33	18.01 ^a	20.41 ^a	66.17	38.42 ^a	4.93
8	KFRI T55	55.5	1.80	3.81	0.044 ^{ab}	0.85	18.57 ^a	20.53 ^a	77.44	39.10 ^a	4.67
9	KFRI T116	58.1	1.64	3.83	0.068 ^a	1.18	17.02 ^a	11.39 ^{ab}	74.89	28.41 ^{ab}	5.00
10	KFRI T142	56.1	1.40	4.03	0.055 ^{ab}	1.05	17.07 ^a	13.59 ^{ab}	79.09	30.66 ^{ab}	3.36
11	KFRI T144	64.8	1.51	3.70	0.028 ^{ab}	0.64	14.85 ^{ab}	8.909 ^{ab}	73.03	23.75 ^{ab}	5.58
S. E		0.58	0.939	0.802	0.039	0.164	0.005	0.007	0.653	0.004	0.915
F-value		0.861	0.386	0.594	2.671*	1.627	3.647*	3.454*	0.773	3.921*	0.431

RWC = Relative water content (%), CATD = Canopy air temperature differences ($^{\circ}\text{C}$), Pn = Photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), Gs = Stomatal conductance ($\text{mol m}^{-2} \text{ s}^{-1}$), E = Transpiration rate ($\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$), Chl. a = Chlorophyll a (mg g^{-1}), Chl. b = Chlorophyll b (mg g^{-1}), CSI = Chlorophyll stability index (mg g^{-1}), Total Chl. = Total chlorophyll (mg g^{-1}), CMSI = Cell membrane stability index (rate)

* Significant at 0.05 levels

Values with same superscript within a column are homogenous

4.2.3 Physiological parameters of plantlets of teak plus tree accessions after rehydration

Observations recorded on physiological parameters of plantlets of teak plus tree accessions after rehydration is presented in Table 6. Result shows that there is no significant difference for the observed physiological parameters ($P < 0.05$) among the plantlets of plus tree accessions after rehydration.

Relative water content of the accessions ranged between 68.9 per cent (KFRI T55) to 83.5 per cent of KFRI T1 with an average of RWC of 78.4 per cent.

For canopy air temperature difference values of the accessions ranged from -1.02°C of KFRI T44 to -1.93°C of KFRI T144 with a mean value of 1.52°C .

Photosynthesis rate among the accessions was found to be ranging from $5.35 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ of KFRI T44 to $6.74 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ of KFRI T16 with overall mean of $6.09 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$.

Stomatal conductance values of the accessions ranged from $0.244 \text{ mol m}^{-2} \text{ s}^{-1}$ (KFRI T16) to $0.127 \text{ mol m}^{-2} \text{ s}^{-1}$ (KFRI T55). The mean stomatal conductance was $0.20 \text{ mol m}^{-2} \text{ s}^{-1}$.

Transpiration values of the accessions ranged between $4.47 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (KFRI T9) and $3.00 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (KFRI T55) 4.19 and the mean transpiration was $3.83 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$.

Chlorophyll a content of the accessions ranged from KFRI T5 15.67 mg g^{-1} of KFRI T5 to 19.37 mg g^{-1} of KFRI T4. The mean chlorophyll a was 18.07 mg g^{-1} .

Values of chlorophyll b content ranged from 13.37 mg g^{-1} (KFRI T16) to 26.55 mg g^{-1} (KFRI T4) among the accessions. The mean chlorophyll b was 18.47 mg g^{-1} .

Total chlorophyll content was in a range of 29.12 mg g⁻¹ (KFRI T5) to 45.92 mg g⁻¹ (KFRI T4) and the average was found to be 36.55 mg g⁻¹.

In chlorophyll stability index, values of accessions ranged from 75.69 per cent of KFRI T1 to 91.90 per cent of KFRI T142 with overall mean of 84.27 per cent.

Cell membrane stability index values of the accessions ranged from 3.77 (KFRI T144) to 2.43 (KFRI T16). The average was found to be 3.08.

Table 6. Physiological parameters of plantlets of teak plus tree accessions after rehydration

SI No.	Accession	RWC	CATD	Pn	Gs	E	Chl.a	Chl.b	CSI	Total Chl.	CMSI
1	KFRI T1	83.5	-1.56	5.69	0.190	3.53	18.27	21.55	75.69	39.81	3.47
2	KFRI T4	80.2	-1.66	6.52	0.194	4.10	19.37	26.55	88.07	45.92	2.89
3	KFRI T5	79.5	-1.61	6.23	0.178	4.12	15.67	13.45	81.71	29.12	2.85
4	KFRI T9	80.6	-1.73	6.20	0.199	4.47	18.30	17.73	84.62	36.03	3.33
5	KFRI T16	81.5	-1.51	6.74	0.244	3.96	17.06	13.37	80.13	30.43	2.43
6	KFRI T24	81.9	-1.48	6.14	0.222	3.56	17.79	16.94	82.87	34.73	3.43
7	KFRI T116	76.6	-1.43	6.34	0.197	3.81	18.50	14.37	83.21	32.86	3.36
8	KFRI T44	72.8	-1.02	5.35	0.152	3.36	19.10	25.63	83.56	44.73	2.98
9	KFRI T55	68.9	-1.07	5.42	0.127	3.00	19.09	22.78	86.18	41.87	2.71
10	KFRI T142	77.8	-1.79	6.02	0.206	4.15	18.04	16.11	91.90	34.15	2.69
11	KFRI T144	79.1	-1.93	6.39	0.242	4.08	17.61	14.74	88.97	32.35	3.77
S. E		0.541	0.653	0.772	0.41	0.431	0.055	0.024	0.399	0.021	0.759
F-value		0.911	0.773	0.631	1.09	1.06	2.244	2.725	1.108	2.807	0.646

RWC = Relative water (%), CATD = Canopy air temperature differences ($^{\circ}\text{C}$), Pn = Photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), Gs = Stomatal conductance ($\text{mol m}^{-2} \text{ s}^{-1}$), E = Transpiration rate ($\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$) Chl. a = Chlorophyll a (mg g^{-1}), Chl. b = Chlorophyll b (mg g^{-1}), CSI = Chlorophyll stability index (mg g^{-1}), Total Chl. = Total chlorophyll (mg g^{-1}), CMSI = Cell membrane stability index (rate)

* Significant at 0.05 levels

Values with same superscript within a column are homogenous

4.3 ANALYSIS OF BIOCHEMICAL PARAMETERS

4.3.1 Biochemical parameters of plantlets of teak plus tree accessions

Biochemical parameters estimated from plantlets of teak plus tree accessions under normal nursery condition is presented in Table 7. There was significant difference among the different plus tree accessions for nitrate reductase activity (NRA), free amino acid (FAA), proline (Pro) and and peroxidase (PDX) ($P < 0.05$). However, the accessions did not differ in terms of total soluble protein content (TSP), glycine betaine (GB), super oxidase dismutase (SOD) and malondialdehyde (MDA).

Total soluble protein ranged from 54.76 mg g^{-1} of accession KFRI T142 to 78.70 mg g^{-1} of KFRI T16 with overall 65.78 mg g^{-1} .

High nitrate reductase activity (NRA) of $7.38 \text{ mmol nitrate g}^{-1}\text{hr}^{-1}$ was exhibited by the accession KFRI T55 which was on par with KFRI T1, T4, T5, T9, T16, T44, T116, T142 and T144. Low value of NRA was observed in KFRI T24 ($2.66 \text{ mmol nitrate g}^{-1}\text{hr}^{-1}$) which was also on par with KFRI T1, T4, T5, T9, T16, T44, T116, T142 and T144.

Lowest Free amino acid among the accessions was observed in KFRI T5 (3.853 mg g^{-1}) which significantly varied from all accession except KFRI T24 (4.736 mg g^{-1}). High quantity of free amino acid was observed in KFRI T1, T16, T44, T55, T116 and T142 which were on par with KFRI T4, T9 and T144.

For proline, significant variation was detected among the accessions. KFRI T55 was the maximum value ($5.47 \text{ } \mu\text{g g}^{-1}$) and was significantly differ from values of other accessions. The accession KFRI T24 ($1.55 \text{ } \mu\text{g g}^{-1}$) had the lowest value which was on par with KFRI T9 and KFRI T16. Intermediate proline content was observed in accessions KFRI T4 and T44 which were on par with KFRI T1, T5, T116, T142 and T144.

Content of Glycine betaine among the accessions ranged from 10.33 $\mu\text{mol g}^{-1}$ of KFRI T5 to 18.92 $\mu\text{mol g}^{-1}$ of KFRI T142. The overall mean was observed to be 14.604 $\mu\text{mol g}^{-1}$.

Values of total soluble sugar content ranged between 30.20 mg g^{-1} (KFRI T55) to 45.40 mg g^{-1} in (KFRI T24). Average TSS observed to be 38.619 mg g^{-1} .

Average super oxide dismutase content in the accessions was estimated to be 0.131 units $\text{protein}^{-1}\text{g}^{-1}$. SOD activity values ranged between 0.048 units $\text{protein}^{-1}\text{g}^{-1}$ to 0.181 units $\text{protein}^{-1}\text{g}^{-1}$ (KFRI T116 and T44 respectively).

Highest peroxidase was observed in KFRI T24 (0.062 $\text{min}^{-1}\text{mg}^{-1}\text{protein}$) which was on par with KFRI T9, KFRI T144, KFRI T5, KFRI T1, KFRI T4, and KFRI T55. While it significantly differed from KFRI T16, T44, T116 and T142. Low peroxidase activity among the accessions was observed in KFRI T16, T116 and T142.

The values of malondialdehyde ranged from 2.16 n mol g^{-1} (KFRI T144) to 3.34 n mol g^{-1} in (KFRI T44) with overall mean 2.573 n mol g^{-1} .

Table 7. Biochemical parameters of plantlets of teak plus tree accessions under normal condition

SI No	Accession	TSP	NRA	FAA	Pro	GB	TSS	SOD	PDX	MDA
1	KFRI T1	60.01	3.97 ^{ab}	11.68 ^a	3.05 ^{bc}	15.96	35.80	0.133	0.058 ^{ab}	2.60
2	KFRI T4	65.19	4.84 ^{ab}	9.911 ^{ab}	3.61 ^b	10.45	37.21	0.135	0.040 ^{abc}	2.39
3	KFRI T5	65.26	3.76 ^{ab}	3.853 ^c	2.69 ^{bcd}	10.33	39.15	0.138	0.041 ^{abc}	2.26
4	KFRI T9	73.17	5.94 ^{ab}	9.552 ^{ab}	1.75 ^{cd}	15.58	37.83	0.177	0.046 ^{abc}	2.43
5	KFRI T16	78.70	5.65 ^{ab}	15.09 ^a	2.03 ^{cd}	16.92	44.14	0.158	0.034 ^c	2.93
6	KFRI T24	66.91	2.66 ^b	4.736 ^{bc}	1.55 ^d	14.98	45.40	0.132	0.062 ^a	2.26
7	KFRI T44	69.79	4.93 ^{ab}	13.49 ^a	3.73 ^b	11.43	45.35	0.181	0.037 ^{bc}	3.34
8	KFRI T55	54.91	7.38 ^a	12.04 ^a	5.47 ^a	12.64	30.20	0.063	0.040 ^{abc}	2.33
9	KFRI T116	77.98	4.55 ^{ab}	14.33 ^a	2.48 ^{bcd}	17.86	34.66	0.048	0.035 ^c	2.26
10	KFRI T142	54.76	4.00 ^{ab}	11.56 ^a	3.13 ^{bc}	18.92	40.81	0.097	0.035 ^c	3.04
11	KFRI T144	56.99	4.82 ^{ab}	9.753 ^{ab}	2.67 ^{bcd}	15.58	34.27	0.173	0.039 ^{abc}	2.16
S. E		0.987	0.042	0.044	0.000	0.999	0.320	0.475	0.002	0.244
F-value		0.245	2.393*	2.364*	15.859*	0.132	1.242	0.997	4.267*	1.401

TSP = Total soluble protein (mg g^{-1}), NRA = Nitrate reductase ($\text{mmol nitrate g}^{-1}\text{hr}^{-1}$), FAA = Free amino acid (mg g^{-1}), Proline ($\mu\text{g g}^{-1}$), GB = Glycine betaine ($\mu\text{mol g}^{-1}$), TSS = Total soluble sugar (mg g^{-1}), SOD = Superoxide dismutase ($\text{units protein}^{-1}\text{g}^{-1}$), PDX = Peroxidase ($\text{min}^{-1} \text{mg}^{-1}$ protein), MDA = Malondialdehyde (n mol g^{-1}).

* Significant at 0.05 levels

Values with same superscript in column are homogenous

4.3.2 Biochemical parameters of plantlets of teak plus tree accessions after drought induction

Observations on different biochemical parameters among plantlets of teak plus tree accessions after induction of drought stress is given in Table 8. Statistical analysis showed significant differences was observed among the accessions for proline (Pro), super oxide dismutase (SOD), peroxidase (PDX) and malondialdehyde (MDA). No difference were noticed in total soluble protein (TSP), nitrate reductase activity (NRA), free amino acid (FAA), glycine betaine (GB) and total soluble sugar (TSS).

Total soluble protein of the tested accessions ranged between 28.03 mg g⁻¹ and 39.82 mg g⁻¹ of KFRI T9 and T1 respectively, with overall mean 33.8 mg g⁻¹.

Nitrate reductase activity values of the accessions ranged between 1.74 mmol nitrate⁻¹hr⁻¹ and 4.08 mmol nitrate⁻¹hr⁻¹ (KFRI T24 and T144 respectively) with overall mean of 3.26 mmol nitrate⁻¹hr⁻¹.

The amounts of free amino acid in the tested accessions ranged from 11.24 mg g⁻¹ to 28.42 mg g⁻¹ of KFRI T24 and T44, respectively with overall mean 22.10 mg g⁻¹.

Accession KFRI T55 had high amount of proline (12.55 µg g⁻¹) which was on par with KFRI T1, T4, T16, T24, and T44. The accessions KFRI T5, T9, T116, T142 and T144 had low amount of proline which were also on par with KFRI T1, T4, T16, T24, and T44.

Glycine betaine values among the tested accessions ranged between 35.20 µmol g⁻¹ and 18.48 µmol g⁻¹ of KFRI T144 and T4 respectively with overall mean 26.24 µmol g⁻¹.

Total soluble sugar content of the accessions ranged between 45.27 mg g⁻¹ and 65.84 mg g⁻¹ of KFRI T144 and T24, respectively. The overall mean value for total soluble sugar among the accessions was 54.48 mg g⁻¹.

Lowest amount of peroxidase of $0.045 \text{ min}^{-1} \text{ mg}^{-1}$ protein was estimated in accession KFRI T116 and was significantly different from all other Intermediate values for peroxidase was observed in accessions KFRI T4, T9, T16, T44, T55, T142 and T144. The highest amount of PDX was present in KFRI T24 ($0.085 \text{ min}^{-1} \text{ mg}^{-1}$ protein) which was on par with KFRI T1 and T5 ($0.081 \text{ min}^{-1} \text{ mg}^{-1}$ protein and $0.077 \text{ min}^{-1} \text{ mg}^{-1}$ protein respectively).

For super oxide dismutase, the lowest amount was noticed in accession KFRI T9 ($0.380 \text{ units protein}^{-1} \text{ g}^{-1}$) which was on par with accessions KFRI T1, T24, T44, T55, T116 and T142. High amount of SOD was observed in KFRI T4, T5, T16 and T144 which was also on par with KFRI T1, T24, T44, T55, T116 and T142.

The high malonidialdehyde content was present in accession KFRI T4 and T44 ($7.43 \text{ n mol g}^{-1}$ and $7.51 \text{ n mol g}^{-1}$ respectively) which were on par with KFRI T1, T9, T16, T55, T116 and T142. Accessions KFRI T5 and T144 recorded values for malonidialdehyde content in ($3.11 \text{ n mol g}^{-1}$ and $3.37 \text{ n mol g}^{-1}$ respectively).

Table 8. Biochemical parameters of plantlets of teak plus tree accessions after drought induction

SI No	Accession	TSP	NRA	FAA	Proline	GB	TSS	SOD	PER	MDA
1	KFRI T1	39.82	3.24	23.40	6.433 ^{ab}	18.94	46.37	0.480 ^{ab}	0.081 ^{ab}	4.10 ^{abc}
2	KFRI T4	34.36	2.19	24.20	7.549 ^{ab}	18.48	57.40	0.488 ^a	0.064 ^{cd}	7.43 ^a
3	KFRI T5	31.62	2.78	18.90	5.901 ^b	21.32	61.50	0.486 ^a	0.077 ^{ab}	3.11 ^c
4	KFRI T9	28.03	3.52	27.41	5.539 ^b	26.31	48.05	0.380 ^b	0.064 ^{cd}	6.07 ^{abc}
5	KFRI T16	38.42	3.62	20.35	9.874 ^{ab}	25.66	52.15	0.499 ^a	0.064 ^{cd}	4.55 ^{abc}
6	KFRI T24	35.07	1.74	11.24	6.578 ^{ab}	27.66	65.84	0.454 ^{ab}	0.085 ^a	4.57 ^{bc}
7	KFRI T44	32.63	3.76	28.42	8.941 ^{ab}	22.41	56.22	0.429 ^{ab}	0.057 ^d	7.51 ^a
8	KFRI T55	28.46	3.94	20.47	12.55 ^a	29.84	50.85	0.442 ^{ab}	0.072 ^{bc}	7.27 ^{ab}
9	KFRI T116	37.01	4.02	23.32	6.003 ^b	29.18	53.82	0.443 ^{ab}	0.045 ^e	4.91 ^{abc}
10	KFRI T142	35.58	3.00	24.24	4.809 ^b	33.64	61.75	0.453 ^{ab}	0.072 ^{bc}	4.58 ^{abc}
11	KFRI T144	31.55	4.08	21.19	6.003 ^b	35.20	45.27	0.492 ^a	0.071 ^{bc}	3.37 ^c
S. E		0.050	0.252	0.007	0.006	0.938	0.203	0.019	0.000	0.001
F-table		2.295	1.382	3.454	3.542*	0.389	1.506	2.878*	28.22*	5.022*

TSP = Total soluble protein (mg g^{-1}), NRA = Nitrate reductase ($\text{mmol nitrate g}^{-1}\text{hr}^{-1}$), FAA = Free amino acid (mg g^{-1}), Proline ($\mu\text{g g}^{-1}$), GB = Glycine betaine ($\mu\text{mol g}^{-1}$), TSS = Total soluble sugar (mg g^{-1}), SOD = Superoxide dismutase ($\text{units protein}^{-1}\text{g}^{-1}$), PER = Peroxidase ($\text{min}^{-1}\text{mg}^{-1}\text{protein}$), MDA = Malondialdehyde (n mol g^{-1}).

* Significant at 0.05 levels

Values with same superscript in coloumn are homogenous

4.3.3 Biochemical parameters of plantlets of teak plus tree accessions after re-watering

Observations taken after re-watering of the plantlets of teak accessions after drought induction is presented in Table 9. The accessions differed significantly in terms of nitrate reductase activity, free amino acid, proline and peroxidase activity. The accessions did not differ with respect to the characters total soluble protein, glycine betaine, total soluble sugar, malondialdehyde and super oxide dismutase.

Total soluble protein of the accessions ranged between 37.45 mg g⁻¹ (KFRI T55) and 72.68 mg g⁻¹ (KFRI T116) with overall mean 57.90 mg g⁻¹.

Low nitrate reductase activity, was observed in accessions KFRI T5, T124 and T142 which were on par with accessions KFRI T1, T4, T9, T16, T44, T116 and T144. High nitrate reductase activity was observed in accession KFRI T55 which was also on par with KFRI T1, T4, T9, T16, T44, T116 and T144.

The high free amino acid content among the accessions was observed in KFRI T44, T116 and T142 which were on par with KFRI T4, T9, T16, T44 and T55. Low free amino acid content was observed in KFRI T5 which also was on par with KFRI T9, T16, T24, T144 and T55.

Low amount of proline was observed in accessions KFRI T5, T9, T116, T142 and T144 which was on par with KFRI T1, T4, T16, T24 and T44. High amount of proline was observed in KFRI T55 which was also on par with KFRI T1, T4, T16, T24 and T44.

The glycine betaine content of the accessions ranged from 25.45 µmol g⁻¹ in (KFRI T116) to 14.50 µmol g⁻¹ (KFRI T55). The overall mean value of glycine content among the accessions was 19.45 µmol g⁻¹.

Average total soluble sugar among the accessions after re-watering was estimated to be 45.80 mg g⁻¹. The lowest TSS was observed in KFRI T1 (38.91 mg g⁻¹) and KFRI T24 had the highest TSS amount (51.63 mg g⁻¹).

Super oxide dismutase activity of the accessions ranged between 0.054 units protein⁻¹g⁻¹ in (KFRI T144) to 0.283 units protein⁻¹g⁻¹ in (KFRI T5) with overall mean 0.20 units protein⁻¹g⁻¹ were at par.

The lowest amount of peroxidase activity of 0.038 min⁻¹ mg⁻¹ protein was observed in accession KFRI T116 which was on par with KFRI T4, T5, T16, T44, T55 and T142. KFRI T24 had the highest amount (0.067 min⁻¹ mg⁻¹ protein) of peroxidase activity which was on par with KFRI T1 and T9. Rest of the accessions had intermediate peroxidase activity.

Values of malondialdehyde ranged from 2.49 n mol g⁻¹ (KFRI T55) to 4.18 n mol g⁻¹ (KFRI T9) with overall mean 3.39 n mol g⁻¹.

Table 9. Biochemical parameters of plantlets of teak plus trees accessions after re-watering.

SI No	Accession	TSP	NRA	FAA	Proline	GB	TSS	SOD	PER	MDA
1	KFRI T1	52.61	3.84 ^{ab}	16.82 ^{ab}	5.570 ^{ab}	18.04	38.91	0.282	0.059 ^{ab}	2.97
2	KFRI T4	65.56	3.61 ^{ab}	16.58 ^{ab}	6.536 ^{ab}	15.10	46.20	0.166	0.048 ^{bcd}	3.77
3	KFRI T5	65.19	2.46 ^b	5.539 ^c	5.109 ^b	17.09	51.59	0.283	0.042 ^{cd}	2.62
4	KFRI T9	64.04	4.41 ^{ab}	15.49 ^{abc}	4.795 ^b	20.07	41.99	0.120	0.060 ^{ab}	4.18
5	KFRI T16	71.94	3.08 ^{ab}	16.01 ^{abc}	8.549 ^{ab}	21.55	48.89	0.159	0.041 ^{cd}	3.97
6	KFRI T24	64.97	1.95 ^b	6.903 ^{bc}	5.695 ^{ab}	21.78	51.63	0.224	0.067 ^a	2.96
7	KFRI T44	59.65	4.15 ^{ab}	22.39 ^a	7.741 ^{ab}	15.87	49.16	0.275	0.041 ^{cd}	3.86
8	KFRI T55	37.45	5.93 ^a	15.17 ^{abc}	10.87 ^a	14.50	40.93	0.265	0.044 ^{cd}	2.49
9	KFRI T116	72.68	3.94 ^{ab}	18.42 ^a	5.197 ^b	25.45	48.07	0.239	0.038 ^d	3.75
10	KFRI T142	53.33	2.54 ^b	18.38 ^a	4.164 ^b	22.30	45.04	0.092	0.045 ^{cd}	3.66
11	KFRI T144	39.31	3.22 ^{ab}	13.81 ^{abc}	5.197 ^b	22.17	41.41	0.054	0.051 ^{bc}	3.06
S. E		0.394	0.014	0.024	0.006	0.999	0.297	0.090	0.000	0.063
F-table		1.116	3.044*	2.725*	3.542*	0.132	1.286	1.965	13.667	2.161

TSP = Total soluble protein (mg g^{-1}), NRA = Nitrate reductase ($\text{mmol nitrate g}^{-1}\text{hr}^{-1}$), FAA = Free amino acid (mg g^{-1}), Proline ($\mu\text{g g}^{-1}$), GB = Glycine betaine ($\mu\text{mol g}^{-1}$), TSS = Total soluble sugar (mg g^{-1}), SOD = Superoxide dismutase ($\text{units protein}^{-1}\text{g}^{-1}$), PER = Peroxidase ($\text{min}^{-1} \text{mg}^{-1} \text{protein}$), MDA = Malondialdehyde (n mol g^{-1}).

* Significant at 0.05 levels

Values with same superscript within a column are homogenous

4.4 EFFECT DROUGHT TREATMENT ON PLANTLETS OF TEAK PLUS TREE ACCESSIONS

4.4.1 Effect of drought on physiological and biochemical parameters of teak plus tree accessions

Results obtained from pair t-test for different physiological and biochemical characters of plantlets of teak plus tree accessions showed that there was highly significant difference between the treatments (normal, drought stress and recovery after drought stress by re watering) The data are presented in Table 10.

Table 10. Effect drought stress and their combination on various growth, physiological and biochemical parameters of teak plus tree accessions.

Parameter	N X D	N X R	R X D
Relative water content (%)	12.0**	4.01**	10.1**
Canopy air temperature differences ($^{\circ}\text{C}$)	14.3**	6.28**	7.73**
Photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)	17.2**	5.32**	9.26**
Transpiration rate ($\text{mol m}^{-2} \text{ s}^{-1}$)	11.1**	6.85**	11.0**
Stomatal conductance ($\text{mol m}^{-2} \text{ s}^{-1}$)	25.5**	10.0**	21.9**
Chlorophyll a (mg g^{-1})	5.24**	4.23**	3.94**
Chlorophyll b (mg g^{-1})	8.24**	5.10**	6.88**
Chlorophyll stability index (mg g^{-1})	13.8**	7.74**	9.58**
Total chlorophyll content (mg g^{-1})	8.05**	5.21**	6.46**
Cell membrane stability index	16.5**	15.2**	7.94**
Total soluble protein (mg g^{-1})	7.01**	4.31**	6.20**
Nitrate reductase activity ($\text{mmol nitrate g}^{-1} \text{ hr}^{-1}$)	6.19**	6.34**	4.17**
Free amino acid (mg g^{-1})	9.78**	6.08**	7.84**
Proline ($\mu\text{g g}^{-1}$)	8.04**	7.32**	10.5**
Glycine betaine (mg g^{-1})	8.56**	8.33**	5.00**
Total soluble sugar (mg g^{-1})	9.43**	6.48**	6.85**
Super oxide dismutase ($\text{units protein}^{-1} \text{ g}^{-1}$)	21.0**	4.69**	11.2**
Peroxidase ($\text{min}^{-1} \text{ mg}^{-1} \text{ protein}$)	10.7**	4.90**	7.46**
Malondialdehyde (n mol g^{-1}).	5.37**	5.56**	3.72**

N= Normal condition D = Drought condition R = Recovery condition

**Significant at 0.01 levels

4.4.2 Effect of drought stress on physiological and biochemical characters of plantlets of teak plus tree accessions

Induction of drought resulted in reduction in most of the physiological parameters of the plantlets of teak plus tree accessions. Out of ten physiological parameters observed only canopy air temperature differences ($^{\circ}\text{C}$) and Cell membrane stability index (rate) increased under drought induction. All other parameters got reduced.

Relative water content decreased in all accessions under drought stress. The percentage decrease varied between 17.8 per cent and 44.6 per cent of accessions KFRI T9 and KFRI T5 respectively. Accessions KFRI T44, T55 and T144 partially recovered after five days of rehydration while other accessions showed almost complete recovery (Figure 1a).

In case of difference in canopy air temperature there was an increase in all the accessions. The percentage increase in difference in canopy air temperature varied between 74.8 per cent of KFRI T144 and 138.6 per cent of KFRI T44. When complete recovery shown by six accessions other accessions (KFRI T9, KFRI T16, KFRI T24, KFRI T44 and KFRI T55) were not able to recover fully after rehydration (Figure 1b).

Two consecutive severe droughts resulted in significant reduction in the net photosynthetic rate among the accessions. Percentage reduction in photosynthetic rate was between 39.8 per cent of KFRI T9 and 52.8 per cent of KFRI T116. Accessions KFRI T4, T5, T16, T24 and T116 recovered almost fully after rehydration while in all others recovery after drought stress with respect to rate of photosynthesis was not full (Figure 2a).

Percentage reduction in stomatal conductance under drought stress ranged from 74.6 per cent of KFRI T116 to 95.7 per cent of (KFRI T5). None of the accessions was able to recover fully after rehydration (Figure 2b).

Transpiration rate also decreased considerably under drought condition and the percentage decrease among the accessions ranged from 76.8 per cent of KFRI T116 and 92.7 per cent of KFRI T5. Five days of rehydration resulted in improvement of transpiration rate in all the accessions. However, none of the accessions recovered fully after rehydration (Figure 2c).

Percentage decrease in content of chlorophyll a in the tested accessions ranged from 4.5 per cent of KFRI T55 to 41.4 per cent of KFRI T16. After five days of re-watering all accessions showed recovery. All accessions except KFRI T5 showed almost full recovery with a comparable value of chlorophyll a content with that of normal irrigated condition (Figure 3a).

There was severe reduction in chlorophyll b in all accessions under drought stress and the percentage reduction ranged from 20.3 per cent of KFRI T24 to 87.1 per cent of KFRI T16. Five days of re-watering resulted in incomplete recovery in all the accessions except KFRI T4 and T24 which showed full recovery after rehydration (Figure 3b).

Percentage reduction in total chlorophyll content under drought among the accessions ranged between 15.2 per cent to 60.8 per cent (KFRI T24 and T16 respectively). Re-watering resulted in recover almost near to the level of normal irrigated condition in accessions of KFRI T4, T9 and T124 while other accessions partial were recovered (Figure 3c).

Compared to other physiological parameters the percentage reduction in chlorophyll stability index was less. It ranged between 15.9 per cent and 30.4 per cent (KFRI T4 and T44 respectively). Incomplete recovery was observed in six accessions after rehydration while other accessions including KFRI T4, T24, T55, T142 and T144 were almost near to the level of normal irrigated condition (Figure 3d).

In response to drought stress, total soluble protein decreased drastically in all accessions and the percentage reduction was in a range of 50.7 per cent to 161 per cent (KFRI T1 and T9 respectively). After rehydration complete recovery was observed in KFRI T5, T16, T24, T116 and T142, and in all other accessions recovery was partial (Figure 4a).

Nitrate reductase activity also got reduced under drought in all the accessions and the percentage reduction ranged between 15.5 per cent of KFRI T116 to 121.5 per cent of KFRI T4. The nitrate reductase activity after rehydration got increased from that under stress condition in all accessions indicating recovery from moisture stress. Only KFRI T1 showed complete recovery (Figure 4b).

Exposing plantlets of teak plus tree accessions to drought lead to significant accumulation of free amino acid, and the accumulation rate was between 0.35 times and 3.91 times (KFRI T16 and T5). Re-watering resulted in intermediate amount of free amino acid compared to normal and stressed plantlets indicating recovery of the plantlets in all the accessions. However only accession KFRI T16 recovered fully (Figure 5a).

Enhanced production of proline was observed in plantlets of all the accessions under stress condition and enhancement ranged between 1.09 times and 3.86 times (KFRI T4 and T16 respectively). Rehydration of the stressed plants resulted in reduction in proline content. However, none of the accessions recovered fully (Figure 5a).

Glycine betaine also got increased under moisture stress condition in all the teak accessions. The percentage increase ranged from 18.7 per cent and 136.1 per cent (KFRI T1 and T55 respectively). Rehydration of the accessions reduced the Glycine betaine content in all the accessions indicating partial recovery from moisture stress (Figure 5c).

Total soluble sugar also got increased in the plantlets of teak plus tree accessions under moisture stress and the percentage increase ranged between

18.2 per cent and 68.4 per cent of KFRI T16 and T55, respectively. Five days of re-watering of the accessions resulted in partial recovery with intermediate values for total soluble sugar under normal and stressed condition (Figure 5d).

Activity of enzyme super oxide dismutase (SOD) was found to increase significantly in all accessions after exposing to drought. Super oxide dismutase increased significantly from 1.37 times to 8.16 times in accessions KFRI T44 and T116, respectively. Rehydration of the accessions showed that there was complete recovery in accessions KFRI T16 and T142 with lesser quantity of SOD accumulated compared to normal condition. All other accessions showed only partial recovery (Figure 6a).

Induction of moisture stress resulted in increase of enzyme peroxidase in all the accessions. The percentage increase ranged between 30.4 per cent of KFRI T116 to and 106.6 per cent of KFRI T142. After rehydration the enzyme activity in all the accessions got reduced indicating recovery from moisture stress. Five of the entries KFRI T1, T5, T24, T55 and T116 showed almost complete recovery (Figure 6b).

Drought stress increased content of malondialdehyde in all the accessions and the percentage increase was between 37.6 per cent and 211.6 per cent of accessions KFRI T5 and T55 respectively. Intermediate content of malondialdehyde observed in all the accessions after rehydration showed that none of the accessions recovered fully from the drought stress with respect to content of malondialdehyde except KFRI T55 (Figure 7a).

Exposing plantlets of teak plus tree accessions to drought lead to significant increase of cell membrane stability index, and the increase rate was between 1.3 times and 15.1 times (KFRI T1 and T16). Re-watering resulted in intermediate rate of cell membrane stability to normal and stressed plantlets indicating recovery of the plantlets in all the accessions. However, none of the accessions recovered fully (Figure 7b).

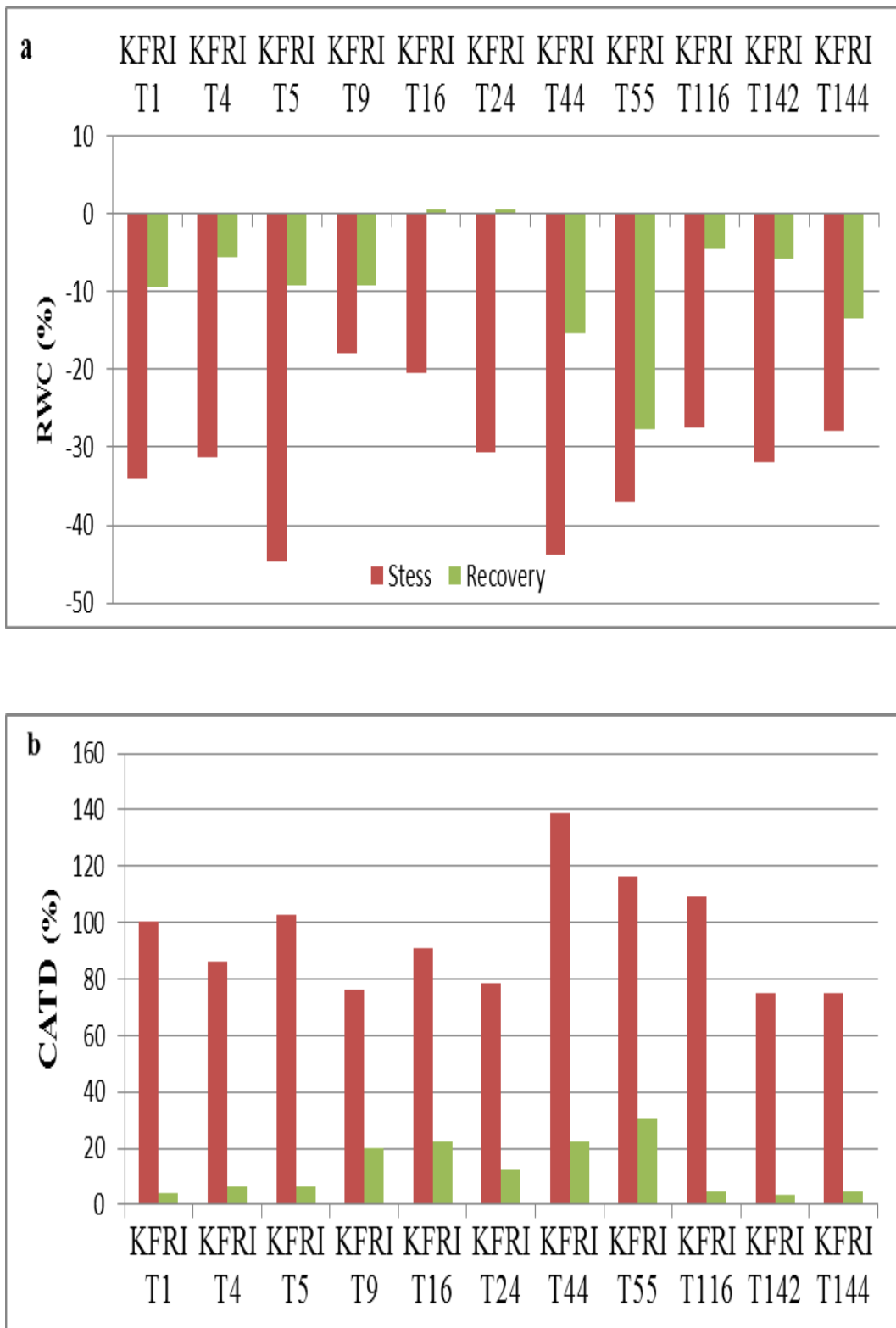


Figure 1 a-b. Change in relative water content and canopy air temperature differences among *Tectona grandis* plus tree accessions under water stress and rehydration.

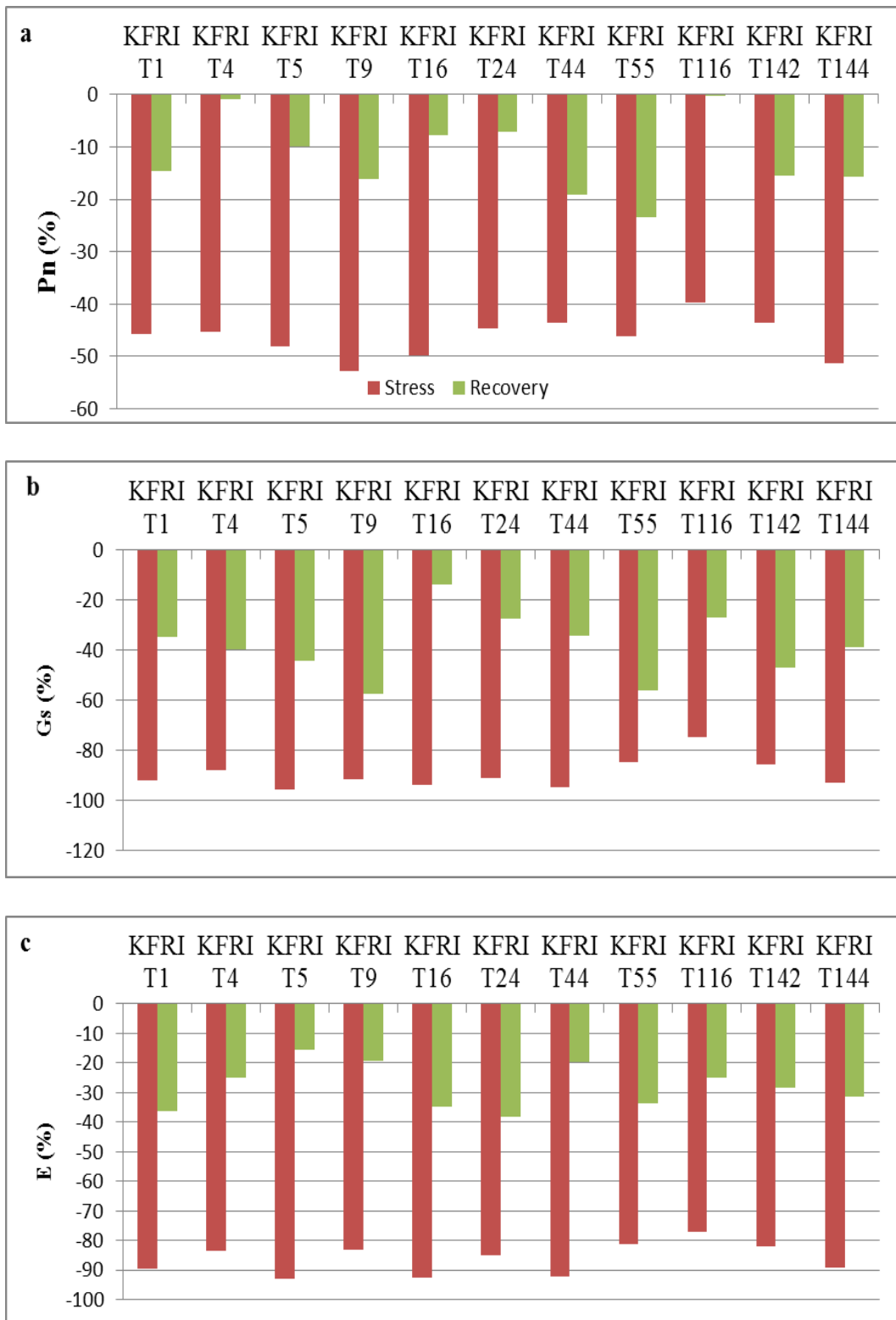


Figure 2 a-c. Change in photosynthetic rate, stomata conductance and transpiration among *Tectona grandis* plus tree accessions under water stress and rehydration.

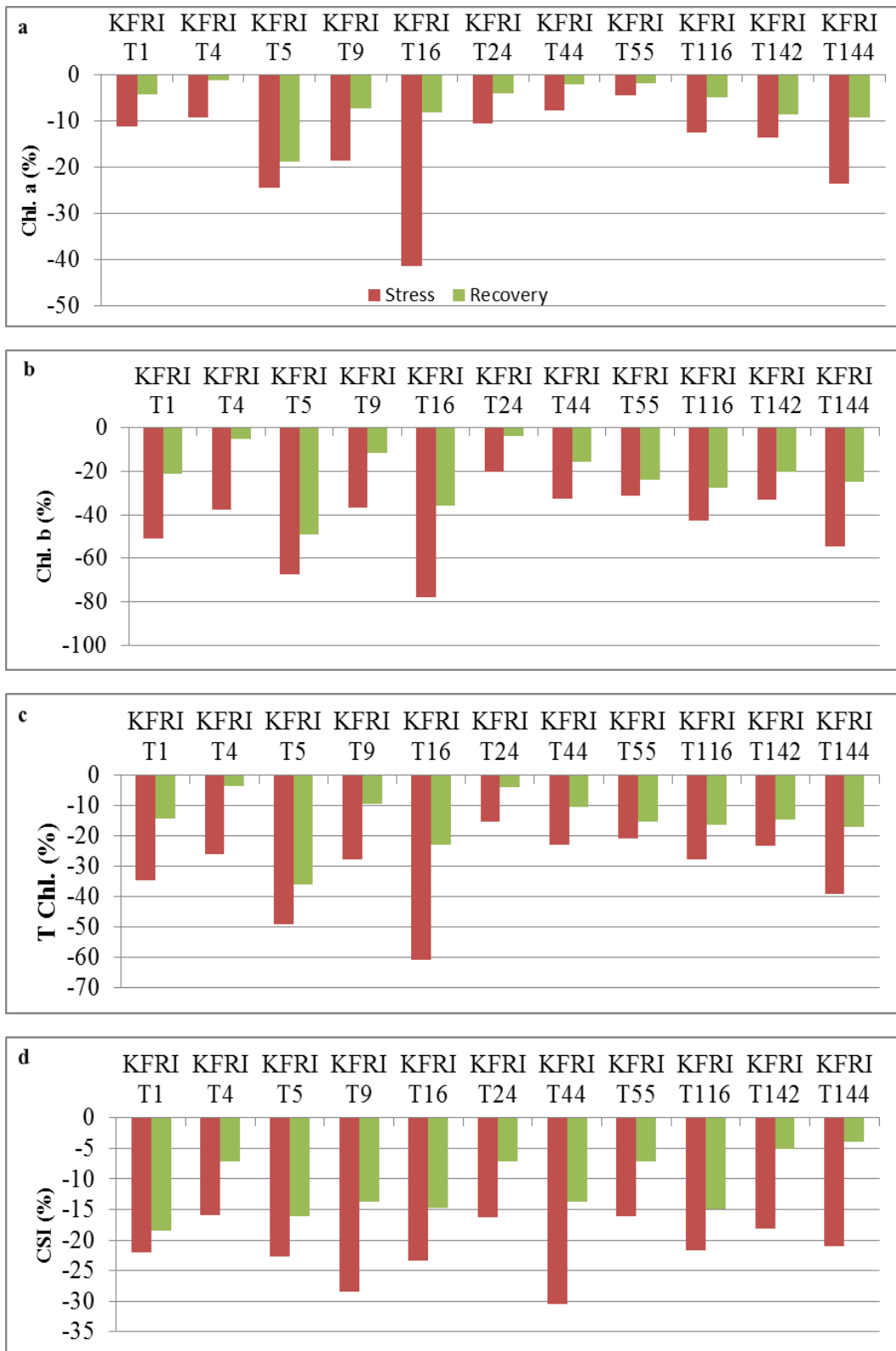


Figure 3 a-d. Change in photosynthetic pigments and chlorophyll stability index among *Tectona grandis* plus tree accessions under water stress and rehydration.

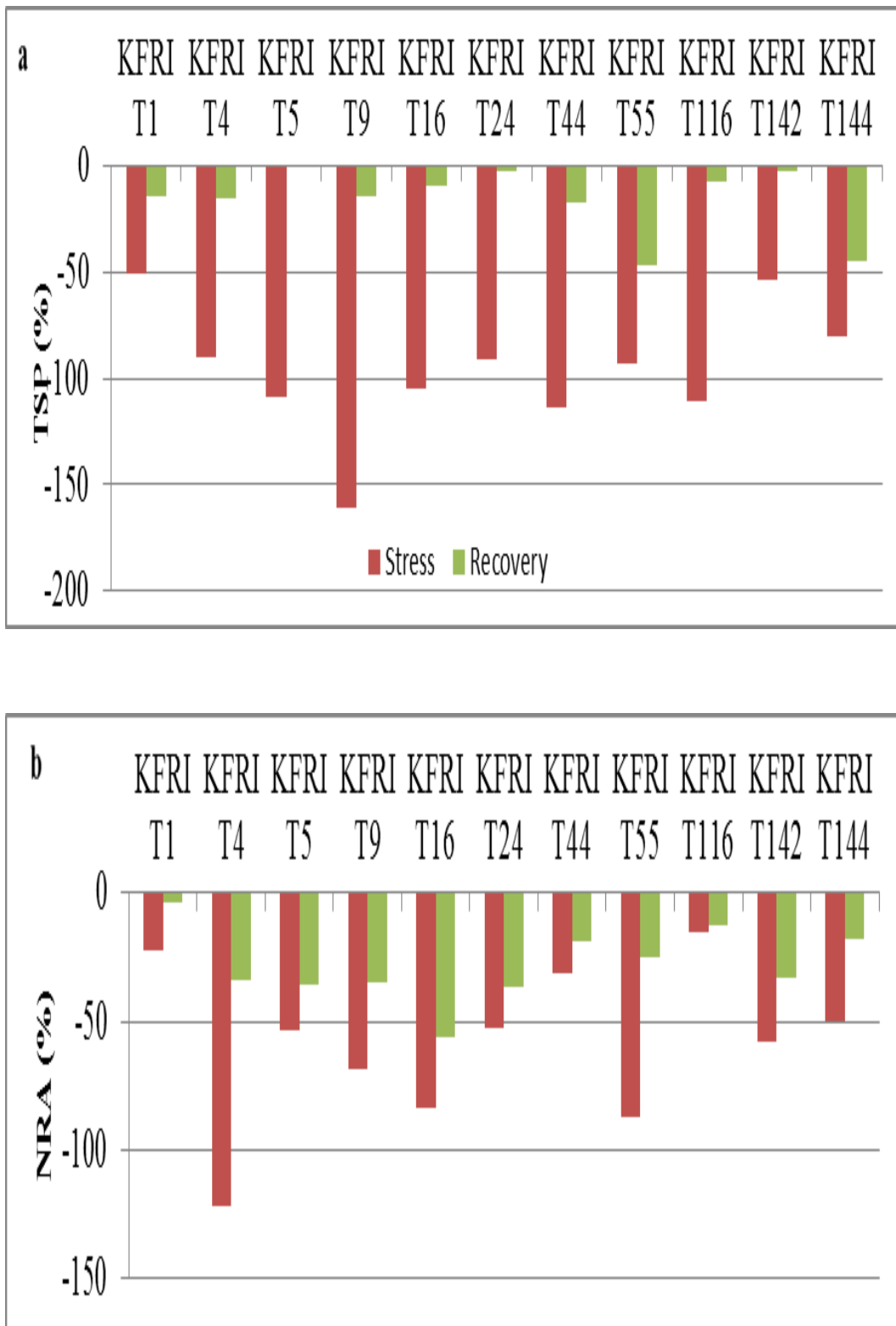


Figure 4 a-b. Change in total soluble protein and nitrate reductase among *Tectona grandis* plus tree accessions under water stress and rehydration.

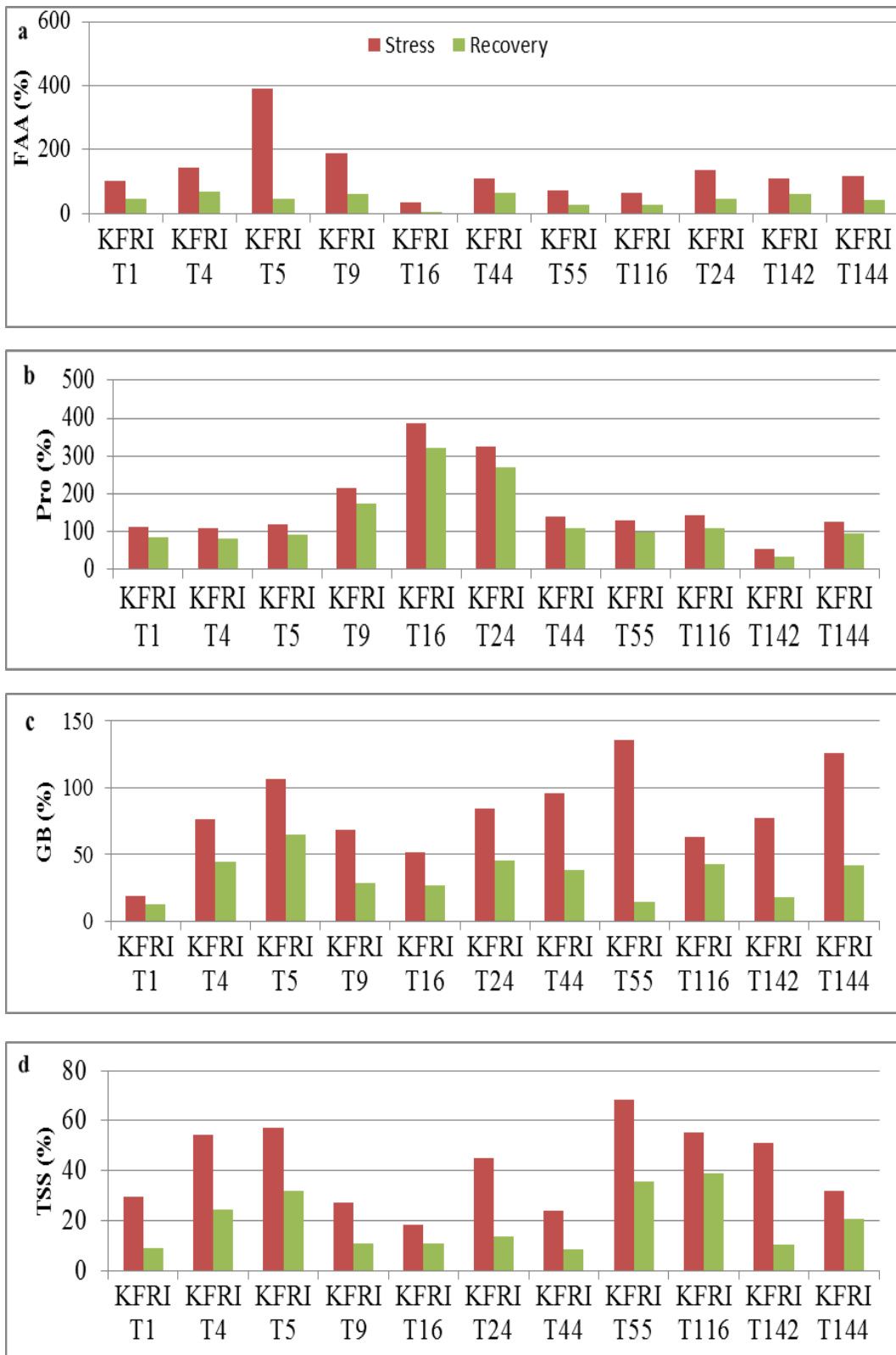


Figure 5 a-d. Change in free amino acid, proline, glycine betaine and total soluble sugar among *Tectona grandis* plus tree accession under water stress and rehydration.

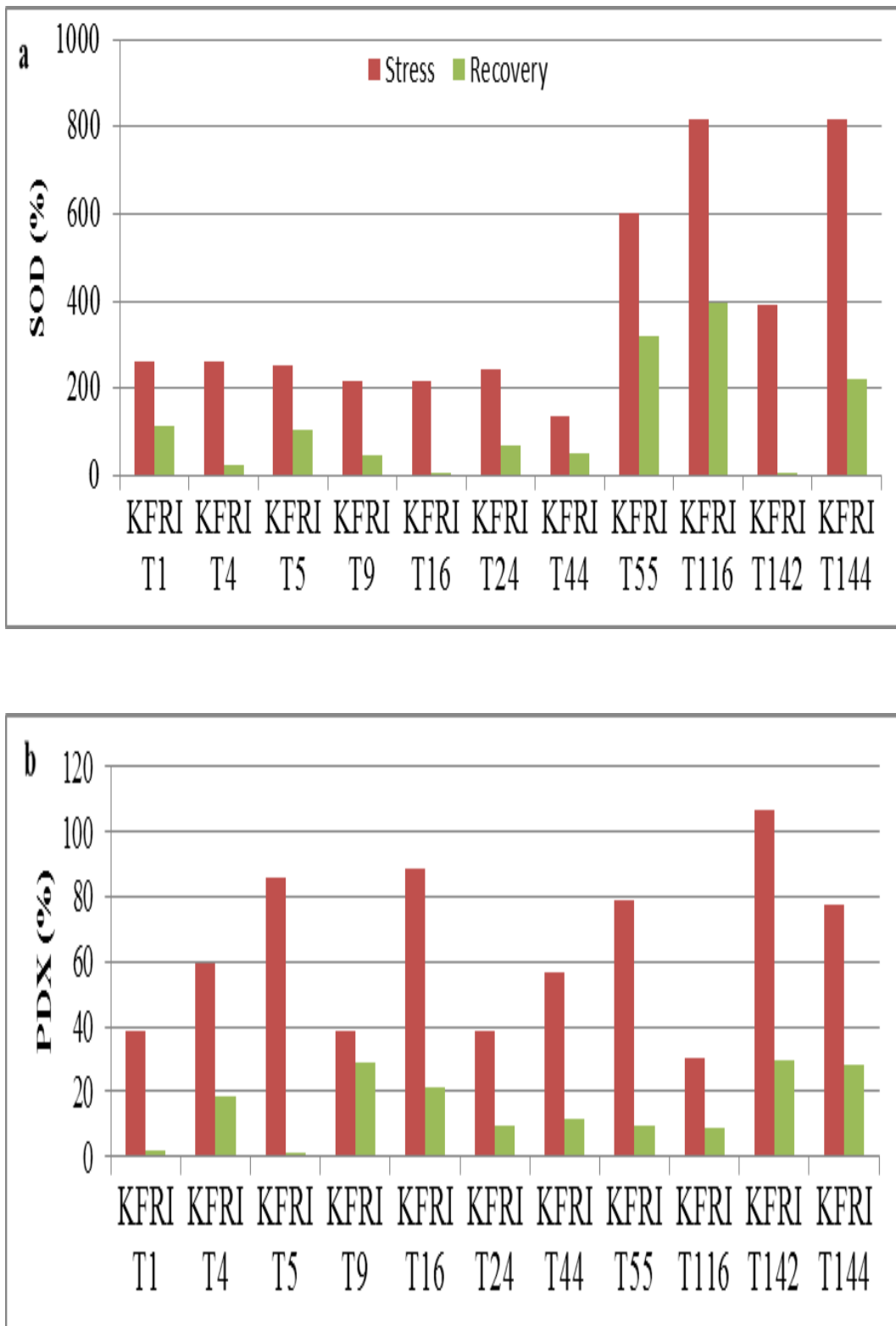


Figure 6 a-b. Change in activity of super oxide dismutase and peoxidase among *Tectona grandis* plus tree accessions under water stress and rehydration.

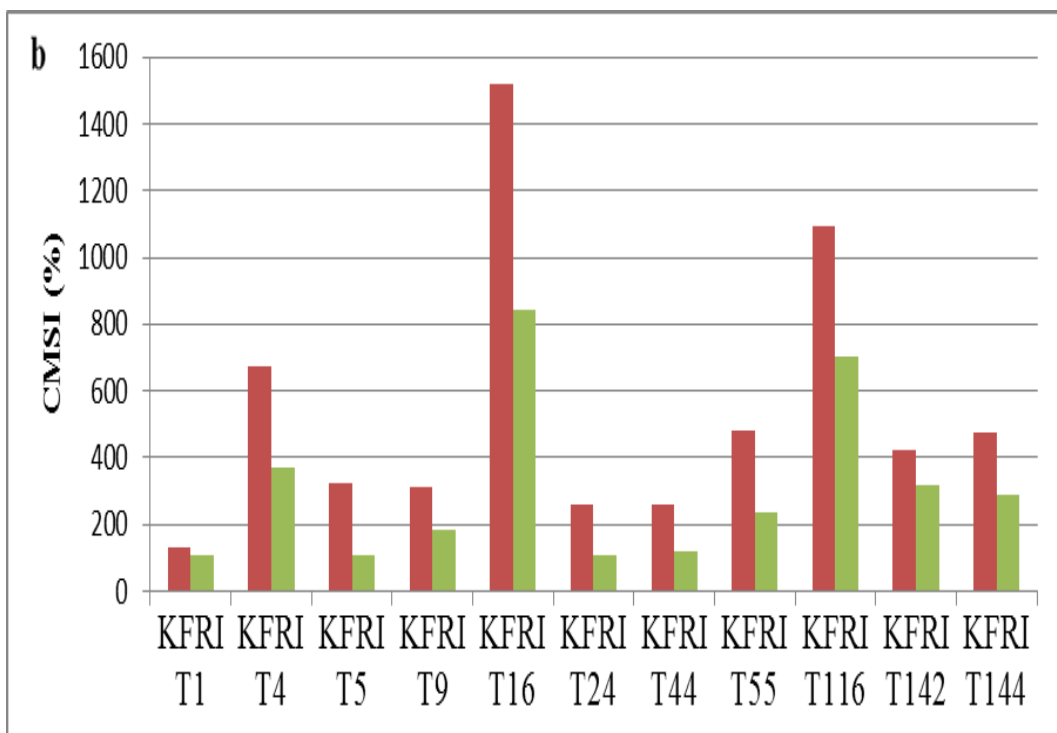
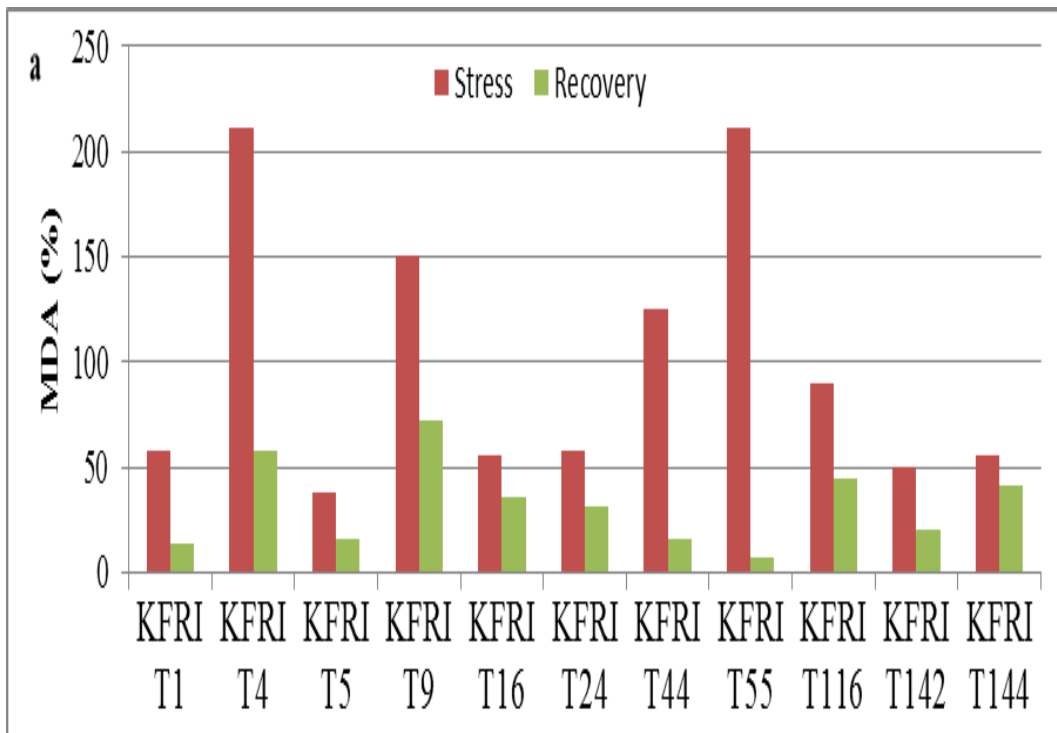


Figure 7 a-b. Change in content of malondialdehyde and cell membrane stability index among *Tectona grandis* plus tree accessions under water stress and rehydration.

4.5 CORRELATION STUDIES

4.5.1 Correlations of physiological and biochemical characters with number of leaves during moisture stress

Correlation of physiological and biochemical characters with number of leaves during moisture stress treatment was calculated and presented in Table 11.

Among the twenty characters used for estimating the correlation under drought stress in teak plus tree accessions, leaf number showed positive correlation with content of chlorophyll a (0.806), chlorophyll b (0.690) and total chlorophyll content (0.806).

Relative water content had significant positive correlation with cell membrane stability index (0.602).

Canopy air temperature difference was negatively correlated with total soluble sugar (-0.609).

Rate of photosynthesis exhibited highly significant positive correlation with transpiration rate (0.564) and positive and significant correlation with chlorophyll stability index (0.613).

Stomatal conductance had highly significant and positive correlation with transpiration rate (0.936) and significant positive correlation with chlorophyll stability index (0.651).

Transpiration rate was positively correlated with chlorophyll stability index (0.621).

Chlorophyll a content was positively correlated with chlorophyll b content (0.903) and content of malondialdehyde (0.544).

Positive correlation was observed between chlorophyll b content, cell membrane index (0.991) and content of malondialdehyde (0.764).

Nitrate reductase activity was negatively correlated with total soluble sugar (-0.725) and peroxidase content (-0.570).

There existed positive correlation between content of proline and content of malondialdehyde (0.553).

Content of glycine was negatively correlated with peroxidase content (-0.702) and positively correlated with malondialdehyde (0.577).

Correlation of peroxidase with malondialdehyde was negative (-0.553). All other inter correlations between the physiological and biochemical characters of the teak plus tree accessions under moisture stress were non-significant.

Table 11: Correlation and inter correlation of physiological and biochemical characters of plantlets of teak at drought condition.

	X1	X2	X3	X4	X5	X6	X7	X8	X9	X10	X11	X12	X13	X14	X15	X16	X17	X18	X19	X20
X1	1																			
X2	-0.297	1																		
X3	0.005	-0.333	1																	
X4	-0.062	0.355	0.386	1																
X5	-0.063	0.230	0.564*	0.936**	1															
X6	-0.096	-0.351	0.278	0.419	0.411	1														
X7	0.032	-0.402	0.194	0.177	0.158	0.903*	1													
X8	0.109	-0.063	0.456	0.651*	0.621*	0.294	0.152	1												
X9	-0.007	-0.395	0.225	0.256	0.240	0.952*	0.991*	0.152	1											
X10	0.602*	-0.193	-0.419	-0.191	-0.299	-0.047	-0.073	-0.116	-0.066	1										
X11	0.136	0.038	-0.245	-0.006	-0.038	-0.289	-0.27	-0.431	-0.281	0.070	1									
X12	-0.005	0.214	0.229	-0.083	0.180	-0.100	0.764**	-0.352	-0.092	-0.195	-0.118	1								
X13	0.232	0.262	0.613*	0.424	0.475	-0.065	-0.172	0.294	-0.142	0.013	-0.306	0.388	1							
X14	0.363	-0.180	0.041	-0.267	-0.19	0.043	0.378	-0.046	0.282	-0.062	-0.252	0.278	-0.098	1						
X15	-0.398	0.125	0.042	0.008	0.168	0.256	0.278	-0.274	0.277	-0.497	0.138	0.513	-0.166	-0.057	1					
X16	0.399	-0.609*	0.265	0.082	-0.014	0.119	0.123	0.362	0.125	0.217	0.298	-0.725**	-0.083	-0.170	-0.449	1				
X17	-0.213	-0.201	-0.028	-0.424	-0.371	-0.466	-0.440	0.186	-0.458	0.022	-0.326	-0.199	-0.194	0.068	-0.373	0.053	1			
X18	0.044	-0.244	-0.127	-0.253	-0.423	0.016	-0.024	0.064	-0.012	0.269	-0.354	-0.570*	-0.048	-0.143	-0.702**	0.344	0.253	1		
X19	0.064	-0.094	0.029	0.137	0.171	0.544*	0.764**	0.044	0.712*	-0.242	0.001	0.166	-0.259	0.553*	0.577*	-0.114	-0.421	-0.553*	1	
X20	-0.178	-0.498	0.235	0.159	0.298	0.806**	0.690**	-0.002	0.690**	-0.111	-0.083	0.165	-0.295	0.112	0.355	-0.036	-0.286	-0.132	0.428	1

X1= Relative water content, X2= Canopy air temperature differences, X3= Photosynthetic rate, X4= Stomatal conductance, X5= Transpiration rate, X6= Chlorophyll a, X7= Chlorophyll b, X8= Chlorophyll stability index, X9= Total chlorophyll, X10= Cell membrane stability index, X11= Total soluble protein, X12= Nitrate reductase, X13= Free amino acid, X14= Proline, X15= Glycine betaine, X16= total soluble sugar, X17= super oxide dismutase, X18= Peroxidase, X19= Malondialdehyde, X20= leaves number.

** Signifigicant at 0.01 level.

* Significant at 0.05 level.

4.5.2 Correlation and inter correlation of physiological and biochemical characters on number of leaves after rehydration

Correlation and intercorrelation of physiological and biochemical characters on number of leaves after rehydration in the plantlets of plus trees of teak accessions were estimated and is presented in Table 12.

Among the twenty characters used for estimating the correlation after rehydration in teak plus tree accessions, leaf number showed highly positive correlation with super oxide dismutase (0.658) and positive correlation with chlorophyll a (0.552), chlorophyll b (0.565) and total chlorophyll content (0.565). It showed highly negative correlation with photosynthetic rate (-0.669) and transpiration (-0.754) while negatively correlated with relative water content (-0.540) and stomatal conductance (-0.561).

Relative water content was correlated positively to canopy air temperature difference (0.671) and transpiration rate (0.830). However, it was having negative correlation to rate of transpiration (-0.781).

Canopy air temperature difference was positively correlated with rate of photosynthesis (0.616), stomatal conductance (0.582), rate of transpiration (0.749), and peroxidase activity (0.559), while it was negatively correlated with nitrate reductase activity (-0.664) and proline content (-0.610).

Highly significant correlation was observed between rate of photosynthesis and stomatal conductance (0.725) and rate of transpiration (0.801) while it was correlated positively with total soluble protein (0.656) and superoxide dismutase (0.531). Rate of photosynthesis was negatively correlated to content of chlorophyll b (-0.564) and total chlorophyll content (-0.557).

Stomatal conductance was having positive correlation only with rate of transpiration (0.599). It was having negative correlation with proline content (-0.702) and superoxide dismutase (-0.669).

Negative correlation was observed between transpiration rate and chlorophyll b content (-0.614), total chlorophyll content (-0.579) and superoxide dismutase (-0.671). It was having highly significant positive correlation only with free amino acid content (0.710).

Highly significant positive correlation was observed between chlorophyll a content and chlorophyll b content (0.789), total chlorophyll content (0.854), and glycine betaine 0.698 and it was having positive correlation with nitrate reductase activity (0.628).

Chlorophyll b content was having positive correlation with total chlorophyll content (0.994), while it had a highly significant negative correlation with free amino acid content (-0.762).

Negative correlation was observed between chlorophyll stability index and superoxide dismutase (-0.652) and none of the other characters was correlated with chlorophyll stability index.

Total chlorophyll content was positively correlated with nitrate reductase (0.556) and number of leaves, while it was negatively correlated with free amino acid content (-0.697).

Cell membrane stability had positive correlation only with peroxidase (0.630) none of the other characters was correlated to it.

Negative correlation was observed between total soluble protein and total soluble sugar (-0.526). None of the other characters was correlated with total soluble protein.

Negative correlation was observed between nitrate reductase activity and proline (-0.618) and total soluble sugar (-0.588).

Free amino acid was negatively correlated with proline content (-0.543).

Glycine betaine had highly significant positive correlation with malonaldehyde content (0.698).

Table 12. Correlation between physiological and biochemical characters of plantlets of teak plus tree after rehydration

	X1	X2	X3	X4	X5	X6	X7	X8	X9	X10	X11	X12	X13	X14	X15	X16	X17	X18	X19	X20
X1	1																			
X2	0.671*	1																		
X3	0.500	0.616*	1																	
X4	0.265	0.582*	0.725**	1																
X5	0.830**	0.749**	0.801**	0.599*	1															
X6	-0.451	-0.432	-0.423	-0.392	-0.412	1														
X7	-0.433	-0.353	-0.564*	-0.457	-0.614*	0.789**	1													
X8	-0.178	-0.365	0.093	0.280	0.030	0.263	0.056	1												
X9	-0.450	-0.379	-0.557*	-0.460	-0.579*	0.854**	0.994**	0.056	1											
X10	0.212	0.307	-0.0120	0.081	0.269	0.077	-0.085	-0.101	-0.058	1										
X11	0.395	0.468	0.656*	0.239	0.456	-0.273	-0.320	-0.510	-0.335	-0.273	1									
X12	-0.781**	-0.664*	-0.515	-0.450	0.000	0.628*	0.520	-0.011	0.556*	-0.077	-0.287	1								
X13	0.501	0.363	0.510	0.389	0.710**	-0.278	-0.762**	0.07	-0.697*	0.346	0.224	-0.453	1							
X14	-0.370	-0.610*	-0.351	-0.702**	-0.486	0.324	0.413	-0.128	0.410	-0.508	0.115	-0.618*	-0.543*	1						
X15	-0.172	-0.357	-0.294	-0.146	-0.209	0.698**	0.457	0.153	0.516	-0.137	-0.165	0.501	-0.023	0.189	1					
X16	0.299	0.080	0.289	0.068	0.132	-0.401	-0.263	-0.085	-0.297	-0.307	-0.526*	-0.588*	0.149	-0.068	-0.357	1				
X17	-0.345	-0.273	0.531*	-0.669*	-0.671*	0.040	0.324	-0.652*	0.282	-0.149	0.100	0.260	-0.458	0.387	-0.105	0.252	1			
X18	0.227	0.559*	0.054	0.082	0.318	0.073	0.084	-0.251	0.085	0.630*	0.003	-0.237	0.100	-0.337	-0.372	-0.164	-0.089	1		
X19	0.117	0.184	0.346	0.495	0.313	0.299	0.048	0.137	0.096	-0.139	0.383	-0.005	0.303	-0.079	0.698**	0.111	-0.411	-0.079	1	
X20	-0.540*	-0.483	-0.669**	-0.561*	-0.754**	0.552*	0.565*	-0.159	0.565*	0.149	-0.448	0.501	-0.325	0.112	0.405	-0.187	0.658*	-0.146	-0.181	1

X1= Relative water content, X2= Canopy air temperature differences, X3= Photosynthetic rate, X4= Stomatal conductance, X5= Transpiration rate, X6= Chlorophyll a, X7= Chlorophyll b, X8= Chlorophyll stability index, X9= Total chlorophyll, X10= Cell membrane stability index, X11= Total soluble protein, X12= Nitrate reductase, X13= Free amino acid, X14= Proline, X15= Glycine betaine, X16= total soluble sugar, X17= super oxide dismutase, X18= Peroxidase, X19= Malondialdehyde, X20= leaves number.

** Significant at the 0.01 level.

* Significant at 0.05 level.

4.6 CLUSTER ANALYSIS

4.6.1 Clustering of accessions based on physiological and biochemical characters under drought condition

Hierarchical cluster analysis was done for the eleven accessions based on the Euclidian squared distance. The data on relative water content, canopy air temperature, photosynthetic rate, transpiration rate, stomatal conductance, chlorophyll a, chllorophyll b, chlorophyll stability index, total chlorophyll content, cell membrane stability index, total soluble protein, nitrate reductase activity, free amino acid, proline, glycine betaine, total soluble sugar, superoxide dismutase, peroxidase and malonidialdehyde of the teak accessions exposed to drought was used for grouping the accessions.

The accessions were grouped into five clusters (Figure 8) and the details of the five clusters are given in the Table (13). The cluster III possesses four of accessions whereas the least number observed for the cluster V.

Table 14 shows inter and intra cluster distances. Intra cluster distances gives the average distance between the elements within a cluster whereas the distance between two clusters gives the inter cluster distances. The diagonal elements shows the intra cluster distances and the off diagonal elements shows the inter cluster distances. It is observed from the table that highest intra cluster distance shown by the cluster II and the highest inter cluster distance was shown by cluster IV and cluster V.

Table13. Clustering of teak plus tree accessions based on physiological biochemical characters under drought stress condition.

Clusters				
I	II	III	IV	V
KFRI T9 KFRI T144	KFRI T1 KFRI T16	KFRI T4 KFRI T55 KFRI T116 KFRI T142	KFRI T5 KFRI T24	KFRI T44

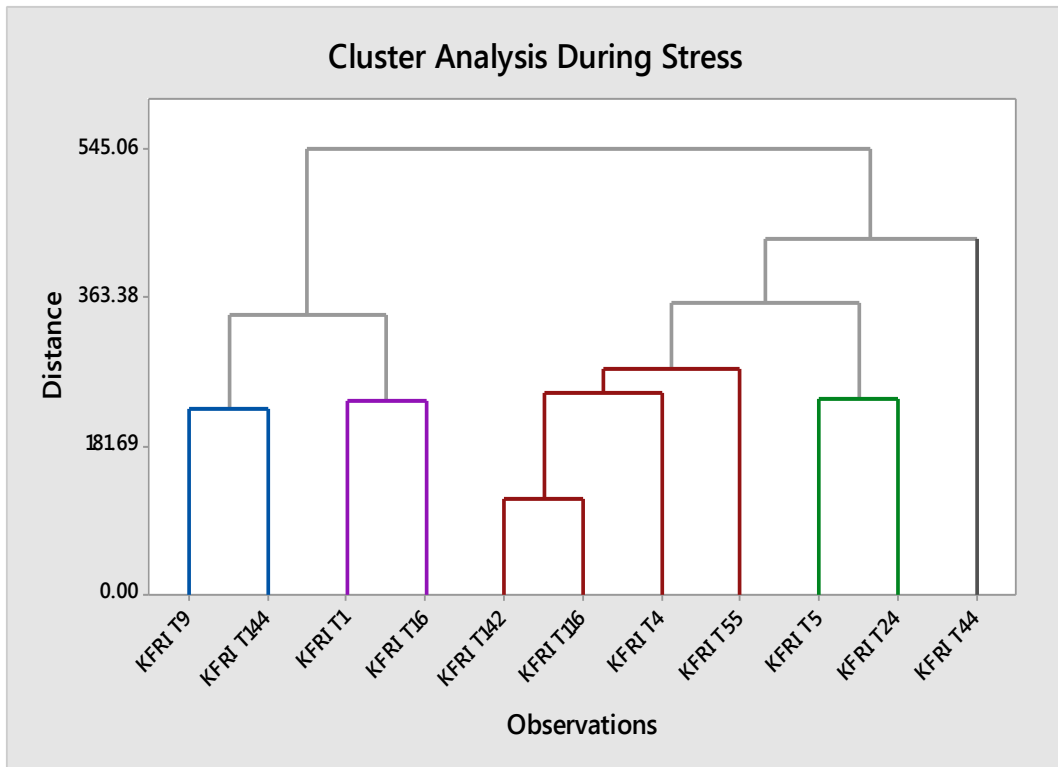


Figure 8. Dendrogram based on on physiological and biochemical attributes in *Tectona grandis* plus tree accessions under moisture stress condition

Table14. Inter and intra cluster distances of the teak accessions under drought stress condition

Matrix showing inter and intra cluster distances					
	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5
Cluster 1	07.5				
Cluster 2	17.9	09.4			
Cluster 3	26.5	14.1	07.7		
Cluster 4	15.0	15.9	20.6	07.7	
Cluster 5	28.0	17.7	21.0	23.1	00.0

4.6.2 Clustering of accessions based on physiological and biochemical characters after rehydration

The teak plus tree accessions were grouped based on physiological and biochemical characters after rehydration and they were grouped into seven clusters. (Table 15, 16 and figure 9).

The cluster VII had three accessions while only one accession was present in cluster I, IV, V and VI.

The diagonal elements of table 15 shows the intra cluster distances and inter cluster distances among the accessions. It is observed from the table that highest intra cluster distance shown by the cluster I and the highest inter cluster distance shown by cluster VI and VII (17).

Table15. Clustering of teak accessions based on physiological and biochemical characters after rehydration

Clusters						
I	II	III	IV	V	VI	VII
KFRI T4 KFRI T9 KFRI T44	KFRI T5 KFRI T24	KFRI T16 KFRI T116	KFRI T142	KFRI T144	KFRI T1	KFRI T55

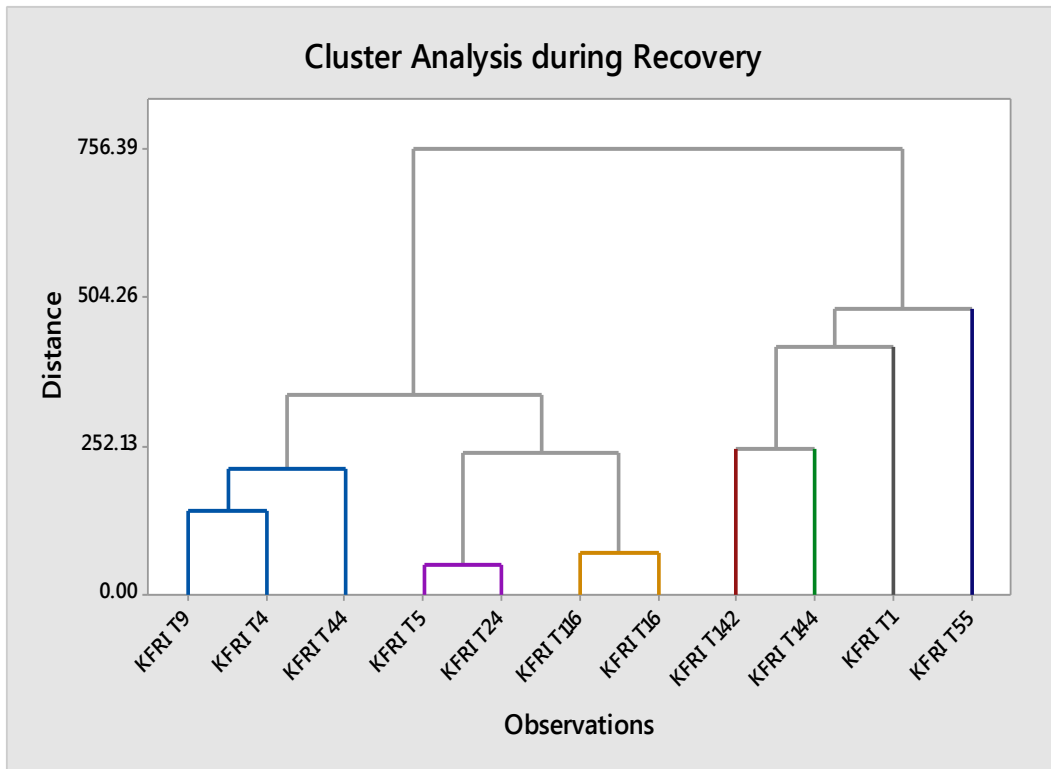


Figure 9. Dendrogram of teak plus tree accessions based on physiological and biochemical characters after rehydration.

Table 16. Inter and intra cluster distances of the cluster diagram based on physiological and biochemical characters of teak accessions after rehydration

Matrix showing inter and intra cluster distances							
	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5	Cluster 6	Cluster 7
Cluster 1	07.9						
Cluster 2	15.0	00.0					
Cluster 3	26.9	15.7	00.0				
Cluster 4	16.8	21.1	29.7	03.5			
Cluster 5	17.1	19.7	21.3	23.0	00.0		
Cluster 6	15.6	22.2	34.8	14.5	25.1	04.3	
Cluster 7	28.4	23.6	16.9	35.3	24.6	39.8	00.0

4.7 PRINCIPAL COMPONENT ANALYSIS

PCA was conducted to study the variations and bring out strong patterns in the data set of independent variables. Further away these vectors are from a PC origin, the more influence they have on that PC. Loading plots also hint at how variables correlate with one another: a small angle implies positive correlation, a large one suggests negative correlation, and 90° angle indicates no correlation between two characteristics.

4.7.1 Principal component analysis at drought condition.

Principal component analysis was carried out using data taken under drought condition. PC1 accounted for 23.0 per cent of the total variability, which was mainly contributed positively by photosynthetic rate, stomatal conductance, transpiration rate, chlorophyll stability index, total chlorophyll content and glycine betaine. PC2 accounted for 18.8 per cent of the total variability (Table 17). Thus first two components together accounted for 41.9 per cent of the total variability, which was mainly contributed positively by relative water content, nitrate reductase, free amino acid, proline and malondialdehyde.

The component loading biplot is given in Fig. 10. It shows that relative water content, nitrate reductase, free amino acid, proline and malonidialdehyde are closely related. These parameters in addition to total chlorophyll content showed negative association with cell membrane stability index, total soluble protein, super oxide dismutase and peroxidase. Canopy air temperature and total soluble sugar showed negative association with proline. Photosynthetic rate, stomatal conductance, transpiration rate, cell membrane stability index, glycine betaine were closely correlated but did not show any association with other characters except total chlorophyll content.

Table 17. Principal component analysis of physiological and biochemical characters of teak plus tree accessions at stress condition

Variable	PC1	PC2
Relative water content (%)	0.132	0.110
Canopy air temperature differences ($^{\circ}\text{C}$)	-0.045	-0.111
Photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)	0.239	-0.306
Transpiration rate ($\text{mol m}^{-2} \text{ s}^{-1}$)	0.332	-0.339
Stomatal conductance ($\text{mol m}^{-2} \text{ s}^{-1}$)	0.394	-0.296
Chlorophyll stability index (mg g^{-1})	0.126	-0.449
Total chlorophyll content (mg g^{-1})	0.223	-0.030
Cell membrane stability index	-0.231	-0.044
Total soluble protein (mg g^{-1})	-0.123	-0.069
Nitrate reductase activity ($\text{mmol nitrate g}^{-1}\text{hr}^{-1}$)	0.269	0.286
Free amino acid (mg g^{-1})	0.320	0.322
Proline ($\mu\text{g g}^{-1}$)	0.048	0.164
Glycine betaine (mg g^{-1})	0.198	-0.241
Total soluble sugar (mg g^{-1})	-0.165	-0.372
Super oxide dismutase ($\text{units protein}^{-1} \text{ g}^{-1}$)	-0.295	-0.078
Peroxidase ($\text{min}^{-1} \text{ mg}^{-1} \text{ protein}$)	-0.322	-0.150
Malondialdehyde (n mol g^{-1})	0.298	0.183
Eigen	3.912	3.204
Proportion	0.230	0.188
Cumulative	0.230	0.419

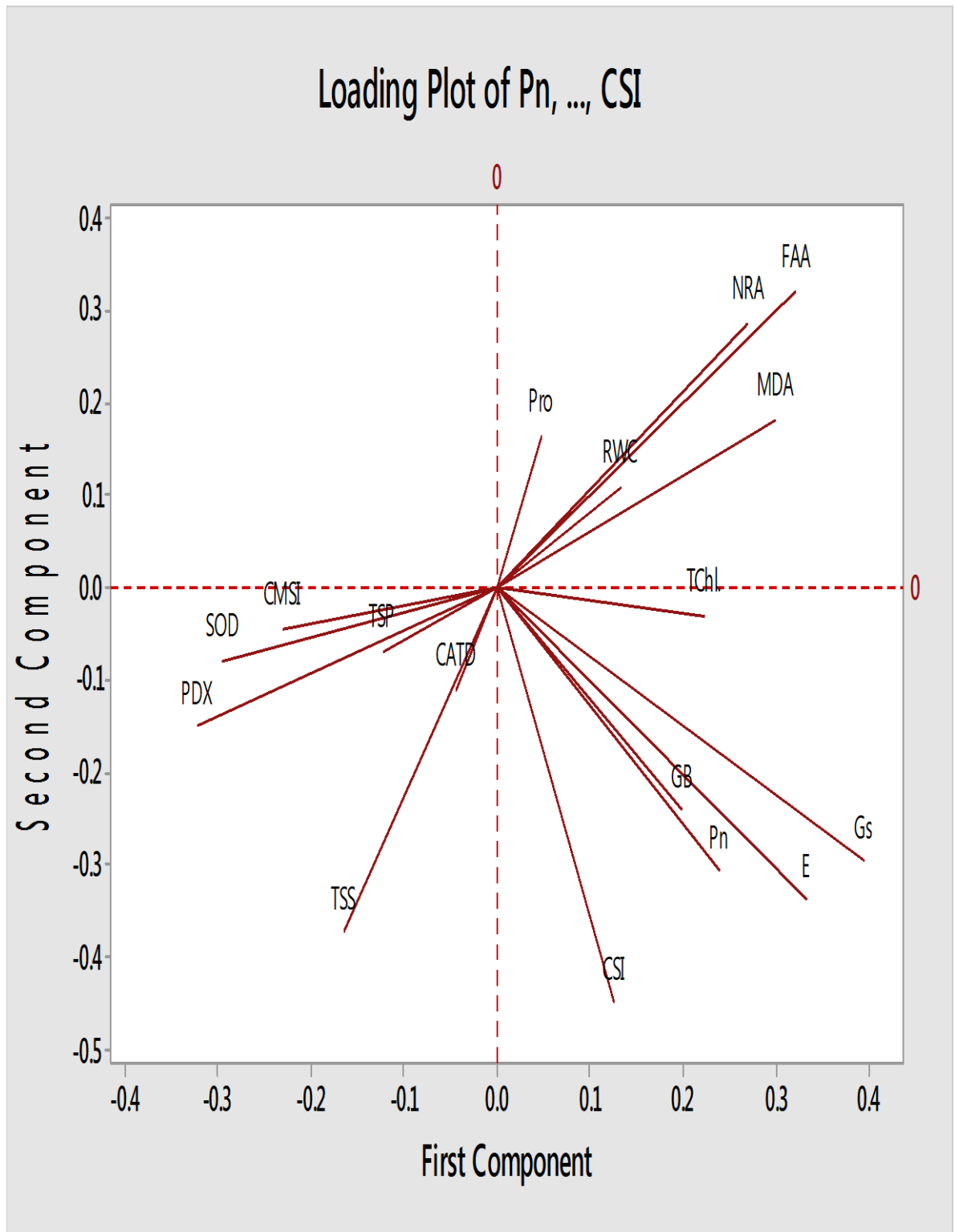


Figure10. Loading plot of physiological and biochemical characters of *Tectona grandis* plus tree accessions at drought condition

4.7.2 Principal component analysis of physiological and biochemical characters of teak plus tree accessions after rehydration

Principal component analysis was carried out using data on physiological and biochemical characters of teak plus tree accessions after rehydration. PC1 accounted for 37.9 per cent of the total variability, which was mainly contributed positively by relative water content, canopy air temperature, cell membrane stability index, total soluble protein, total soluble sugar and peroxidase. PC2 accounted for 15.3 per cent of the total variability, which was mainly contributed positively by chlorophyll content, nitrate reductase activity and free amino acid (Table 18). First two components which together accounted for 53.2 per cent of the total variability, which was mainly contributed positively by photosynthetic rate, stomatal conductance, transpiration rate, chlorophyll stability index, glycine betaine and malondiadehyde

The component loading biplot given in Fig. 11. It shows that relative water content, canopy air temperature, cell membrane stability index and total soluble protein are closely related. These parameters showed negative association with total chlorophyll content and nitrate reductase. Total soluble sugar and peroxidase were negatively related with free amino acid. Both set showed no association with malondialdehyde and super oxide dismutase which were negatively related. Photosynthetic rate, stomatal conductance, transpiration and glycine betaine were negatively associated with proline.

Table 18. Principal component analysis of physiological and biochemical characters of teak plus tree accessions after rehydration

Variable	PC1	PC2
Relative water content (%)	0.320	-0.155
Canopy air temperature differences ($^{\circ}\text{C}$)	0.292	-0.116
Photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)	0.320	0.097
Transpiration rate ($\text{mol m}^{-2} \text{ s}^{-1}$)	0.296	0.244
Stomatal conductance ($\text{mol m}^{-2} \text{ s}^{-1}$)	0.362	0.067
Chlorophyll stability index (mg g^{-1})	0.003	0.425
Total chlorophyll content (mg g^{-1})	-0.271	0.119
Cell membrane stability index	0.106	-0.079
Total soluble protein (mg g^{-1})	0.191	-0.091
Nitrate reductase activity ($\text{mmol nitrate g}^{-1}\text{hr}^{-1}$)	-0.320	0.194
Free amino acid (mg g^{-1})	-0.138	0.427
Proline ($\mu\text{g g}^{-1}$)	-0.283	-0.038
Glycine betaine (mg g^{-1})	0.283	0.103
Total soluble sugar (mg g^{-1})	0.125	-0.233
Super oxide dismutase ($\text{units protein}^{-1}\text{g}^{-1}$)	-0.215	-0.436
Peroxidase ($\text{min}^{-1} \text{mg}^{-1} \text{protein}$)	0.109	-0.196
Malondialdehyde (n mol g^{-1})	0.124	0.405
Eigen	6.435	2.602
Proportion	0.379	0.153
Cumulative	0.379	0.532

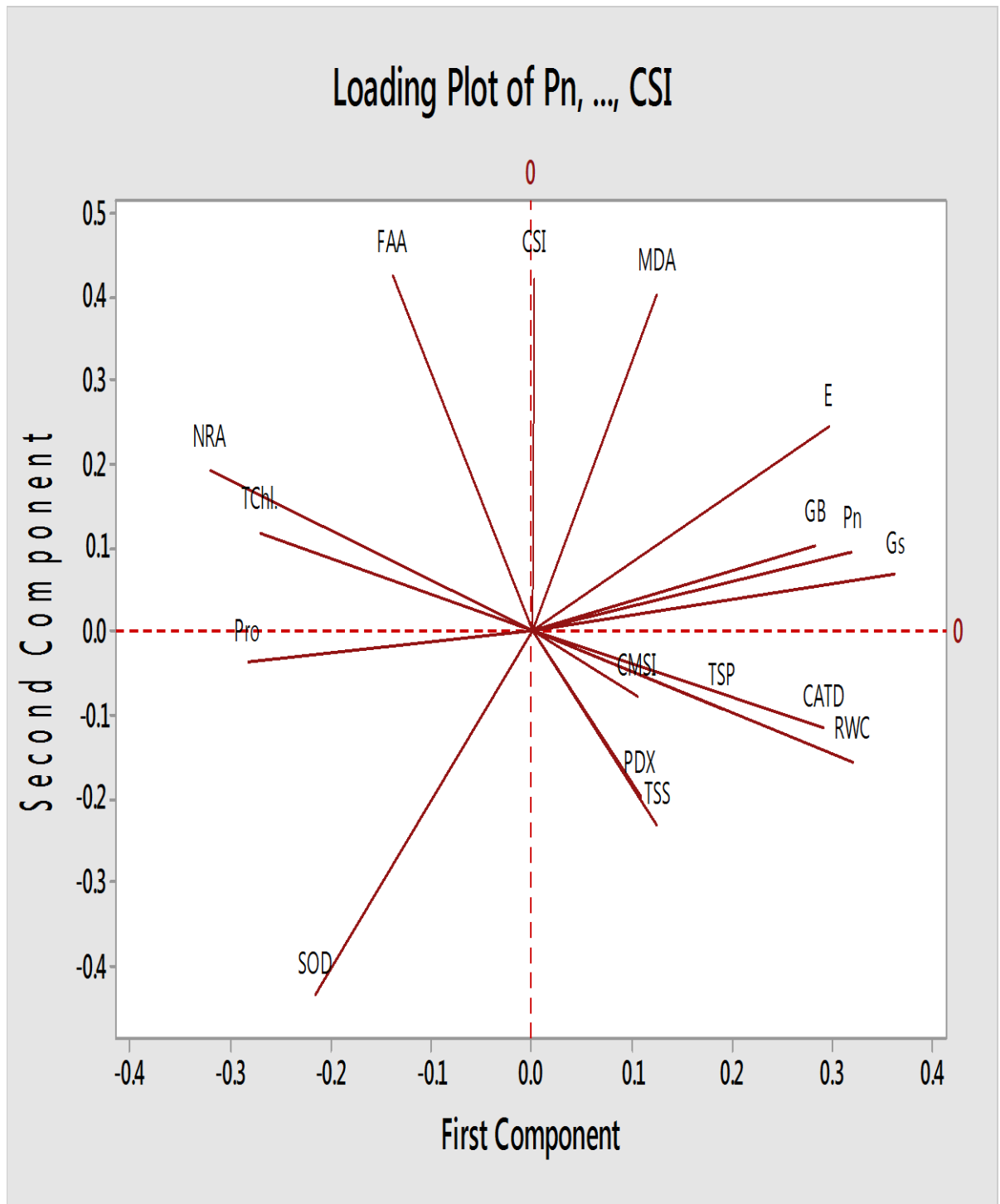


Figure 11. Loading plot of physiological and biochemical characters of *Tectona grandis* plus tree accessions after rehydration.

4.8 SELECTION INDEX

Selection index values were worked out to select accession tolerant to drought stress and recovery (based on biochemical parameters and chlorophyll content), using principle component analysis. Principal component analysis was performed on all parameters where accumulation indicates the tolerance of the plant and the first principal component was taken as the index value for selection (Table 17 and 18) to form the following equations:

$$\text{Tolerance to stress} = 0.233 \times \text{T Chl.} - 0.231 \times \text{TSP} + 0.269 \times \text{NRA} + 0.320 \times \text{FAA} + 0.480 \times \text{Pro} + 0.198 \times \text{GB} - 0.165 \times \text{TSS} - 0.295 \times \text{SOD} - 0.322 \times \text{PDX} + 0.298 \times \text{MDA}$$

$$\text{Recovery from stress} = \text{T Chl.} + 0.106 \times \text{CMSI} + 0.191 \times \text{TSP} - 0.320 \times \text{NRA} - 0.138 \times \text{FAA} - 0.283 \times \text{Pro} + 0.283 \times \text{GB} + 0.125 \times \text{TSS} - 0.215 \times \text{SOD} + 0.425 \times \text{PDX} + 0.109 \times \text{MDA}$$

Based on these equations selection index were by calculate the score representing total response to drought and recovery in each accession. Under drought condition, KFRI T55 showed the strong response, while KFRI T24 showed the weakest response. Under recovery, KFRI T116 showed the strongest response while, KFRI T55 showed the weakest response.

Table 19. Total score of biochemical responses of teak accessions during stress and recovery.

Accession	Stress	Recovery
KFRI T1	5.58	-0.69
KFRI T4	5.24	-2.13
KFRI T5	0.93	2.80
KFRI T9	9.00	1.56
KFRI T16	2.00	5.62
KFRI T24	-0.02	3.18
KFRI T44	8.96	-3.26
KFRI T55	9.71	-5.28
KFRI T116	6.21	4.71
KFRI T142	6.80	1.62
KFRI T144	7.63	-3.25



DISCUSSION



DISCUSSION

5.1. VARIABILITY STUDY

Every tree improvement programs begins with the assessment of the natural variability existing in the species. Forest trees are assumed to be genetically variable in order to survive, grow and reproduce under numerous environments (Antonovicks, 1971). Evaluation of different genotypes of tree species for morphological, physiological and biochemical characteristics is thus an important aspect of a tree improvement programme. Variation in response to environmental stress and its recovery indicate that there are considerable opportunities for selection to improve productivity in sites with possible threat of these stresses. One of the possibilities to determine such variation is to determine the extent, cause and nature of variations present in seedling stage assuming good juvenile-mature correlation in the species. Although *Tectona grandis* has a significant importance in tropical forest, there are few reports on exploration, identification and documentation on the variation in teak to stresses like drought.

5.2.1 Evaluation of morphometric traits of plus tree accessions at optimal growth stage

In the first experiment, an attempt was made to identify the best performing plus tree accessions of one-year age that grow in optimal growing conditions in nursery. The variables such as seedling height, collar diameter, leaves number and branches number were measured at three-month interval (first, third and six) to study the performance.

The seedling height was found to be significantly different among the accessions at first month, while at third and sixth months they were on par (Table 1). Average height at first, third and sixth month were 26.5 cm, 44.5 cm and 67.6 cm respectively. After sixth month KFRI T116 showed the highest value 74.60 cm.

The accessions did not differ in terms of collar diameter at all stages (Table 1). Average collar diameter at first, third and sixth month was 7.4 mm,

10.6 mm, 15.7 mm respectively. After sixth month KFRI T1 showed the highest value 17.0 mm.

Number of branches and number of leaves too did not differ significantly among the various accessions (Table 1). Average of number of branches at first, third and sixth month was 0.5, 1.3 and 2.0 respectively. Average of number leaves at first, third and sixth month was 10.4, 17.5 and 22.0 respectively.

Results obtained for the qualitative characters between accessions indicate that there was no difference in terms of morphometric characters among the plus tree accession up to six-month stage in nursery. Early differences in morphometric characters in tree seedlings are not entirely unknown. However, at early lag phase of growth it is entirely plausible that the differences are not obvious. It could be reasonable to expect that the differences will start emerging later on once the seedlings enter into the log phase of their growth.

5.2.2 Evaluation of morphometric traits of plus tree accessions under induced water stress

The availability of water is one of the major limiting factors for normal plant growth. Plants growing in tropical climate often face certain degree of drought stress. Plant growth is influenced by various internal and external factors besides its genetic makeup and is an important tool for assessing productivity.

The accessions subjected to stress did not show any differences among them and has mean height of 72.6 cm. Similar results were noticed in collar diameter too. Mean collar diameter of the plantlets after induction of stress was 16.55 mm. (Table 2). However, the stress seems to have reduced the growth rates. The reason of growth inhibition under water deficit situation is quite common since cell division dependent on cell turgidity and hence growth. Many studies showed reduction in terms of growth under water stress in other tree seedling like *Erythrina* (Muthuchelian *et al.*, 1986), *Albizia* (Sundaravalli *et al.*, 2005), *Eucalyptus microtheca* (Li *et al.*, 2000), *Populus* species (Yin *et al.*, 2005b),

Populus cathayana (Xiao *et al.*, 2009) and *Populus davidiana* (Zhang *et al.*, 2005).

Branching habit of trees effect the quality of final product (Zobel and Talbert, 1984). Undesirable branching decreases the volume and economic value of the timber (Codesido and Fernandez-lobez, 2008). Significant variation in number of branches was detected among the accessions under stress (Table 2). Number of branches under moisture stress in teak reduced to 1.7 from 2.0 when drought stress was induced. Significant reduction in leaves number occurred when plants subjected to water stress (17.1) from 20.0. Significant variation in number of leaves was observed amongst the accession under stress. Branches and leaves reduction are common responses to drought stress in tropical forests (Corlett, 2016). The reason for branches and leaves senescence would provide a more extreme mechanism of water conservation by reducing the transpiration surface and reduce the transpiration loss (Rood *et al.*, 2000). In extreme cases, in deciduous species like teak, the tree sheds its entire leaves and remains dormant to tide over the stress period.

5.3 EFFECT OF DROUGHT STRESS ON PLUS TREE ACCESSIONS

5.3.1 Relative water content and canopy air temperature difference

Plant water status controls the physiological functioning and growth (Kramer, 1969). Relative water content (RWC), a key indicator of turgidity, provides a measure of the plant water status. It is usually used as an index of the degree of cell and tissue dehydration in plants (Anjum *et al.*, 2011). Our study revealed that RWC decreased in all teak accessions during drought stress (Table 9). Drought stress-induced decrease in RWC has been reported in many previous studies (Yang and Miao, 2010; Binks, 2016; Karimi *et al.*, 2018; El-yamani *et al.*, 2019). It was found that in *Hevea brasiliensis* seedlings, there was a 20% decrease in RWC after 9 days without irrigation, when compared to control treatment (Chen *et al.*, 2010). After resuming irrigation, recovery was showed by all

accession except KFRI T55 (Fig. 1) indicating more plastic damages to the accession from the stress.

Plant temperature has been recognized as one of the most important factor to obtain a good measure of plant water stress levels (Stockle and Dugas, 1992). Under conditions of water stress, plants often attain higher temperature, which increases their vulnerability to light stress and photo inhibition (Carpentier, 1997). Canopy air temperature difference is a convenient measure of plants avoiding heat stress through transpirational cooling. Consequently, when water becomes limiting, this pathway becomes unavailable and plant canopy temperature rises. Thus CATD is often used as a handy indication of the stress levels of the plant. Present study revealed that CATD in leaves increased significantly due to moisture stress (Table 9). This result is similar to other studies that revealed that drought stressed plants had higher canopy temperature than non-stressed plants (Siddique *et al.*, 2000). It was found that in *Tectona grandis* seedlings, there was an increase in CATD under water deficit compared to well water seedlings (Sneha *et al.*, 2017). In recovery stage, partial recovery was seen KFRI T9, T16, T24, T44 and T55 (Fig. 9), While the remaining accessions recovered completely. The ability to recover fully indicates the flexibility in these accessions to moisture stress. The accessions that did not decrease the leaf temperature transpired less than the others. Similar behaviour observed in six scots pine provenances, where in Seidel *et al.* (2016) observed an increase in CATD during water stress and after re-irrigation the differences became less or near to normal.

5.3.2 Photosynthetic activity

Inducing drought significantly reduced photosynthetic rate (Pn), stomatal conductance (Gs) and transpiration rate (E). Decrease of 46.4 per cent in photosynthesis, 89.5 per cent in stomatal conductance and 86.4 per cent in transpiration rate was observed in the study. (Table 4 and 9). The photosynthetic activity reduction during drought initially is probably prompted by stomatal closure, leading to limitation of ambient CO₂ diffusion to the mesophyll. Stomatal

closure also limits transpiration. In eucalyptus species under drought stress, a strong reduction in the rate of photosynthesis, stomatal conductance and transpiration was observed (Lima *et al.*, 2003). Drought stress decreasing photosynthesis stomatal conductance, and transpiration rates was noticed in young apple trees too (Wang *et al.*, 2018).

Moreover, among the accessions Gs exhibited higher variation response even before the leaf water content changed due drought treatment (Table 3). Stomatal conductance can have protective effects because it allows the plant to save water and to improve its efficient use (Chaves *et al.*, 2009). This indicates that Gs can be used as good indicator of drought stress tolerance in plants.

Photosynthesis reduction is mainly caused by stomatal limitation under mild to moderate drought condition when both Gs and sub-stomatal CO₂ concentration (Ci) decline, while non-stomatal limitation is the main reason for the decrease in photosynthesis when Ci increases and Gs reaches a minimum inflection point (Pérez-Lopez *et al.*, 2012; Zhou *et al.*, 2013). This agree with our findings that the decrease in photosynthesis due to stress might be as result of both stomatal and non-stomatal limitation mechanisms as reported by Epron *et al.* (1992) and Maxwell and Johnson (2000) too. Higher values of Pn are usually accompanied by higher rates of E and Gs for several species in the tropical region (Santiago and Wright, 2007). However, the maintenance of the Pn associated with lower values of Gs and E under conditions of water stress, indicate the existence of adaptation mechanisms. According to Chaves and Oliveira (2004), the initial phase of water deficit is the stomata partial closure, observed through a fast reduction of the Gs. Then, a later phase can occur with a decrease in the transpiration rate and photosynthetic carbon assimilation, which both causes an elevation in the water use efficiency and intrinsic water use efficiency, in order to diminish water loss.

The recovery from drought stress is dependent on not only the duration time of rehydration but also the degree and duration time of drought (Gomes *et*

al., 2012; Sircelj *et al.*, 2007). After rehydration, the results showed that the impaired photosynthetic activity (Pn) gradually recovered along with stomatal conductance and transpiration in most accessions (Fig. 2). The lack of recovery may be due to limitation of stomatal and mesophyll conductance and biochemical limitations (Galmes *et al.*, 2007). The complete recovery in photosynthetic rate in KFRI T4 and T116 indicated that increases in stomatal aperture with duration of re-watering facilitated diffusion of CO₂ from the atmosphere to the carboxylation site of Rubisco (Abid *et al.*, 2018). Moreover, the recovery of photochemistry and down regulation of heat dissipation in all accessions occurred in the short period. These observations on photosynthetic rate, stomatal conductance and transpiration rate results are in accordance with those previously observed in teak (Rajendrudu and Naidu, 1997; Husen, 2010), where a significant drop in photosynthetic rate, stomatal conductance and transpiration rate in trees submitted to water stress was observed. These values recovered near to the values of optimal irrigation. Hence it can be concluded that the role played by stomatal closure and physiological processes were clear in decreasing net photosynthetic rates during drought stress.

5.3.3 Photosynthetic pigment and chlorophyll stability index

Photosynthetic pigments are important to plants mainly for harvesting light and production of reducing powers. Both the chlorophyll a (Chl. a) and chlorophyll b (Chl. b) are vulnerable to water deficit (Farooq *et al.*, 2009). Chlorophyll Stability Index (CSI) is a function of temperature and is found to co-relate with drought tolerance. It is a measure of integrity of membrane or heat stability of pigments under drought stress conditions (Kaloyereas, 1958). It is an important parameter used to measure drought tolerance of a plant. A higher CSI helps plants to withstand stress through better availability of chlorophyll. This leads to increased photosynthetic rate (Mohan *et al.*, 2000).

The result showed that Chl. a, Chl. b, chlorophyll stability index and total chlorophyll content in teak accessions were reduced under water stress (Table 9). The decrease in chlorophyll content due to pigment photo-oxidation and

chlorophyll degradation under drought stress has been considered as a typical symptom of oxidative stress. It is well known that exposing plants to water stress lead to photosynthetic pigments degradation (Duan *et al.*, 2005; Manivannan *et al.*, 2007; Elsheery and Cao, 2008; Guerfel *et al.*, 2009). Both drought stress and temperature stress causes significant decrease of CSI in all wheat genotypes (Sairam *et al.*, 1997). Loss of chlorophyll contents under water stress is considered as a main cause of inactivation of photosynthetic pigments due to several causes. Smirnoff (1993) reported that the decrease of chlorophyll content was mainly the result of damage to chloroplasts caused by reactive oxygen species. The reduction of chlorophyll content under drought might be due to the fact that drought stress blemishes the chlorophyll content through internal modification in the thylakoid membrane (Sivakumar *et al.*, 2017). The reduction of photosynthetic pigments may be due to reduction in the lamellar content of the light harvesting Chl. a/b protein that accounts for the elevated Chl. a/b ratio and/or due to water stress-induced decrease in the availability of mineral nutrients, and/or to the damages to chloroplasts by reactive oxygen species (ROS), and or either slow synthesis or fast breakdown of chlorophyll pigments (Khanna-Chopra *et al.*, 1980; Ashraf, 2003). The decrease in photosynthetic pigment concentrations could also reduce damages to the photosynthetic machinery, since under low water availability there would be lower CO₂ concentrations inside the chloroplast caused by stomatal closure (Hortensteiner, 2009; Oliveira *et al.*, 2014). Drought stress caused a large decline in the chlorophyll a content, the chlorophyll b content, and the total chlorophyll content in all sunflower varieties investigated (Manivannan *et al.*, 2007). Juby (2019) reported that drought stress decreased chlorophyll stability index in all cocoa hybrids.

At recovery stage, all accessions recovered near to normal stage (Fig. 3). Majority of accessions showed complete recovery in chlorophyll a and total chlorophyll while in chlorophyll b and chlorophyll stability index, the recovery was slower. The faster recovery of Chl. a indicate that less effect of drought response compare to Chl. b. Observations in *Anadenanthera columbrina*

(Rivas *et al.*, 2013), *Moringa oleifera* (Oliveira *et al.*, 2014) and *Sterculia foetida* (Frosi *et al.*, 2017) indicate that higher concentration of chlorophylls and carotenoids under stress, could provide a faster recovery of the photosynthetic activity after rehydration.

5.3.4 Total soluble protein

A decrease in the protein concentration would be a typical symptom of oxidative stress and has frequently been observed in drought stressed plants (Seel *et al.*, 1992; Moran *et al.*, 1994). Many studies have revealed that excess light energy absorbed by photosynthetic pigments is responsible for PSII photo-damage (Gan and Amasino, 1997; Quirino *et al.*, 2000; Barber and Andersson, 1992) and to damage of thylakoid proteins (Kato and Sakamoto, 2009; Aro *et al.*, 1993). Present study indicates that water deficit affected all accessions significantly and caused total soluble protein (TSP) degradation (Table 9). The degradation might be due to increase protease activity (Palma *et al.*, 2002) or other catabolic enzymes, which get activated under drought, stress (Zhang and Davies, 1987). It could also be due to fragmentation of proteins due to toxic effects of reactive oxygen species (Baisak *et al.*, 1994; Kramer and Boyer, 1995). Under normal conditions, D1 protein remains at a certain level by the balance between the damage and repair of D1 (Baena-Gonzalez and Aro, 2002). Previous research has shown that, to prevent the accumulation of photodamaged D1 and PSII, plants develop a repair process consisting of several steps as follows: proteolytic degradation of the D1 protein; synthesis of the precursor to the D1 protein (pre-D1); insertion of the newly synthesized precursor into the thylakoid membrane concomitant with the assembly of other PSII proteins; maturation of the D1 protein by C-terminal processing of pre-D1; and finally, assembly of the oxygen-evolving machinery (Aro *et al.*, 1993; 2004).

After recovery five accession showed complete reversal while other showed partial recovery (Fig. 4). Similar results obtained in teak (Husen, 2010),

cowpea (Rivas *et al.*, 2016), wheat (Abid *et al.*, 2018) and apple (Wang *et al.*, 2018).

5.3.5 Nitrate reductase activity

The nitrate reductase activity (NRA), a substrate inducible enzyme mediates conversion of nitrate to nitrite (Beevers and Hageman, 1969). In our study inducing drought stress in all accessions showed significant reduction in NRA (Table 9). The reduction in the activity might be either due to reduction in enzyme level (Bardzik *et al.*, 1971) or due to the inactivation of the enzyme (Nicholas *et al.*, 1976) caused by stress condition. Ferrario-Mery *et al.* (1998) reported decline of NRA during water stress is mainly attributed to low NO³⁻ absorption and availability resulting from water uptake deprivation. Decline of NRA consequent to moisture stress has been reported in many plant species, such as maize (Foyer *et al.*, 1998), potato (Ghosh *et al.*, 2000), winter wheat (Xu and Yu, 2006) and cocoa (Juby, 2019). Sivaramakrishnan *et al.* (1988) studied the midseason drought indicating that there is a sharp decline in NRA under water stress situation.

The reduction of NRA in all accessions was reversed partially after relief of stress except KFRI T1 (Fig. 4). The reversal may be attributed to a restoration of the polyribosomal system (Hsiao, 1970). This confirm to earlier reports on wheat (Plaut, 1974), *Zea mays* L, (Shaner and Boyer, 1976), sugarcane (Devi *et al.*, 2018), where under drought stage NRA decline severely while after resuming irrigation NRA recover to near to normal.

5.3.6 Osmotic adjustment

The physiological mechanisms that allow a species to tolerate prolonged periods of drought stress can involve numerous attributes including the accumulation of osmotically active solutes like free amino acid (FAA), proline (Pro), glycine betaine (GB) and total soluble sugar (TSS), so that turgor in cell may be maintained during drought (Farooq *et al.*, 2009; Silva *et al.*, 2009). In this

study, FAA content increased under drought treatment, as compared with normal watering stage (Table 9). FAA is an important mechanism that alleviates some of the detrimental effects of water-deficit stress (Morgan, 1984). Accumulation of amino acids may be due to the hydrolysis of protein and may occur in response to the change in osmotic adjustment of cellular contents (Greenway and Munns, 1980). This conforms to earlier reports of (Yadav *et al.*, 2005; Manivannan *et al.*, 2007).

Our study found that proline increased due to water deficit (Table 9). Proline is an amino acid and known to act as an osmolyte, protecting the plasma membrane integrity (Mansour, 1998), as a sink of energy or reducing power (Verbruggen *et al.*, 1996), as a scavenger of reactive oxygen species and their derivatives (Hong *et al.*, 2000; Bashir, *et al.*, 2007) and as a source for carbon and nitrogen (Peng *et al.*, 1996), thereby helping the plants to tolerate stress effects (Manivannan *et al.*, 2007). Thus, the proline accumulation appeared to play adaptive roles in plant stress tolerance (Manivannan *et al.*, 2007) and is known to be the first response of plants exposed to water-deficit stress (Anjum *et al.*, 2011).

Sugars have different functions in plants from energy storage to signaling. Plants utilize several sugar-based strategies to adapt to environmental stresses (Anderson and Kohron, 2001; Chaves *et al.*, 2003). Present study indicated that TSS increased significantly in all accession when subjected to water stress stage (Table 9). The increase of TSS contents through inversion of carbohydrates may contribute to enhanced desiccation tolerance and allows metabolic activity to be maintained. Chaves (1991) mentioned that TSS content tends to be maintained in the leaves of drought-stressed plants, although rates of carbon assimilation were partially reduced. The maintenance of TSS content in the absence of declined carbon assimilation may be at the expense of starch, which drastically declines. Hence, it can be concluded that the TSS act as osmoticum that protect specific macromolecules and contribute to the stabilization of membrane structures (Bartels and Sunkar, 2005).

The glycine betaine (GB) content increased under drought stress in all teak accessions (Table 9). The accumulation of GB might serve as an intercellular osmoticum and it could be closely correlated with elevation of osmotic pressure (Wyn-Jones and Story, 1978). GB may maintain the osmoticum, provided that the basal metabolism of the plant can sustain a high rate of synthesis of these compounds to facilitate osmotic adjustment for tolerance to water stress (Kishore *et al.*, 1995). Xing and Rajashekar (1999) reported that when *Phaseolus vulgaris* was treated with GB, plant showed a slower decrease in leaf water potential during stress and developed wilting symptoms much later than untreated plants.

The accumulation of osmoprotectants in plants under drought stress conditions have been reported to increase the osmotic adjustment capability, resulting in a stronger drought tolerance (Zu *et al.*, 2017). Previous studies of teak drought stress showed a significant increase in leaf free amino acid, proline, and total soluble sugar contents after drought stress (Castro *et al.*, 2007; Husen, 2010). Similarly GB increased in cocoa hybrids due to exposure to stress (Juby, 2019). It has also been demonstrated that GB treated bean plants showed a better ability to recover from wilting following the removal of the stress (Ashraf and Foolad, 2007).

After re-watering, all accessions did not show complete recovery in proline and glycine betaine, while complete recovery was noticed for free amino acid and total soluble sugar (Fig. 5). Consistent with the results in this study Husen (2010), Abid *et al.* (2018), Zhang *et al.* (2018) and Dien *et al.* (2019) found that rehydration after water stress significantly reduced the content of osmoregulants that were accumulated during stress.

5.3.7 Antioxidant enzymes

When the degree and duration of water stress exceed the tolerance ability of plants, plants will not be able to dissipate the excess radiation intercepted and this excess energy will lead to an increase in the production of reactive oxygen

species (ROS) including $O_2^{\bullet-}$ and H_2O_2 (Wang *et al.*, 2018). Consequently, the accumulation of ROS will lead to damages to plant proteins, lipids, carbohydrates, DNA (Gill and Tuteja, 2010). To avoid this disturbance a series of antioxidant enzymes are deployed by plants to scavenge ROS induced by adverse environments (Zhu *et al.*, 2005). Present study observed an increase in superoxide dismutase (SOD) and peroxidase (PDX) due to drought stress (Table 9). The increase of SOD and PDX indicates that protective mechanisms (photoprotection) are employed by the plant to tolerate moisture stress. SOD plays a central role in the enzymatic defense system in removing superoxide anions (Bowler *et al.*, 1994). The imbalance between the antioxidant defense systems and reactive oxygen species (ROS) production will ultimately results in irreversible damage and cell death (Apel and Hirt, 2004; Gill and Tuteja, 2010). Significant increase in SOD activity under drought stress was observed in *Argan spinosa* (Chakhchar *et al.*, 2015, 2016), *Olea europea* (Ahmed *et al.*, 2009; Boughalleb and Mhamdi, 2011), *Saccharum officinarum* (Patade *et al.*, 2011), *Populus cathayana* (Zhang *et al.*, 2012) and *Jatropha curcas* (Pompelli *et al.*, 2010). This significant increase in SOD activity indicates the ability to eliminate the increased ROS in teak seedling indicating an adaptation to alleviate oxidative stress. The increase of PDX activity could reflect the changed mechanical properties of the cell wall. PDX activity is an adaptive trait that helps to overcome the damage to the tissue metabolism by reducing toxic levels of H_2O_2 produced during cell metabolism. PDX offers protection against oxidative stress by converting H_2O_2 to water and oxygen (Blokhina *et al.*, 2003).

Upon relieving of stress, five of eleven accessions reduced the SOD to normal levels, while the rest showed partial recovery (Figure 6). Increase in SOD and PDX activities due to stress and rapid reversal was noticed in *Phoebe zhennan* saplings (Hu *et al.*, 2015), warm-temperate woody species (Li *et al.*, 2019) and olive trees (Sofa *et al.*, 2008).

5.3.8 Malondialdehyde and cell membrane

Malondialdehyde in cells is a reactive aldehyde produced as result of lipid peroxidation of polyunsaturated fatty acids by ROS. Cell membrane stability index (CMSI) and malondialdehyde (MDA) increased significantly during drought stress and caused considerable damage to cellular membranes in all teak accessions (Table 9). Despite photoprotection (physio-protective mechanisms) and antioxidant substances to counter the ROS damage, cells membrane damage by ROS occurred in all teak accessions during water stress. Drought stress-induced damage on the cellular membranes has been reported in many previous studies (Yin *et al.*, 2005a; Duan *et al.*, 2005; Elsheery and Cao, 2008; Xiao *et al.*, 2009). Increased MDA level indicates further oxidative damage to teak leaves.

Resuming irrigation lead to slow recovery in CMSI and MDA except KFRI T55 (Fig. 7). Rapid increase in CMSI and MDA during water stress and reversible after rehydration has been reported in teak (Husen, 2010), *Populus cathayana*, *Populus populations* (Liao *et al.*, 2018), Mexican lime (Shekafandeh *et al.*, 2019), suger cane (Devi *et al.*, 2018) and *Artemisia halodendron* (Chen *et al.*, 2019).

4.5. CORRELATION OF DROUGHT STRESS AND DROUGHT RECOVERY ON NUMBER OF LEAVES

Many studies have been conducted to find out how certain enzymes regulate the activity of plants during drought stress and how this affects the physiological parameters in order to give tolerance to plants (Deltoro *et al.*, 1998; Yordanov *et al.*, 2000). Understanding the correlation of physiological and biochemical responses to water deficit will help in breeding plant cultivars having high yield and stability under drought conditions (Yordanov *et al.*, 2000).

Among the physiological characters studied, only chlorophyll content and photosynthetic pigments showed significant positive correlation with the

dependent variable (number of leaves). The correlation coefficient observed were (0.81) for chlorophyll a (0.69) for Chlorophyll b and (0.69) for total chlorophyll (Table 10). These characters had significant positive correlation with drought recovery too (table 11). It has been already proved that photosynthetic pigments are having direct correlation with drought stress and drought recovery in plants (Chen *et al.*, 2016). Photosynthetic pigment absorbs energy from light and hence, the foliar chlorophyll content is an important factor affecting the performance of plant photosynthesis (Taiz and Zeiger, 2006). Maintaining lower chlorophyll content under serious drought stress may help plants to reduce photo-oxidative damage, which occurs when photosynthesis is inhibited and light excitation energy is in excess (Aranjuelo *et al.*, 2010). Several studies had showed that drought stress visibly decreases the chlorophyll a, chlorophyll b and total chlorophyll content of different crops (Mafakheri *et al.*, 2010), which indicate that the presence of low levels of chlorophyll in leaves was a general symptom of stress. The excessive excitation energy absorbed by photosynthetic pigment in photosystem II will lead to an impairment of photosynthetic function, progressing to an accumulation of reactive oxygen species (ROS) and resulting in oxidative stress (Pinto-Marijuan and Munne-Bosch, 2014). Alternatively, maintaining higher chlorophyll content during water stress contributes to rapid recovery of photosynthesis (Chen *et al.*, 2016). Hence, it can be concluded that chlorophyll content was significantly correlated with drought stress and drought recovery in plants.

Maintaining optimal water status is crucial to optimal physiological functioning and growth. Relative water content (RWC) had significant negative relation with number of leaves during drought recovery and no correlation with drought stress (Tables 10 and 11). This finding disagrees with many studies that have suggested that high RWC is closely related to drought resistance (Altinkut *et al.*, 2001; Keles and Oncel, 2004). Observations at recovery agree with Muthuramu *et al.* (2010) who reported that 'days to attain 70 per cent RWC had negative and significant correlation with leaf drying during drought recovery

in rice. This result suggested that existence of high number of retained leaves may serve as an indicator of plant water status, and that a plant's ability to maintain adequate water status improves drought adaptability by enhancing stress relief and drought recovery. It can be concluded that leaf water potential, is a key indicator of the degree of cell and tissue hydration.

Transpiration and photosynthetic rates had negative and significant correlation with number of leaves during water recovery while no correlation at drought stress (Tables 10 and 11). This is in disagreement with many studies which reported drought condition causing water loss which lowers rates of carbon uptake and hence lowers rates of net photosynthesis per unit area of photosynthesizing surface in leaves (Orians and Solbrig, 1977). This indicates that during the stress, plant dropped the leaf in order to reduce transpiration rate while maintain the photosynthesis. This agrees with Orians and Solbrig (1977) who mentioned a plant gains by dropping a leaf if the cost of maintaining it during a stress period exceeds the cost of producing a new one.

Super oxide dismutase (SOD) had positive significant correlation with number of leaves during drought recovery while no correlation during drought stage (Tables 10 and 11) suggesting that the retained leaves still suffer from oxidative stress. Roldan *et al.* (2008) reported increase of SOD activity level in shoots of *Juniperus oxycedrus* seedlings after recovery period.

In the present study neither drought stress or drought recovery shows correlation with the osmotic regulators and the number of leaves (Tables 10 and 11). Several studies have suggested a positive correlation of osmotic regulation with drought response (Shangguan *et al.*, 1999; Hura *et al.*, 2007; Juby, 2018). Chen *et al.* (2016) reported no correlation between osmoregulation potential and total soluble in maize when induced to water stress and drought recover.

In the present study, the correlation analysis between relative values of physiological and biochemical changes and number of leaves indicated that relative water content, photosynthetic rate, stomatal conductance, transpiration

rate, chlorophyll a, chlorophyll b, total chlorophyll content and super oxide dismutase could be used as reliable reference indicators in the selection of drought-adaptive genotypes. More specifically, all eight indicators were related to drought recovery while chlorophyll a, chlorophyll b and total chlorophyll content were related to drought stress (Tables 10 and 11).

Hence, it can be concluded that drought recovery may play a more important role than drought stress in drought adaptation of teak. Recently, increasing importance has been attached to drought recovery in crops, particularly in light of global climatic change (Chaves *et al.*, 2009; Luo, 2010; Perrone *et al.*, 2012; Vankova *et al.*, 2012; Fang and Xiong, 2015). Fang and Xiong (2015) considered drought recovery as one of the major components of drought resistance, along with drought avoidance, drought tolerance and drought escape.

5.6 PRINCIPAL COMPONENTS ANALYSIS OF PHYSIOLOGICAL AND BIOCHEMICAL PARAMETERS AT DROUGHT STRESS AND DROUGHT RECOVERY

The principal components reflected different aspects of physiological and biochemical responses of genotypes more intuitively. In the present study, the roles of drought stress and drought recovery were comparatively analyzed. These results revealed that the physiological and biochemical bases of drought stress and drought recovery (Fig. 11 and 12) are definitely different and that difference may reduce the damage associated with drought stress on plant photosynthetic systems contributing to rapid recovery after re-watering.

5.7 CLUSTER ANALYSIS OF PHYSIOLOGICAL AND BIOCHEMICAL PARAMETERS AT DROUGHT STRESS AND DROUGHT RECOVERY

Cluster analysis attempted to differentiate the accessions based on physiological and biochemical characteristics of drought response. Data obtained from physiological and biochemical traits of eleven accessions in both drought stress and recovery stages were analyzed using hierarchical euclidean cluster

analysis, which allows discriminating individuals maintaining homogeneity within the group and heterogeneity between the groups.

Eleven plus trees accessions were grouped into six clusters in case of drought stress (Table 15) while seven clusters in case of drought recovery (Table 16). It helped to identify the most distant accessions and most closely placed ones for further breeding. For drought stress, the cluster II possessed maximum number of accessions whereas the remaining had two in each.

For drought recovery, the cluster VII possessed maximum number of accessions whereas the least number was observed for the cluster I, IV, V and VI. So, KFRI T1, T9, T16 and T116 differed in their morphological, physiological and biochemical characters than others.

Greater variability in the base population ensures more success in tree improvement programme. The primary objective of germplasm conservation is to collect and preserve the genetic variability in indigenous collection of a species to make it available for present and future generations. Our results showed the clustering of the genotypes into groups differing in drought stress and recovery of selected teak accessions. Beside, both stages had considerable variations that allowed separating the groups of similarities and thus indicated that tolerance to drought in teak plus trees might be having a wide genetic background.



SUMMARY



SUMMARY

The study entitled “**Drought response in plus trees of teak (*Tectona grandis* Linn. f.) provenances of Kerala**” was conducted in College of Forestry, Vellanikkara during 2018-2019. In the study, an attempt was done to evaluate the variability of drought stress and drought recovery on morphological, physiological and biochemical responses among eleven plus trees of teak from different provenances of Kerala as well as to understand the role of physiological and biochemical responses in teak during drought stress and recovery.

The salient features of the study are summarised below:

Experiment-I: Screening of ramet performance

- One year ramets of teak plus tree from different provenance of Kerala were used to evaluate the variability among them.
- At the first, third and six month in nursery, the plus trees were evaluated for height, diameter, leaves number and branches number.
- At the sixth month, variation was absent among the eleven teak accession in term of height, diameter, leaves number and branches number.
- Physiological and biochemical characters also were investigated at six month stage, and the variation was detected in characters of stomatal conductance, nitrate reductase activity, free amino acid, proline and peroxidase.

Experiment-II: Screening of ramet at drought and recovery

- Drought stress was induced by withholding water consecutively, till leaves exhibited symptoms of wilting. Plantlets were exposed to two consecutive drought treatments with an intervening period of stress relief through rehydration for six days.
- During drought stress and recovery, biometric, physiological and biochemical character were observed.

- After inducing drought stress, growth parameters slowed down in all accessions between plus trees.
- All physiological parameters except canopy air temperature differences and cell membrane stability index were lower under water stress. All physiological parameters reversed near or to level of normal after relieving water stress.
- During water stress, variability of physiological characters among tested accessions were observed in stomatal conductance, chlorophyll a, chlorophyll b and total chlorophyll content. While, no variation in all physiological parameters among accessions after rehydration.
- Most of biochemical parameters like free amino acid, proline, glycine betaine, total soluble sugar, super oxide dismutase, peroxidase and malondialdehyde increased after inducing drought. However, total soluble protein and nitrate decreased under drought induction. After rehydration, most of these physiological and biochemical parameters rapidly returned to the level of normal irrigated condition.
- Variability on biochemical characters were observed in proline, super oxide dismutase, peroxidase and malondialdehyde during drought stress among accession. In drought recovery, character like nitrate reductase, free amino acid, proline and peroxidase showed variability.
- Among the twenty characters used for estimating the correlation under drought stress in teak plus tree accessions, leaf number showed positive correlation with content of chlorophyll a, chlorophyll b and total chlorophyll content.
- Relative water content, photosynthetic rate, stomatal conductance, transpiration rate chlorophyll a, chlorophyll b, total chlorophyll content and superoxide dismutase had a positive correlation with number of leaves during drought recovery.

- Both drought stress and recovery are key determinants of seedling plant drought adaptation, drought recovery may play the more important role as more character correlated with number of leaves.
- A hierarchical cluster analysis was done for the eleven accessions based on the Euclidian squared distance. During drought stress, the accessions grouped into five clusters; cluster II had three of accessions, whereas remaining clusters had two accessions each.
- In case of drought recovery, the accession was grouped into seven clusters; cluster VII had three accessions, while only one accession was present in cluster I, IV, V and VI.
- Principal component analysis showed that in case of drought stress, first two components accounted for 41.9 per cent of the total variability, which was mainly contributed positively by relative water content, nitrate reductase, free amino acid, proline and malondialdehyde.
- At drought recovery, first two components of PCA together accounted for 53.2 per cent of the total variability, which was mainly contributed positively by photosynthetic rate, stomatal conductance, transpiration rate, chlorophyll stability index, glycine betaine and malondiadehyde.
- Selection index were worked out to select accession tolerant to drought stress and recovery based on biochemical parameters and total chlorophyll content using first principle component as index. It was found that KFRI T55 was most tolerant and quickest to recover after relieving stress among accession.
- These results could be useful in selection of drought tolerant clones of Teak.



REFERENCE



REFERENCES

- Abid, M., Ali, S., Qi, L. K., Zahoor, R., Tian, Z., Jiang, D., Snider, J. L., and Dai, T. 2018. Physiological and biochemical changes during drought and recovery periods at tillering and jointing stages in wheat (*Triticum aestivum* L.). *Sci. Rep.*, 8(1): 4615.
- Ahmed, C. B., Rouina, B. B., Sensoy, S., Boukhris, M., and Abdallah, F. B. 2009. Changes in gas exchange, proline accumulation and antioxidative enzyme activities in three olive cultivars under contrasting water availability regimes. *Environ. Exp. Bot.*, 67(2): 345-352.
- Ain-Lhout, F., Zunzunegui, M., Barradas, M. D., Tirado, R., Clavijo, A., and Novo, F. G. 2001. Comparison of proline accumulation in two Mediterranean shrubs subjected to natural and experimental water deficit. *Plant Soil*, 230(2): 175-183.
- Akinci, S. and Losel, D. M. 2009. The soluble sugars determination in Cucurbitaceae species under water stress and recovery periods. *Adv. Environ. Biol.*, 3(2): 175-183.
- Alcantara, B. K. and Veasey, E. A. 2013. Genetic diversity of teak (*Tectona grandis* LF) from different provenances using microsatellite markers. *Riv. Árvore*, 37(4): 747-758.
- Alcazar, R., Marco, F., Cuevas, J. C., Patron, M., Ferrando, A., Carrasco, P., Tiburcio, A. F., and Altabella, T. 2006. Involvement of polyamines in plant response to abiotic stress. *Biotechnol. Lett.*, 28(23): 1867-1876.
- Allen, C. D., Breshears, D. D., and Mc-Dowell, N. G. 2015. On underestimation of global vulnerability to tree mortality and forest die-off from hotter drought in the Anthropocene. *Ecosphere*, 6(8): 1-55.

- Allen, D. J. and Ort, D. R. 2001. Impacts of chilling temperatures on photosynthesis in warm-climate plants. *Trends Plant Sci.*, 6(1): 36-42.
- Altinkut, A., Kazan, K., Ipekci, Z., and Gozukirmizi, N. 2001. Tolerance to paraquat is correlated with the traits associated with water stress tolerance in segregating F2 populations of barley and wheat. *Euphytica*, 121(1): 81-86.
- Amani, I., Fischer, R. A., and Reynolds, M. P. 1996. Canopy temperature depression association with yield of irrigated spring wheat cultivars in a hot climate. *J. Agron. Crop Sci.*, 176(2):119-129.
- Ameys, M., Wertin, T. M., Bauweraerts, I., Mc-Guire, M. A., Teskey, R. O. and Steppe, K. 2012. The effect of induced heat waves on *Pinus taeda* and *Quercus rubra* seedlings in ambient and elevated CO₂ atmospheres. *New Phytol.*, 196(2): 448-461.
- Anderson, C. M. and Kohorn, B. D. 2001. Inactivation of *Arabidopsis* SIP1 leads to reduced levels of sugars and drought tolerance. *J. Plant Physiol.*, 158(9): 1215-1219.
- Anjum, F., Yaseen, M., Rasool, E., Wahid, A., and Anjum, S. 2003. Water stress in barley (*Hordeum vulgare* L.) I. Effect on morphological characters. *Seeds*, 105: 266-271.
- Anjum, S. A., Xie, X. Y., Wang, L. C., Saleem, M. F., Man, C., and Lei, W. 2011. Morphological, physiological and biochemical responses of plants to drought stress. *Afr. J. Agric. Res.*, 6(9): 2026-2032.
- Antonovics, J. 1971. The effect of heterogenous environment on the genetics of natural population. *Am. Sci.*, 59: 593-595.
- Apel, K. and Hirt, H. 2004. Reactive oxygen species: metabolism, oxidative stress, and signal transduction. *Annu. Rev. Plant Biol.*, 55: 373-399.

- Aranjuelo, I., Molero, G., Erice, G., Avice, J. C., and Nogues, S. 2010. Plant physiology and proteomics reveals the leaf response to drought in alfalfa (*Medicago sativa* L.). *J. Exp. Bot.*, 62(1): 111-123.
- Araujo, W. L., Tohge, T., Ishizaki, K., Leaver, C. J., and Fernie, A. R. 2011. Protein degradation—an alternative respiratory substrate for stressed plants. *Trends Plant Sci.*, 16(9): 489-498.
- Aro, E. M., Suorsa, M., Rokka, A., Allahverdiyeva, Y., Paakkarinen, V., Saleem, A., Battchikova, N., and Rintamäki, E. 2004. Dynamics of photosystem II: a proteomic approach to thylakoid protein complexes. *J. Exp. Bot.*, 56(411): 347-356.
- Aro, E. M., Virgin, I., and Andersson, B. 1993. Photoinhibition of photosystem II. Inactivation, protein damage and turnover. *Biochim. Biophys. Acta (BBA)-Bioenerg.*, 1143(2): 113-134.
- Asada, K. 1999. The water-water cycle in chloroplasts: scavenging of active oxygens and dissipation of excess photons. *Annu. Rev. Plant Biol.*, 50(1): 601-639.
- Ashraf, M. 2003. Relationships between leaf gas exchange characteristics and growth of differently adapted populations of Blue panicgrass (*Panicum antidotale* Retz.) under salinity or waterlogging. *Plant Sci.*, 165(1): 69-75.
- Ashraf, M. F. M. R. and Foolad, M. 2007. Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Environ. Exp. Bot.*, 59(2): 206-216.
- Baena-Gonzalez, E. and Aro, E. M. 2002. Biogenesis, assembly and turnover of photosystem II units. *Philos. Trans. R. Soc. Lond. Series B: Biol. Sci.*, 357(1426): 1451-1460.

- Bai, W. M., Wang, Z. W., Chen, Q. S., Zhang, W. H., and Li, L. H. 2008. Spatial and temporal effects of nitrogen addition on root life span of *Leymus chinensis* in a typical steppe of Inner Mongolia. *Funct. Ecol.*, 22(4): 583-591.
- Baisak, R., Rana, D., Acharya, P.B., and Kar, M. 1994. Alteration in the activities of active oxygen scavenging enzymes of wheat leaves subjected to water stress. *Plant Cell Physiol.* 35: 489-495.
- Bajji, M., Lutts, S., and Kinet, J. M. 2001. Water deficit effects on solute contribution to osmotic adjustment as a function of leaf ageing in three durum wheat (*Triticum durum* Desf.) cultivars performing differently in arid conditions. *Plant Sci.*, 160(4): 669-681.
- Barber, J. and Andersson, B. 1992. Too much of a good thing: light can be bad for photosynthesis. *Trends Biochem. Sci.*, 17(2): 61-66.
- Bardzik, J. M., Marsh, H. V., and Havis, J. R. 1971. Effects of water stress on the activities of three enzymes in maize seedlings. *Plant Physiol.*, 47(6): 828-831.
- Barros, J. A., Cavalcanti, J. H. F., Medeiros, D.B., Nunes-Nesi, A., Avin-Wittenberg, T., Fernie, A.R., and Araujo, W.L. 2017. Autophagy deficiency compromises alternative pathways of respiration following energy deprivation in *Arabidopsis thaliana*. *Plant Physiol.*, 175(1): 62-76.
- Barrs, H. D. 1968. Determination of water deficits in plant tissues. In: Kozloowski, T. (ed.), *Water deficits and plant growth*. Academic Press, New York, 236-368.
- Bartels, D. and Sunkar, R. 2005. Drought and salt tolerance in plants. *Crit. Rev. Plant Sci.*, 24(1): 23-58.

- Bashir, F., Siddiqi, T. O., and Iqbal, M. 2007. The antioxidative response system in *Glycine max* (L.) Merr. exposed to Deltamethrin, a synthetic pyrethroid insecticide. *Environ. Pollut.*, 147(1): 94-100.
- Bates, L. S., Waldren, R. P., and Teare, I. D. 1973. Rapid determination of free proline for water-stress studies. *Plant Soil*, 39(1): 205-207.
- Beevers, L. and Hageman, R. H. 1969. Nitrate reduction in higher plants. *Annu. Rev. Plant Physiol.* 20(1): 495-522.
- Benhassaine-Kesri, G., Aid, F., Demandre, C., Kader, J. C., and Mazliak, P. 2002. Drought stress affects chloroplast lipid metabolism in rape (*Brassica napus*) leaves. *Physiol. Plant.*, 115(2): 221-227.
- Benson, E. J., Hartnett, D. C., and Mann, K. H. 2004. Belowground bud banks and meristem limitation in tall grass prairie plant populations. *Am. J. Bot.*, 91(3): 416-421.
- Bernacchi, C. J., Portis, A. R., Nakano, H., Von Caemmerer, S., and Long, S. P. 2002. Temperature response of mesophyll conductance. Implications for the determination of Rubisco enzyme kinetics and for limitations to photosynthesis in vivo. *Plant Physiol.*, 130(4): 1992-1998.
- Berry, J. A., Beerling, D. J., and Franks, P. J. 2010. Stomata: key players in the earth system, past and present. *Curr. Opinion Plant Biol.*, 13(3): 232-239.
- Bhatt, R. M. and Rao, N. S. 2005. Influence of pod load on response of okra to water stress. *Indian J. Plant Physiol.*, 10(1): 54.
- Binks, O., Meir, P., Rowland, L., da Costa, A.C.L., Vasconcelos, S. S., de Oliveira, A. A. R., Ferreira, L., Christoffersen, B., Nardini, A., and Mencuccini, M. 2016. Plasticity in leaf-level water relations of tropical rainforest trees in response to experimental drought. *New Phytol.*, 211(2): 477-488.

- Blokhina, O., Virolainen, E. and Fagerstedt, K. V. 2003. Antioxidants, oxidative damage and oxygen deprivation stress: a review. *Ann. Bot.*, 91(2): 179-194.
- Boughalleb, F. and Mhamdi, M. 2011. Possible involvement of proline and the antioxidant defense systems in the drought tolerance of three olive cultivars grown under increasing water deficit regimes. *Agri. J.*, 6(6): 378-391.
- Bowler, C., Van Camp, W., Van Montagu, M., Inze, D., and Asada, K. 1994. Superoxide dismutase in plants. *Crit. Rev. Plant Sci.*, 13(3): 199-218.
- Brodribb, T. J., Bowman, D. J., Nichols, S., Delzon, S., and Burlett, R. 2010. Xylem function and growth rate interact to determine recovery rates after exposure to extreme water deficit. *New Phytol.*, 188(2): 533-542.
- Brodribb, T.J. and Mc-Adam, S.A. 2011. Passive origins of stomatal control in vascular plants. *Sci.*, 331(6017): 582-585.
- Cano, F. J., Lopez, R., and Warren, C. R. 2014. Implications of the mesophyll conductance to CO₂ for photosynthesis and water-use efficiency during long-term water stress and recovery in two contrasting *Eucalyptus* species. *Plant Cell Environ.*, 37(11): 2470-2490.
- Carpentier, R. 1997. Influence of high light intensity on photosynthesis: photoinhibition and energy dissipation. *Handb. Photosynthesis*, 2:338-353.
- Casson, S. A. and Hetherington, A. M. 2010. Environmental regulation of stomatal development. *Curr. Opinion Plant Biol.*, 13(1): 90-95.
- Castillo, F. J., Penel, C., and Greppin, H. 1984. Peroxidase release induced by ozone in *Sedum album* leaves: involvement of Ca²⁺. *Plant Physiol.*, 74(4): 846-851.

- Castro, D. da S., dos Santos, A.O., da Silva Lobato, A. K., da Silva Gouvea, D. D., Neto, C. F. O., da Cunha, R. L. M., and da Costa, R. C. L. 2007. Concentrações de prolina e carboidratos solúveis totais em folhas de teca (*Tectona grandis* L. f) submetida ao estresse hidrico. *Rev. Bras. Biocienc*, 5(S2): 921-923.
- Chakhchar, A., Lamaoui, M., Aissam, S., Ferradous, A., Wahbi, S., El Mousadik, A., Ibensouda-Koraichi, S., Filali-Maltouf, A., and El Modafar, C. 2016. Differential physiological and antioxidative responses to drought stress and recovery among four contrasting *Argania spinosa* ecotypes. *J. Plant Interact.*, 11(1): 30-40.
- Chakhchar, A., Wahbi, S., Lamaoui, M., Ferradous, A., El Mousadik, A., Ibensouda-Koraichi, S., Filali-Maltouf, A., and El Modafar, C. 2015. Physiological and biochemical traits of drought tolerance in *Argania spinosa*. *J. Plant interact.*, 10(1): 252-261.
- Chandra, S. (2003). Effects of leaf age on transpiration and energy exchange of *Ficus glomerata*, a multipurpose tree species of central Himalayas. *Physiol. Mol. Biol. Plants*, 9: 255-260.
- Chaves, M. M. 1991. Effects of water deficits on carbon assimilation. *J. Exp. Bot.*, 42(1): 1-16.
- Chaves, M. M. and Oliveira, M. M. 2004. Mechanisms underlying plant resilience to water deficits: prospects for water-saving agriculture. *J. Exp. Bot.*, 55(407): 2365-2384.
- Chaves, M. M., Flexas, J., and Pinheiro, C. 2009. Photosynthesis under drought and salt stress: regulation mechanisms from whole plant to cell. *An. Bot.*, 103(4): 551-560.
- Chaves, M. M., Maroco, J. P., and Pereira, J. S. 2003. Understanding plant responses to drought-from genes to the whole plant. *Funct. Plant Biol.*, 30(3): 239-264.

- Chen, D., Wang, S., Cao, B., Cao, D., Leng, G., Li, H., Yin, L., Shan, L., and Deng, X. 2016. Genotypic variation in growth and physiological response to drought stress and re-watering reveals the critical role of recovery in drought adaptation in maize seedlings. *Front. Plant Sci.*, 6: 1241.
- Chen, J. W., Zhang, Q., Li, X. S., and Cao, K. F. 2010. Gas exchange and hydraulics in seedlings of *Hevea brasiliensis* during water stress and recovery. *Tree physiol.*, 30(7): 876-885.
- Chen, J., Zhao, X., Zhang, Y., Li, Y., Luo, Y., Ning, Z., Wang, R., Wang, P. and Cong, A. 2019. Effects of Drought and Rehydration on the Physiological Responses of *Artemisia halodendron*. *Water*, 11(4): 793.
- Chen, W. P., Li, P. H., and Chen, T. H. H. 2000. Glycinebetaine increases chilling tolerance and reduces chilling-induced lipid peroxidation in *Zea mays* L. *Plant Cell Environ.*, 23(6): 609-618.
- Choat, B., Brodribb, T. J., Brodersen, C. R., Duursma, R. A., Lopez, R., and Medlyn, B. E. 2018. Triggers of tree mortality under drought. *Nature*, 558(7711): 531-539.
- Codesido, V. and Fernandez-Lopez, J., 2008. Juvenile genetic parameter estimates for vigour, stem form, branching habit and survival in three *radiata pine* (*Pinus radiata* D. Don) progeny tests in Galicia, NW Spain. *Eur. J. For. Res.*, 127(4): 315-325.
- Corlett, R. T. 2016. The impacts of droughts in tropical forests. *Trends Plant Sci.*, 21(7): 584-593.
- Curtis, E. M., Knight, C. A., Petrou, K., and Leigh, A. 2014. A comparative analysis of photosynthetic recovery from thermal stress: a desert plant case study. *Oecologia*, 175(4): 1051-1061.

- Dalvi, U. S. 2015. Studies on biochemical and molecular responses to biotic (*Fusarium WILT*) and abiotic (*Drought*) stresses in Chickpea (*Cicer arietinum* L). Doctoral dissertation, Mahatma Phule Krishi Vidyapeeth, Rahuri
- Deltoro, V. I., Calatayud, A., Gimeno, C., Abadía, A., and Barreno, E. 1998. Changes in chlorophyll a fluorescence, photosynthetic CO₂ assimilation and xanthophyll cycle interconversions during dehydration in desiccation-tolerant and intolerant liverworts. *Planta*, 207(2): 224-228.
- Devi, K., Gomathi, R., Kumar, R. A., Manimekalai, R., and Selvi, A. 2018. Field tolerance and recovery potential of sugarcane varieties subjected to drought. *Indian J. Plant Physiol.*, 23(2): 71-282.
- Dhanda, S. S., Sethi, G. S., and Behl, R. K. 2004. Indices of drought tolerance in wheat genotypes at early stages of plant growth. *J. Agron. Crop Sci.* 190(1): 6-12.
- Dhindsa, R. S. and Matowe, W. 1981. Drought tolerance in two mosses: correlated with enzymatic defence against lipid peroxidation. *J. Exp. Bot.*, 32(1): 79-91.
- Dien, D. C., Mochizuki, T., and Yamakawa, T. 2019. Effect of various drought stresses and subsequent recovery on proline, total soluble sugar and starch metabolisms in Rice (*Oryza sativa* L.) varieties. *Plant Prod. Sci.*, 22(4): 530-545.
- Dlamini, C. S. 2010. Provenance and family variation in germination and early seedling growth in *Sclerocarya birrea* sub-species *caffra*. *J. Hortic. For.*, 2(9): 229-235.
- Drossopoulos, J. B., Karamanos, A. J., and Niavis, C. A. 1987. Changes in ethanol soluble carbohydrates during the development of two wheat cultivars subjected to different degrees of water stress. *Ann. Bot.*, 59(2): 173-180.

- Duan, B., Lu, Y., Yin, C., Junttila, O., and Li, C. 2005. Physiological responses to drought and shade in two contrasting *Picea asperata* populations. *Physiol. Plant.*, 124(4): 476-484.
- Dupuy, B. and Verhaegen, D. 1993. Plantation grown teak (*Tectona grandis*) in Cote d'ivoire. *Bois. For.Trop.*, 235: 9-24.
- Egenti, L. C. (1977). The international provenance trials of teak (*Tectona grandis* L. f.) in Nigeria. In: Proceedings of Joint IUFRO Workshop, S2 - 02 - 08, S2 - 03 - 01. Brisbane Vol. II: 754 - 760.
- Elsheery, N. I. and Cao, K. F. 2008. Gas exchange, chlorophyll fluorescence, and osmotic adjustment in two mango cultivars under drought stress. *Acta Physiol. Plant.*, 30(6): 769-777.
- El-yamani, M., Boussakouran, A. and Rharrabti, Y., 2019. Physiological and biochemical responses of young olive trees (*Olea europaea* L.) to water stress during flowering. *Arch. Biol. Sci.*, 71(1): 123-132.
- Epron, D., Dreyer, E., and Breda, N. 1992. Photosynthesis of oak trees [*Quercus petraea* (Matt.) Liebl.] during drought under field conditions: diurnal course of net CO₂ assimilation and photochemical efficiency of photosystem II. *Plant Cell Environ.*, 15(7): 809-820.
- Ethier, G. J. and Livingston, N. J. 2004. On the need to incorporate sensitivity to CO₂ transfer conductance into the Farquhar–von Caemmerer–Berry leaf photosynthesis model. *Plant Cell Environ.*, 27(2): 137-153.
- Fallon, K. M. and Phillips, R. 1989. Responses to water stress in adapted and unadapted carrot cell suspension cultures. *J. Exp. Bot.*, 40(6): 681-687.
- Fang, Y. and Xiong, L. 2015. General mechanisms of drought response and their application in drought resistance improvement in plants. *Cell Mol. Life Sci.*, 72(4): 673-689.

- Farooq, M., Wahid, A., Kobayashi, N., Fujita, D. B. S. M. A., and Basra, S. M. A. 2009. Plant drought stress: effects, mechanisms and management. In *Sustain. Agric.*, Springer, Dordrecht, pp. 153-188.
- Fazeli, F., Ghorbanli, M. and Niknam, V. 2006. Effect of drought on water relations, growth and solute accumulation in two sesame cultivars. *Pakist. J. Biol. Sci.*, 9(9): 1829-1835.
- Ferguson, J. H. A. (1938) Selectie op stemkwaliteit (Selection of stem quality). *Tectona* 31 (9/10): 727-740.
- Ferrario-Mery, S., Valadier, M. H., and Foyer, C. H. 1998. Overexpression of nitrate reductase in tobacco delays drought-induced decreases in nitrate reductase activity and mRNA. *Plant Physiol.*, 117(1): 293-302.
- Foyer, C. H., Valadier, M. H., Migge, A., and Becker, T.W. 1998. Drought-induced effects on nitrate reductase activity and mRNA and on the coordination of nitrogen and carbon metabolism in maize leaves. *Plant Physiol.*, 117(1): 283-292.
- Frosi, G., Harand, W., Oliveira, M. T. D., Pereira, S., Cabral, S. P., Montenegro, A. A. D. A., and Santos, M.G. 2017. Different physiological responses under drought stress result in different recovery abilities of two tropical woody evergreen species. *Acta Bot. Bras.*, 31(2): 153-160.
- Galmes, J., Medrano, H., and Flexas, J. 2007. Photosynthetic limitations in response to water stress and recovery in Mediterranean plants with different growth forms. *New Phytol.*, 175: 81–93.
- Gan, S. and Amasino, R. M. 1997. Making sense of senescence (molecular genetic regulation and manipulation of leaf senescence). *Plant Physiol.*, 113(2): 313.

- Gartner, K., Nadezhdina, N., Englisch, M., Cermak, J., and Leitgeb, E. 2009. Sap flow of birch and Norway spruce during the European heat and drought in summer 2003. *For. Ecol. Manag.*, 258(5): 590-599.
- Gaspar, T., Franck, T., Bisbis, B., Kevers, C., Jouve, L., Hausman, J.F., and Dommès, J. 2002. Concepts in plant stress physiology. Application to plant tissue cultures. *Plant Growth Reg.*, 37(3): 263-285.
- Ghosh, S. C., Asanuma, K. I., Kusutani, A., and Toyota, M. 2000. Effects of moisture stress at different growth stages on the amount of total nonstructural carbohydrate, nitrate reductase activity and yield of potato. *Jpn. J. Trop. Agric.*, 44(3): 158-166.
- Gill, S. S. and Tuteja, N. 2010. Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. *Plant Physiol. and biochem.*, 48(12): 909-930.
- Gollan, T., Schurr, U., and Schulze, E. D. 1992. Stomatal response to drying soil in relation to changes in the xylem sap composition of *Helianthus annuus*. I. The concentration of cations, anions, amino acids in, and pH of, the xylem sap. *Plant Cell Environ.*, 15(5): 551-559.
- Gomes, M. T. G., da Luz, A. C., dos Santos, M. R., Batitucci, M. D. C. P., Silva, D. M., and Falqueto, A. R. 2012. Drought tolerance of passion fruit plants assessed by the OJIP chlorophyll a fluorescence transient. *Sci. Hortic.*, 142: 49-56.
- Gorham, J., Hughes, L. L., and Wyn Jones, R. G. 1981. Low-molecular-weight carbohydrates in some salt-stressed plants. *Physiol. plant*, 53(1): 27-33.
- Greenway, H. and Munns, R. 1980. Mechanisms of salt tolerance in nonhalophytes. *Annu. Rrev. Plant physiol.*, 31(1): 149-190.
- Grieve, C. M. and Grattan, S. R. 1983. Rapid assay for determination of water soluble quaternary ammonium compounds. *Plant Soil*, 70(2): 303-307.

- Guerfel, M., Baccouri, O., Boujnah, D., Chaïbi, W., and Zarrouk, M. 2009. Impacts of water stress on gas exchange, water relations, chlorophyll content and leaf structure in the two main Tunisian olive (*Olea europaea* L.) cultivars. *Sci. Hortic.*, 119(3): 257-263.
- Hedge, J. E., Hofreiter, B. T., and Whistler, R. L. 1962. Carbohydrate chemistry. *Academic Press, New York*, 17 p.
- Hieng, B., Ugrinović, K., Sustar-Vozlic, J., and Kidric, M. 2004. Different classes of proteases are involved in the response to drought of *Phaseolus vulgaris* L. cultivars differing in sensitivity. *J. Plant Physiol.*, 161(5): 519-530.
- Hildebrandt, T. M. 2018. Synthesis versus degradation: directions of amino acid metabolism during *Arabidopsis* abiotic stress response. *Plant Mol. Biol.*, 98(1-2): 121-135.
- Hildebrandt, T. M., Nesi, A. N., Araújo, W. L., and Braun, H. P. 2015. Amino acid catabolism in plants. *Mol. Plant*, 8(11): 1563-1579.
- Hirota, T., Izumi, M., Wada, S., Makino, A., and Ishida, H. 2018. Vacuolar protein degradation via autophagy provides substrates to amino acid catabolic pathways as an adaptive response to sugar starvation in *Arabidopsis thaliana*. *Plant Cell Physiol.*, 59(7): 1363-1376.
- Holmstrom, K. O., Somersalo, S., Mandal, A., Palva, T. E., and Welin, B. 2000. Improved tolerance to salinity and low temperature in transgenic tobacco producing glycine betaine. *J. Exp. Bot.*, 51(343): 177-185.
- Hong, Z., Lakkineni, K., Zhang, Z., and Verma, D. P. S. 2000. Removal of feedback inhibition of Δ^1 -pyrroline-5-carboxylate synthetase results in increased proline accumulation and protection of plants from osmotic stress. *Plant Physiol.*, 122(4): 1129-1136.

- Hong-Bo, S., Xiao-Yan, C., Li-Ye, C., Xi-Ning, Z., Gang, W., Yong-Bing, Y., Chang-Xing, Z., and Zan-Min, H. 2006. Investigation on the relationship of proline with wheat anti-drought under soil water deficits. *Colloids Surf B Biointerfaces*, 53(1): 113-119.
- Hortensteiner, S. 2009. Stay-green regulates chlorophyll and chlorophyll-binding protein degradation during senescence. *Trends Plant Sci.*, 14(3): 155-162.
- Horváth, E., Pal, M., Szalai, G., Paldi, E., and Janda, T. 2007. Exogenous 4-hydroxybenzoic acid and salicylic acid modulate the effect of short-term drought and freezing stress on wheat plants. *Biol. Plant.*, 51(3): 480-487.
- Hsiao, T. C. 1970. Rapid changes in levels of polyribosomes in *Zea mays* in response to water stress. *Plant Physiol.*, 46(2): 281-285.
- Hu, Y., Wang, B., Hu, T., Chen, H., Li, H., Zhang, W., Zhong, Y., and Hu, H. 2015. Combined action of an antioxidant defence system and osmolytes on drought tolerance and post-drought recovery of *Phoebe zhennan* S. Lee saplings. *Acta Physiol. Plant.*, 37(4): 84.
- Huang, J., Hirji, R., Adam, L., Rozwadowski, K. L., Hammerlindl, J. K., Keller, W. A., and Selvaraj, G. 2000. Genetic engineering of glycinebetaine production toward enhancing stress tolerance in plants: metabolic limitations. *Plant Physiol.*, 122(3): 747-756.
- Hudak, I., Dobranszki, J., Sardi, E., and Hevesi, M. 2010. Changes in carbohydrate content of potato calli during osmotic stress induced by mannitol. *Acta Biol. Hung.*, 61(2): 234-236.
- Hueve, K., Bichele, I., Rasulov, B., and Niinemets, U. L. O. 2011. When it is too hot for photosynthesis: heat-induced instability of photosynthesis in relation to respiratory burst, cell permeability changes and H₂O₂ formation. *Plant Cell Environ.*, 34(1): 113-126.

- Hura, T., Grzesiak, S., Hura, K., Thiemt, E., Tokarz, K., and Wędzony, M. 2007. Physiological and biochemical tools useful in drought-tolerance detection in genotypes of winter triticale: accumulation of ferulic acid correlates with drought tolerance. *Ann. Bot.*, 100(4): 767-775.
- Husen, A. 2010. Growth characteristics, physiological and metabolic responses of teak (*Tectona grandis* Linn. f.) clones differing in rejuvenation capacity subjected to drought stress. *Silvae Genet.*, 59(1-6): 124-136.
- Iturbe-Ormaetxe, I., Escuredo, P.R., Arrese-Igor, C., and Becana, M. 1998. Oxidative damage in pea plants exposed to water deficit or paraquat. *Plant Physiol.*, 116(1): 173-181.
- Izanloo, A., Condon, A.G., Langridge, P., Tester, M., and Schnurbusch, T. 2008. Different mechanisms of adaptation to cyclic water stress in two South Australian bread wheat cultivars. *J. Exp. Bot.*, 59(12): 3327-3346.
- Jayasankar, S., Babu, L.C., Sudhakara, K., and Dhanesh Kumar, P. 1999. Evaluation of provenances for seedling attributes in teak (*Tectona grandis* Linn. F.). *Silvae Genet.*, 48: 115-121.
- Jayasankar, S., Sudhakara, K., and Babu, L. C. 2003. Provenance variation in growth, physiology, anatomical characteristics and foliar nutrient status of teak (*Tectona grandis*) seedlings. *J. Trop. For. Sci.*, 15(1): 37-50.
- Jones, H.G. and Corlett, J.E. 1992. Current topics in drought physiology. *J. Agric. Sci.*, 119(3): 291-296.
- Jones, N. 1970. The relationship between the form and value of some tree species in West Africa. *Second world consultation on forest tree breeding*, Washington, DC, USA, 1, pp.317-332.
- Jongdee, B., Fukai, S., and Cooper, M. 2002. Leaf water potential and osmotic adjustment as physiological traits to improve drought tolerance in rice. *Field Crops Res.*, 76(2-3): 153-163.

- Juby, B. 2019. Breeding for drought tolerance in Cocoa (*Theobroma cacao* L.). M. Sc. Thesis, Kerala Agricultural University, Thrissur, Vellanikkara.
- Kadambi, K. 1945. Teak-seed Origin Experiments in Mysore. *Indian For.*, 71(8): 265-269.
- Kaiser, W. M., Weiner, H., and Huber, S. C. 1999. Nitrate reductase in higher plants: a case study for transduction of environmental stimuli into control of catalytic activity. *Physiol. Plant.*, 105(2): 384-389.
- Kaiser, W.M. 1987. Effects of water deficit on photosynthetic capacity. *Physiol. Plant.*, 71(1): 142-149.
- Kaloyereas, S. A. 1958. A New Method of Determining Drought Resistance. *Plant Physiol.*, 33(3): 232.
- Karimi, S., Rahemi, M., Rostami, A. A., and Sedaghat, S. 2018. Drought Effects on Growth, Water Content and Osmoprotectants in Four Olive Cultivars with Different Drought Tolerance. *Int. J. Fruit Sci.*, 18(3): 254-267.
- Kato, Y. and Sakamoto, W. 2009. Protein quality control in chloroplasts: a current model of D1 protein degradation in the photosystem II repair cycle. *J. Biochem.*, 146(4): 463-469.
- Kedharnath S 1980. Gentic improvement of forest trees, position paper presented at the Second Forestry Conference, Dehradun, India
- Kedharnath, S. 1982. Plus tree selection-a tool in forest tree improvement. In *Improvement of forest biomass: symposium proceedings/edited by PK Khosla*. Solan, India: Indian Society of Tree Scientists, pp.13-21.
- Kedharnath, S. and Matthews, J. D. 1962. Improvement of Teak by selection and breeding. *Indian For.*, 88(4): 277-284.

- Kedharnath, S. and Sing, P. 1975. Studies on natural variation in susceptibility of *Tectona* to leaf skeletoniser, *Pyrausta machaeralis* (Lepidoptera: Pyralidae), paper presented at FAO/IUFRO Symp. on Forest diseases and insects; New Delhi--India
- Kedharnath, S., Chetty, C.R. and Rawat, M.S. 1969. Estimation of genetic parameters in teak (*Tectona grandis*) without raising progeny. *Indian For.*, 95(4): 238-245.
- Keiding, H. 1966. Aim and prospects of teak breeding in Thailand. *Nat. Hist. Bull. Siam Soc.*, 21(1): 45 - 62.
- Keles, Y. and Oncel, I. 2004. Growth and solute composition in two wheat species experiencing combined influence of stress conditions. *Russ. J. Plant Physiol.*, 51(2): 203-209.
- Keogh, R. M. 1979. Does teak have a future in tropical America? A survey of *Tectona grandis* in the Caribbean, Central America, Venezuela and Colombia. *Unasylva*. 31(126): 13-19.
- Keogh, R. M. 1982. Teak (*Tectona grandis* Linn. f.) provisional site classification chart for the Caribbean, Central America, Venezuela and Colombia. *For. Ecol. Manag.*, 4(2):143-153.
- Keogh, R. M. 2009. The future of teak and the high-grade tropical hardwood sector.
- Khanna-Chopra, R., Chaturverdi, G. S., Aggarwal, P. K., and Sinha, S. K. 1980. Effect of potassium on growth and nitrate reductase during water stress and recovery in maize. *Physiol. plant.*, 49(4): 495-500.
- Kishor, P. K., Hong, Z., Miao, G. H., Hu, C. A. A., and Verma, D. P. S. 1995. Overexpression of [δ]-pyrroline-5-carboxylate synthetase increases proline production and confers osmotolerance in transgenic plants. *Plant physiol.*, 108(4): 1387-1394.

- Kollert, W. and Cherubini, L. 2012. Teak resources and market assessment (2010) FAO planted forests and trees working paper FP/47/E. Rome, Italy.
- Kollert, W. and Kleine, M. 2017. *global teak study: analysis, evaluation and future potential of teak resources*. Vienna: International Union of Forestry Organizations. IUFRO. World Series Volume 36.
- Kong, W., Liu, F., Zhang, C., Zhang, J. and Feng, H. 2016. Non-destructive determination of Malondialdehyde (MDA) distribution in oilseed rape leaves by laboratory scale NIR hyperspectral imaging. *Sci. Rep.*, 6: 35393.
- Kramer, P. J. 1969. *Plant and soil water relationships: a modern synthesis*. Academic Press, San Diego, 482 p.
- Kramer, P. J. and J. S. Boyer. 1995. *Water relations of plants and soils*. Academic Press, San Diego, 482 p.
- Kusaka, M., Lalusin, A. G., and Fujimura, T. 2005. The maintenance of growth and turgor in pearl millet (*Pennisetum glaucum* [L.] Leeke) cultivars with different root structures and osmo-regulation under drought stress. *Plant Sci.*, 168(1): 1-14.
- Laurie, M. V. 1938. Branching and seed origin in Coorg teak plantations. *Indian For.*, 64(10): 594-600.
- Lemoine, R., La Camera, S., Atanassova, R., Dédaldéchamp, F., Allario, T., Pourtau, N., Bonnemain, J. L., Laloi, M., Coutos-Thévenot, P., Maurousset, L. and Faucher, M. 2013. Source-to-sink transport of sugar and regulation by environmental factors. *Front. Plant Sci.*, 4: 272.
- Li, C., Berninger, F., Koskela, J., and Sonninen, E. 2000. Drought responses of *Eucalyptus microtheca* provenances depend on seasonality of rainfall in their place of origin. *Funct. Plant Biol.*, 27(3): 231-238.

- Li, Q., Wang, N., Liu, X., Liu, S., Wang, H., Zhang, W., Wang, R., and Du, N. 2019. Growth and physiological responses to successional water deficit and recovery in four warm-temperate woody species. *Physiol. plant.*, 167: 645-660.
- Li, S., Feifel, M., Karimi, Z., Schuldt, B., Choat, B., and Jansen, S. 2015. Leaf gas exchange performance and the lethal water potential of five European species during drought. *Tree physiol.*, 36(2): 179-192.
- Liao, T., Wang, Y., Xu, C.P., Li, Y., and Kang, X.Y. 2018. Adaptive photosynthetic and physiological responses to drought and rewatering in triploid *Populus* populations. *Photosynthetica*, 56(2): 578-590.
- Lima, W. D. P., Jarvis, P. and Rhizopoulou, S. 2003. Stomatal responses of *Eucalyptus* species to elevated CO₂ concentration and drought stress. *Sci. Agric.*, 60(2): 231-238.
- Lotfi, N., Vahdati, K., Hassani, D., Kholdebarin, B., and Amiri, R. 2009, February. Peroxidase, guaiacol peroxidase and ascorbate peroxidase activity accumulation in leaves and roots of walnut trees in response to drought stress. In *VI International Walnut Symposium* 861: 309-316.
- Lowry, O. H., Rosebrough, N. J., Farr, A. L., and Randall, R. J. 1951. Total protein estimation by Lowry's method. *J. Biol. Chem.* 193: 265.
- Luo, L. J. 2010. Breeding for water-saving and drought-resistance rice (WDR) in China. *J. Exp. Bot.*, 61(13): 3509-3517.
- Mafakheri, A., Siosemardeh, A., Bahramnejad, B., Struik, P. C., and Sohrabi, Y. 2010. Effect of drought stress on yield, proline and chlorophyll contents in three chickpea cultivars. *Aust. J. Crop Sci.*, 4(8): 580.

- Manivannan, P., Jaleel, C. A., Sankar, B., Kishorekumar, A., Somasundaram, R., Lakshmanan, G. A., and Panneerselvam, R. 2007. Growth, biochemical modifications and proline metabolism in *Helianthus annuus* L. as induced by drought stress. *Colloids Surf B Biointerfaces*, 59(2): 141-149.
- Mansfield, T. A. and Atkinson, C. J. 1990. Stomatal behaviour in water stressed plants. *Plant Biol.*, 12: 241-264.
- Mansour, M. M. F. 1998. Protection of plasma membrane of onion epidermal cells by glycinebetaine and proline against NaCl stress. *Plant Physiol. Biochem.*, 36(10): 767-772.
- Masia, A. 2003. Physiological effects of oxidative stress in relation to ethylene in postharvest produce. *Postharvest oxidative stress in horticultural crops*, pp.165-197.
- Matthews, J. D. 1961. *Report to the government of India on a program of forest genetics and forest tree breeding research*. Food and Agriculture Organization of the United Nations ETAF Report. No. 1349. Rome.
- Maxwell, K. and Johnson, G. N. 2000. Chlorophyll fluorescence-a practical guide. *J. Exp. Bot.*, 51(345): 659-668.
- Mc-Cue, K. F. and Hanson, A. D. 1990. Drought and salt tolerance: towards understanding and application. *Trends Biotechnol.*, 8: 358-362.
- Midgley, S. J., Stevens, P. R., and Arnold, R. J. 2017. Hidden assets: Asia's smallholder wood resources and their contribution to supply chains of commercial wood. *Aust. For.*, 80(1): 10-25.
- Mitchell, J. H., Siamhan, D., Wamala, M. H., Risimeri, J. B., Chinyamakobvu, E., Henderson, S. A. and Fukai, S. 1998. The use of seedling leaf death score for evaluation of drought resistance of rice. *Field Crops Res.*, 55(1-2): 129-139.

- Miyashita, K., Tanakamaru, S., Maitani, T., and Kimura, K. 2005. Recovery responses of photosynthesis, transpiration, and stomatal conductance in kidney bean following drought stress. *Environ. Exp. Bot.*, 53(2): 205-214.
- Mohan, M. M., Narayanan, S. L., and Ibrahim, S. M. 2000. Chlorophyll stability index (CSI): its impact on salt tolerance in rice. *International Rice Research Notes*, 25(2): 38-39.
- Moller, I. M., Jensen, P. E., and Hansson, A. 2007. Oxidative modifications to cellular components in plants. *Annu. Rev. Plant Biol.*, 58: 459-481.
- Moore, S. and Stein, W. H. 1948. Photometric nin-hydrin method for use in the chromatography of amino acids. *J. Biol. Chem.*, 176: 367-388.
- Moran, J. F., Becana, M., Iturbe-Ormaetxe, I., Frechilla, S., Klucas, R. V., and Aparicio-Tejo, P. 1994. Drought induces oxidative stress in pea plants. *Planta*, 194(3): 346-352.
- Morgan, J. M. 1984. Osmoregulation and water stress in higher plants. *Annu. Rev. Plant Physiol.*, 35(1): 299-319.
- Murata, N., Takahashi, S., Nishiyama, Y., and Allakhverdiev, S. I. 2007. Photoinhibition of photosystem II under environmental stress. *Biochim. et Biophys. Acta (BBA)-Bioenerg.*, 1767(6): 414-421.
- Muthuchelian, K., Krishna, R., Paliwal, K. and Gnanam, A. 1986. Physiological responses of a tree legume to leaf water deficits. *Physiological and biochemical and genetic aspects of crop plants in relation to environmental stress*, pp. 30-31.
- Muthuramu, S., Jebaraj, S., and Gnanasekaran, M. 2010. Association analysis for drought tolerance in rice (*Oryza sativa* L.). *Res. J. Agric. Sci.*, 1(4): 426-429.

- Nayyar, H. and Gupta, D. 2006. Differential sensitivity of C3 and C4 plants to water deficit stress: association with oxidative stress and antioxidants. *Environ. Exp. Bot.*, 58(1-3): 106-113.
- Nemeskeri, E., Molnar, K., Vigh, R., Nagy, J., and Dobos, A. 2015. Relationships between stomatal behaviour, spectral traits and water use and productivity of green peas (*Pisum sativum* L.) in dry seasons. *Acta physiol. plant.*, 37(2): 34.
- Neto, C. O., Rocha, M. E., Abade, M. T., Nogueira, G. A., Nascimento, V. R., Cardoso, K. P., Brito, A. E., Machado, L. C., Martins, J. T., Costa, T. C., and Okumura, R. S. 2018. Antioxidant enzymes activity in the *Elaeis guineensis* Jacq. submitted to drought. *J. Agric. Sci.*, 10(8): 245-252.
- Nicholas, J. C., Harper, J. E., and Hageman, R. H. 1976. Nitrate reductase activity in soybeans (*Glycine max* [L.] Merr.): I. effects of light and temperature. *Plant Physiol.*, 58(6): 731-735.
- O'Brien, M. J., Ong, R., and Reynolds, G. 2017. Intra-annual plasticity of growth mediates drought resilience over multiple years in tropical seedling communities. *Glob. Change Biol.*, 23(10): 4235-4244.
- Oliveira, M. T., Medeiros, C. D., Frosi, G., and Santos, M. G. 2014. Different mechanisms drive the performance of native and invasive woody species in response to leaf phosphorus supply during periods of drought stress and recovery. *Plant Physio. and Biochem.*, 82: 66-75.
- Ommen, O. E., Donnelly, A., Vanhoutvin, S., Van Oijen, M., and Manderscheid, R. 1999. Chlorophyll content of spring wheat flag leaves grown under elevated CO₂ concentrations and other environmental stresses within the 'ESPACE-wheat' project. *Eur. J. Agron.*, 10(3-4): 197-203.
- Orians, G. H. and Solbrig, O. T. 1977. A cost-income model of leaves and roots with special reference to arid and semiarid areas. *Am. Nat.*, 111(980): 677-690.

- Ozdemir, F., Bor, M., Demiral, T., and Türkan, İ. 2004. Effects of 24-epibrassinolide on seed germination, seedling growth, lipid peroxidation, proline content and antioxidative system of rice (*Oryza sativa* L.) under salinity stress. *Plant Growth Reg.*, 42(3): 203-211.
- Palma, J. M., Sandalio, L. M., Corpas, F. J., Romero-Puertas, M. C., Mc-Carthy, I., and Luis, A. 2002. Plant proteases, protein degradation, and oxidative stress: role of peroxisomes. *Plant Physiol. and Biochem.*, 40(6-8): 521-530.
- Pantuwan, G., Fukai, S., Cooper, M., Rajatasereekul, S., and O'toole, J.C. 2002. Yield response of rice (*Oryza sativa* L.) genotypes to drought under rainfed lowland: 3. Plant factors contributing to drought resistance. *Field Crops Res.*, 73(2-3): 181-200.
- Patade, V. Y., Bhargava, S., and Suprasanna, P. 2011. Salt and drought tolerance of sugarcane under iso-osmotic salt and water stress: growth, osmolytes accumulation, and antioxidant defense. *J. Plant Interact.*, 6(4): 275-282.
- Pattanagul, W. and Madore, M. A. 1999. Water deficit effects on raffinose family oligosaccharide metabolism in *Coleus*. *Plant Physiol.*, 121(3): 987-993.
- Pelah, D., Wang, W., Altman, A., Shoseyov, O., and Bartels, D. 1997. Differential accumulation of water stress-related proteins, sucrose synthase and soluble sugars in *Populus* species that differ in their water stress response. *Physiol. Plant.*, 99(1): 153-159.
- Peng, Z., Lu, Q. and Verma, D. P. S. 1996. Reciprocal regulation of Δ 1-pyrroline-5-carboxylate synthetase and proline dehydrogenase genes controls proline levels during and after osmotic stress in plants. *Mol. and Gen. Genet.*, 253(3): 334-341.
- Perez-Lopez, U., Robredo, A., Lacuesta, M., Mena-Petite, A., and Munoz-Rueda, A. 2012. Elevated CO₂ reduces stomatal and metabolic limitations on photosynthesis caused by salinity in *Hordeum vulgare*. *Photosynthesis Res.*, 111(3): 269-283.

- Perrone, I., Pagliarani, C., Lovisolo, C., Chitarra, W., Roman, F., and Schubert, A. 2012. Recovery from water stress affects grape leaf petiole transcriptome. *Planta*, 235(6): 1383-1396.
- Pierre, M. and Savoure, A. 1990. Effects of water-stress and SO₂ pollution on spruce endoproteases. *Plant Phys. Biochem. (Paris)*, 28(1): 95-104.
- Pinto-Marijuan, M. and Munne-Bosch, S. 2014. Photo-oxidative stress markers as a measure of abiotic stress-induced leaf senescence: advantages and limitations. *J. Exp. Bot.*, 65(14): 3845-3857.
- Plaut, Z. 1974. Nitrate reductase activity of wheat seedlings during exposure to and recovery from water stress and salinity. *Physiol. plant.*, 30(3): 212-217.
- Pompelli, M. F., Barata-Luís, R., Vitorino, H. S., Goncalves, E. R., Rolim, E. V., Santos, M. G., Almeida-Cortez, J. S., Ferreira, V. M., Lemos, E. E., and Endres, L. 2010. Photosynthesis, photoprotection and antioxidant activity of purging nut under drought deficit and recovery. *Biomass and Bioenerg.*, 34(8): 1207-1215.
- Premachandra, G. S., Saneoka, H., Fujita, K., and Ogata, S. 1992. Leaf water relations, osmotic adjustment, cell membrane stability, epicuticular wax load and growth as affected by increasing water deficits in sorghum. *J. Exp. Bot.*, 43(12): 1569-1576.
- Quirino, B. F., Noh, Y. S., Himelblau, E., and Amasino, R. M. 2000. Molecular aspects of leaf senescence. *Trends Plant Sci.*, 5(7): 278-282.
- Rajendrudu, G. and Naidu, C. V. 1997. Effects of water stress on leaf growth and photosynthetic and transpiration rates of *Tectona grandis*. *Biol. Plant.*, 40(2): 229-234.
- Ranney, T. G., Bassuk, N. L., and Whitlow, T. H. 1991. Osmotic adjustment and solute constituents in leaves and roots of water-stressed cherry (*Prunus*) trees. *J. Am. Soc. Hortic. Sci.*, 116(4): 684-688.

- Rao, K. M. and Sresty, T. V. S. 2000. Antioxidative parameters in the seedlings of pigeonpea (*Cajanus cajan* (L.) Millspaugh) in response to Zn and Ni stresses. *Plant Sci.*, 157(1): 113-128.
- Rauf, S., Al-Khayri, J. M., Zaharieva, M., Monneveux, P., and Khalil, F. 2016. Breeding strategies to enhance drought tolerance in crops. In *Advances in plant breeding strategies: agronomic, abiotic and biotic stress traits*, Springer, Cham pp. 397-445.
- Rawat, K. and Bakshi, M. 2011. Provenance variation in cone, seed and seedling characteristics in natural populations of *Pinus wallichiana* AB Jacks (Blue Pine) in India. *Ann. For. Res.*, 54(1): 39-55.
- Ren, J., Sun, L. N., Zhang, Q. Y., and Song, X. S. 2016. Drought tolerance is correlated with the activity of antioxidant enzymes in *Cerasus humilis* seedlings. *BioMed research international*, 2016.
- Rhodes, D. and Hanson, A. D. 1993. Quaternary ammonium and tertiary sulfonium compounds in higher plants. *Ann. Rev. Plant Biol.*, 44(1): 357-384.
- Rivas, R., Falcao, H. M., Ribeiro, R. V., Machado, E. C., Pimentel, C. and Santos, M. G. 2016. Drought tolerance in cowpea species is driven by less sensitivity of leaf gas exchange to water deficit and rapid recovery of photosynthesis after rehydration. *South African J. Bot.*, 103: 101-107.
- Rivas, R., Oliveira, M. T., and Santos, M. G. 2013. Three cycles of water deficit from seed to young plants of *Moringa oleifera* woody species improves stress tolerance. *Plant Physiol. Biochem.*, 63: 200-208.
- Rodrigues, B. M., Souza, B. D., Nogueira, R. M., and Santos, M. G. 2010. Tolerance to water deficit in young trees of jackfruit and sugar apple. *Riv. Ciênc. Agron.*, 41(2): 245-252.

- Roldan, A., Diaz-Vivancos, P., Hernandez, J. A., Carrasco, L., and Caravaca, F. 2008. Superoxide dismutase and total peroxidase activities in relation to drought recovery performance of mycorrhizal shrub seedlings grown in an amended semiarid soil. *J. Plant Physiol.*, 165(7): 715-722.
- Rood, S. B., Patino, S., Coombs, K., and Tyree, M. T. 2000. Branch sacrifice: cavitation-associated drought adaptation of riparian cottonwoods. *Trees*, 14(5): 248-257.
- Roy-Macauley, H., Zuily-Fodil, Y., Kidric, M., Thi, A. P., and de Silva, J. V. 1992. Effect of drought stress on proteolytic activities in *Phaseolus* and *Vigna* leaves from sensitive and resistant plants. *Physiol. Plant.*, 85(1): 90-96.
- Ruehr, N. K., Grote, R., Mayr, S., and Arneth, A. 2019. Beyond the extreme: recovery of carbon and water relations in woody plants following heat and drought stress. *Tree Physiol.*, 39(8): 1285-1299.
- Ruehr, N. K., Offermann, C. A., Gessler, A., Winkler, J. B., Ferrio, J. P., Buchmann, N. and Barnard, R. L. 2009. Drought effects on allocation of recent carbon: from beech leaves to soil CO₂ efflux. *New Phytol.*, 184(4): 950-961.
- Sairam, R. K., Deshmukh, P. S., and Shukla, D. S. 1997. Tolerance of drought and temperature stress in relation to increased antioxidant enzyme activity in wheat. *J. Agron. Crop Sci.*, 178(3): 171-178.
- Sakamoto, A. and Murata, N. 2000. Genetic engineering of glycinebetaine synthesis in plants: current status and implications for enhancement of stress tolerance. *J. Exp. Bot.*, 51(342): 81-88.
- Santiago, L. S. and Wright, S. J. 2007. Leaf functional traits of tropical forest plants in relation to growth form. *Funct. Ecol.*, 21(1): 19-27.

- Saxe, H., Cannell, M. G., Johnsen, O., Ryan, M. G. and Vourlitis, G. 2001. Tree and forest functioning in response to global warming. *New phytol.*, 149(3): 369-399.
- Schultz, H. R. and Matthews, M. A. 1993. Growth, osmotic adjustment, and cell-wall mechanics of expanding grape leaves during water deficits. *Crop Sci.*, 33(2): 287-294.
- Schwalm, C. R., Anderegg, W. R., Michalak, A. M., Fisher, J. B., Biondi, F., Koch, G., Litvak, M., Ogle, K., Shaw, J. D., Wolf, A., and Huntzinger, D. N. 2017. Global patterns of drought recovery. *Nature*, 548(7666): 202.
- Seel, W.E., Hendry, G.A.F. and Lee, J.A. 1992. The combined effects of desiccation and irradiance on mosses from xeric and hydric habitats. *J. Exp. Bot.*, 43(8): 1023-1030.
- Seidel, H., Schunk, C., Matiu, M., and Menzel, A. 2016. Diverging drought resistance of scots pine provenances revealed by infrared thermography. *Front. Plant Sci.*, 7:1247.
- Seiler, J. R. and Johnson, J. D. 1985. Photosynthesis and transpiration of loblolly pine seedlings as influenced by moisture-stress conditioning. *For. Sci.*, 31(3): 742-749.
- Sen Gupta, I. N. 1939. Summary of results of data of the All Indian Co-operative Teak Seed Origin Investigation. Paper 11. Item. 4. Proc. 5th Silv. Conf., Dehra Dun, pp.109-115.
- Seth, S. K. and Khan, W. M. A. 1958. Regeneration of teak forests. Dhra Dun, India, Recommendations of teal study tour and Symposium, December 1957-January 1958. *Forest Research Institute*.
- Shaner, D. L. and Boyer, J. S. 1976. Nitrate Reductase Activity in Maize (*Zea mays* L.) Leaves: II. Regulation by Nitrate Flux at Low Leaf Water Potential. *Plant Physiol.*, 58(4): 505-509.

- Shangguan, Z., Shao, M., and Dyckmans, J. 1999. Interaction of osmotic adjustment and photosynthesis in winter wheat under soil drought. *J. Plant Physiol.*, 154(5-6): 753-758.
- Shao, H.B., Chu, L.Y., Shao, M.A., Jaleel, C.A., and Hong-mei, M. 2008. Higher plant antioxidants and redox signaling under environmental stresses. *Comptes Rendus biol.*, 331(6): 433-441.
- Shekafandeh, A., Assar, P., and Taghipour, L. 2019. Cell membrane stability and water relations of different scions grafted onto Mexican lime under water stress conditions and recovery. *Plant Physiol. Rep.*, 24(2): 182-191.
- Sheokand, S., Kumari, A., and Sawhney, V. 2008. Effect of nitric oxide and putrescine on antioxidative responses under NaCl stress in chickpea plants. *Physiol. Mol. Biol. Plants*, 14(4): 355-362.
- Siddique, M. R. B., Hamid, A. I. M. S., and Islam, M.S. 2000. Drought stress effects on water relations of wheat. *Bot. Bull. Acad. Sin.*, 41
- Silva, F. L., Pedrozo, C. A., Barbosa, M. H. P., Resende, M. D. V., Peternelli, L. A., de Almeida Costa, P. M., and Vieira, M. S. 2009. Analise de trilha para os componentes de produção de cana-de-açúcar via blup. *Riv. Ceres*, 56(3): 308-314.
- Sircelj, H., Tausz, M., Grill, D. and Batic, F. 2005. Biochemical responses in leaves of two apple tree cultivars subjected to progressing drought. *J. Plant Physiol.*, 162(12): 1308-1318.
- Sircelj, H., Tausz, M., Grill, D. and Batic, F. 2007. Detecting different levels of drought stress in apple trees (*Malus domestica* Borkh.) with selected biochemical and physiological parameters. *Sci. Hortic.*, 113(4): 362-369.

- Sivakumar, R., Nandhitha, G. K., and Nithila, S. 2017. Impact of drought on chlorophyll, soluble protein, abscisic acid, yield and quality characters of contrasting genotypes of tomato (*Solanum lycopersicum*). *Curr. J. App. Sci. Technol.*, pp. 1-10.
- Sivaramakrishnan, S., Patell, V.Z., Flower, D. J., and Peacock, J. M. 1988. Proline accumulation and nitrate reductase activity in contrasting sorghum lines during mid-season drought stress. *Physiol. Plant.*, 74(3): 418-426.
- Smirnoff, N. 1993. The role of active oxygen in the response of plants to water deficit and desiccation. *New phytol.*, 125(1): 27-58.
- Sneha, C., Santoshkumar, A. V., and Sunil, K. M. 2017. Detection of Water Stress in Teak (*Tectona grandis*) Seedlings Using Canopy-Air Temperature Difference. *Indian For.*, 143(7): 665-670.
- Sofo, A., Dichio, B., Xiloyannis, C., and Masia, A. 2004. Lipoxygenase activity and proline accumulation in leaves and roots of olive trees in response to drought stress. *Physiol. plant.*, 121(1): 58-65.
- Sofo, A., Manfreda, S., Fiorentino, M., Dichio, B., and Xiloyannis, C. 2008. The olive tree: a paradigm for drought tolerance in Mediterranean climates. *Hydrol.. Earth Syst. Sci. Discussions*, 12(1): 293-301.
- Souza, R. P., Machado, E. C., Silva, J. A. B., Lagôa, A. M. M. A., and Silveira, J. A. G. 2004. Photosynthetic gas exchange, chlorophyll fluorescence and some associated metabolic changes in cowpea (*Vigna unguiculata*) during water stress and recovery. *Environ. Exp. Bot.*, 51(1): 45-56.
- Sreekanth, P.M. and Balasundaran, M., 2013. Clonal seed orchard of teak (*Tectona grandis* Lf): genetic diversity measures primary basis for future environmental uncertainty. *Tree Genet. Mol. Breed.*, 3(5): 4-11.
- Srivastava, H. S. 1980. Regulation of nitrate reductase activity in higher plants. *Phytochemistry*, 19(5): 725-733.

- Stewart, C. R. 1971. Effect of wilting on carbohydrates during incubation of excised bean leaves in the dark. *Plant Physiol.*, 48(6): 792-794.
- Stockle, C. O. and Dugas, W. A. 1992. Evaluating canopy temperature-based indices for irrigation scheduling. *Irrig. Sci.*, 13(1): 31-37.
- Subramanian, K., Mandal, A. K., Rambabu, N., Chundamannil, M., and Nagarajan, B. 2000. Site, technology and productivity of teak plantations in India. *Site, technology and productivity of teak plantations. FORSPA Publication*, (24): 51-68.
- Sundaravalli, V. M., Paliwal, K., and Ruckmani, A. 2005. Effect of water stress on photosynthesis, protein content and nitrate reductase activity of *Albizzia* seedlings. *J. Plant Biol.*, 32(1): 13.
- Surendran, T. 2014. *Studies on clonal propagation of plus trees of teak for identifying superior trees for future plantation programmes*. KFRI [Kerala Forest Research Institute] Research Report No. 484, Peechi, India, 23 p.
- Taiz, L. and E. Zeiger. 2002. *Plant Physiology*. (3rd Ed) Sinauer Associates Inc. Publishers, Massachusetts, USA. 787 p.
- Takahashi, S. and Murata, N. 2008. How do environmental stresses accelerate photoinhibition?. *Trends Plant Sci.*, 13(4): 178-182.
- Todd, G. W. and Basler, E. 1965. Fate of various protoplasmic constituents in droughted wheat plants. *Phyton.*, 22(1): 79-85.
- Torres-Ruiz, J. M., Diaz-Espejo, A., Morales-Sillero, A., Martín-Palomo, M. J., Mayr, S., Beikircher, B., and Fernández, J. E. 2013. Shoot hydraulic characteristics, plant water status and stomatal response in olive trees under different soil water conditions. *Plant Soil*, 373(1-2): 77-87.
- Vakshasya, R. K., Uniyal, D. P., and Rawat, M. S. 1988. Teak plus trees for specific traits. *Indian For.*, 114(3): 168-169.

- Vankova, R., Dobra, J., and Storchova, H. 2012. Recovery from drought stress in tobacco: an active process associated with the reversal of senescence in some plant parts and the sacrifice of others. *Plant Signal. Behav.*, 7(1): 19-21.
- Vasav, V. P., Narkhede, S. S., Rane, A. D., Gunaga, R. P., Rewale, A. P., and Bhave, S. G. 2011. Evaluation of progenies of candidate plus trees of *Pongamia pinnata* (L.) Pierre. for seed germination and seedling vigour. *Curr. Sci.*, 100(4): 465-467.
- Verbruggen, N., Hua, X. J., May, M., and Van Montagu, M. 1996. Environmental and developmental signals modulate proline homeostasis: evidence for a negative transcriptional regulator. *Proc. Natl. Acad. Sci. U.S.A.*, 93(16): 8787-8791.
- Vierstra, R. D. 1993. Protein degradation in plants. *Annu. Rev. Plant boil.*, 44(1): 385-410.
- Wang, L. F. 2014. Physiological and molecular responses to drought stress in rubber tree (*Hevea brasiliensis* Muell. Arg.). *Plant Physiol. and Biochem.*, 83: 243-249.
- Wang, Z., Li, G., Sun, H., Ma, L., Guo, Y., Zhao, Z., Gao, H., and Mei, L. 2018. Effects of drought stress on photosynthesis and photosynthetic electron transport chain in young apple tree leaves. *Biol. Open*, 7(11): 035279.
- Wang, Z., Quebedeaux, B., and Stutte, G.W. 1995. Osmotic adjustment: effect of water stress on carbohydrates in leaves, stems and roots of apple. *Funct. Plant Biol.*, 22(5): 747-754.
- Wassmann, R., Jagadish, S. V. K., Sumfleth, K., Pathak, H., Howell, G., Ismail, A., Serraj, R., Redona, E., Singh, R. K., and Heuer, S. 2009. Regional vulnerability of climate change impacts on Asian rice production and scope for adaptation. *Adv. Agron.*, 102: 91-133.

- Wei, P., Yang, Y., Wang, F., and Chen, H. 2015. Effects of drought stress on the antioxidant systems in three species of *Diospyros* L. *Hortic. Environ. Biotechnol.*, 56(5): 597-605.
- Weimberg, R., Lerner, H. R., and Poljakoff-Mayber, A. 1984. Changes in growth and water-soluble solute concentrations in *Sorghum bicolor* stressed with sodium and potassium salts. *Physiol. Plant.*, 62(3): 472-480.
- White, D. A., Turner, N. C., and Galbraith, J. H. 2000. Leaf water relations and stomatal behavior of four allopatric *Eucalyptus* species planted in Mediterranean southwestern Australia. *Tree Physiol.* 20(17): 1157-1165.
- White, K. J. 1993. Research results: a selection of annotated references of teak (*Tectona grandis* Lf). Forestry Research Support Programme for Asia and Pacific *FORSPA Occasional Papers (FAO)*, No. 19, 22 p.
- Wright J. W. 1976. *Introduction to Forest Genetics*. Academic Press, New York, 463 p.
- Wyn-Jones, R. G. and Storey, R. 1978. Salt stress and comparative physiology in the Gramineae, 2. Glycinebetaine and proline accumulation in two salt-and water-stressed barley cultivars. *Aust. J. Plant Physiol.*, 5(6): 817-829.
- Xiao, S. H. U., Xu, Y. A. N. G., and Zhiling, Y. A. N. G. 2012. Variation in seed and seedling traits among fifteen Chinese provenances of *Magnolia officinalis*. *Not. Bot. Hort. Agrobot.*, 40(2): 274-283.
- Xiao, X., Yang, F., Zhang, S., Korpelainen, H., and Li, C. 2009. Physiological and proteomic responses of two contrasting *Populus cathayana* populations to drought stress. *Physiol. plant.*, 136(2): 150-168.
- Xing, W. and Rajashekar, C. B. 1999. Alleviation of water stress in beans by exogenous glycine betaine. *Plant Sci.*, 148(2): 185-192.

- Xu, Z. Z. and Yu, Z. W. 2006. Nitrogen metabolism in flag leaf and grain of wheat in response to irrigation regimes. *J. Plant Nutr. Soil Sci.*, 169(1): 118-126.
- Xu, Z., Zhou, G., and Shimizu, H. 2010. Plant responses to drought and rewatering. *Plant Signal. Behav.*, 5(6): 649-654.
- Yadav, S. K., Lakshmi, N. J., Maheswari, M., Vanaja, M., and Venkateswarlu, B. 2005. Influence of water deficit at vegetative, anthesis and grain filling stages on water relation and grain yield in sorghum. *Indian J. Plant Physiol.*, 10(1): 20.
- Yamada, M., Morishita, H., Urano, K., Shiozaki, N., Yamaguchi-Shinozaki, K., Shinozaki, K., and Yoshida, Y. 2005. Effects of free proline accumulation in petunias under drought stress. *J. Exp. Bot.*, 56(417): 1975-1981.
- Yang, F. and Miao, L. F. 2010. Adaptive responses to progressive drought stress in two poplar species originating from different altitudes. *Silva Fenn.*, 44(1): 23-37.
- Yang, W. J., Rich, P. J., Axtell, J. D., Wood, K. V., Bonham, C. C., Ejeta, G., Mickelbart, M. V., and Rhodes, D. 2003. Genotypic variation for glycinebetaine in sorghum. *Crop Sci.*, 43(1): 162-169.
- Yi, X. P., Zhang, Y. L., Yao, H. S., Luo, H. H., Gou, L., Chow, W. S., and Zhang, W. F. 2016. Rapid recovery of photosynthetic rate following soil water deficit and re-watering in cotton plants (*Gossypium herbaceum* L.) is related to the stability of the photosystems. *J. Plant Physiol.* 194: 23-34.
- Yin, C., Peng, Y., Zang, R., Zhu, Y., and Li, C. 2005 a. Adaptive responses of *Populus kangdingensis* to drought stress. *Physiol. Plant.*, 123(4): 445-451.
- Yin, C., Wang, X., Duan, B., Luo, J., and Li, C. 2005 b. Early growth, dry matter allocation and water use efficiency of two sympatric *Populus* species as affected by water stress. *Environ. Exp. Bot.*, 53(3): 315-322.

- Yordanov, I., Velikova, V., and Tsonev, T. 2000. Plant responses to drought, acclimation, and stress tolerance. *Photosynthetica*, 38(2): 171-186.
- Yu, T. F., Xu, Z. S., Guo, J. K., Wang, Y. X., Abernathy, B., Fu, J. D., Chen, X., Zhou, Y. B., Chen, M., Ye, X. G., and Ma, Y. Z. 2017. Improved drought tolerance in wheat plants overexpressing a synthetic bacterial cold shock protein gene SeCspA. *Sci. Rep.*, 7: 44050.
- Zhang, J. and Davies, W. J. 1987. Increased synthesis of ABA in partially dehydrated root tips and ABA transport from roots to leaves. *J. Exp. Bot.*, 38(12): 2015-2023.
- Zhang, S., Chen, L., Duan, B., Korpelainen, H., and Li, C. 2012. *Populus cathayana* males exhibit more efficient protective mechanisms than females under drought stress. *For. Ecol. Manag.*, 275: 68-78.
- Zhang, X., Wu, N., and Li, C. 2005. Physiological and growth responses of *Populus davidiana* ecotypes to different soil water contents. *J. Arid Environ.*, 60(4): 567-579.
- Zhang, Y., Chen, Q., Lan, J., Luo, Y., Wang, X., Chen, Q., Sun, B., Wang, Y., Gong, R., and Tang, H. 2018. Effects of drought stress and rehydration on physiological parameters and proline metabolism in kiwi fruit seedling. *Int. J. Agric. Biol.*, 20: 2891-2896.
- Zhou, S., Duursma, R. A., Medlyn, B. E., Kelly, J. W., and Prentice, I. C. 2013. How should we model plant responses to drought? An analysis of stomatal and non-stomatal responses to water stress. *Agric. For. Meteorol.*, 182: 204-214.
- Zhu, X., Gong, H., Chen, G., Wang, S., and Zhang, C. 2005. Different solute levels in two spring wheat cultivars induced by progressive field water stress at different developmental stages. *J. Arid Environ.*, 62(1): 1-14.

- Zobel, B. and Talbert, J. 1984. *Applied forest tree improvement*. John Wiley & Sons, New York, USA. 505p.
- Zou, G. H., Liu, H. Y., Mei, H. W., Liu, G. L., Yu, X. Q., Li, M. S., Wu, J. H., Chen, L., and Luo, L. J. 2007. Screening for drought resistance of rice recombinant inbred populations in the field. *J. Int. Pl. Bio.* 49: 1508-1516.
- Zu, X., Lu, Y., Wang, Q., Chu, P., Miao, W., Wang, H., and La, H. 2017. A new method for evaluating the drought tolerance of upland rice cultivars. *Crop J.*, 5(6): 488-498.

**DROUGHT RESPONNS IN PLUS TREES OF TEAK (*Tectona grandis* Linn. f.)
PROVENANCES OF KERALA**

By

MOHAMMED ALI ELTOUM HASSAN

(2017-17-014)

ABSTRACT OF THE THESIS

**Submitted in partial fulfilment of the
requirement for the degree of**

MASTER OF SCIENCE IN FORESTRY

Faculty of Forestry

Kerala Agricultural University



DEPARTMENT OF FOREST BIOLOGY AND TREE IMPROVEMENT

COLLEGE OF FORESTRY

VELLANIKKARA, THRISSUR- 680 656

KERALA, INDIA

2020

ABSTRACT

A study on 'Drought response in plus trees of teak (*Tectona grandis* Linn. f.) provenances of Kerala' was conducted in College of Forestry, Vellanikkara during 2018-2019 to evaluate variability in seedling biometric, physiological and biochemical characters of plant during drought stress and drought recovery. Drought stress was induced by withholding water till leaves exhibited symptoms of wilting. Plantlets were exposed to two consecutive drought treatments with an intervening period of stress relief through rehydration.

Seedling vigor was screened at six months. No variability was present in morphometric characters like height, diameter, number of leaves and branches. After inducing drought stress, growth parameters slowed down in all accessions between plus trees. Induction of drought resulted in reduction in most of the physiological parameters of the plantlets of teak plus tree accessions. Relative water content, photosynthetic rate, stomatal conductance, transpiration rate, chlorophyll a, chlorophyll b, total chlorophyll and chlorophyll stability index showed reduction. However, under drought stress, plantlets showed increased of canopy air temperature differences and cell membrane stability index. Most of biochemical parameters like free amino acid, proline, glycine betaine, total soluble sugar, super oxide dismutase, peroxidase and malondialdehyde increased after inducing drought. However, total soluble protein and nitrate decreased under drought induction. After rehydration, most of these physiological and biochemical parameters rapidly returned to the level of normal irrigated condition.

The stomatal conductance was only differed in the accessions before stress was induced. However, after stress, it was found that accessions differed in stomatal conductance, photosynthetic pigments and total chlorophyll content. In drought recovery, there was no variation in physiological parameters among accessions. Variability on biochemical characters were observed in nitrate reductase, free amino acid, proline and peroxidase among accessions during pre-stress stage, while in drought stress it was observed that proline, super oxide dismutase, peroxidase and

malondialdehyde differed among accession. In drought recovery, characters such as nitrate reductase, free amino acid, proline and peroxidase showed variability.

Correlation studies showed that among the physiological and biochemical character only chlorophyll a, chlorophyll b and total chlorophyll showed positive correlation with the number of leaves during drought stress, while in recovery, positive correlation was shown by super oxide dismutase, chlorophyll a, chlorophyll b and total chlorophyll content and negative correlation was observed in photosynthetic rate, transpiration, relative water content and stomatal conductance.

Hierarchical cluster analysis was done for the eleven accessions based on the Euclidian squared distance. During drought stress, the accessions grouped into five clusters; cluster III possesses four of accessions whereas the least number observed for the cluster V. In recovery, it was grouped into seven clusters; cluster VII had three accessions, while only one accession was present in cluster I, IV, V and VI.

First two components of principle component analysis during drought stress accounted for 41.9 per cent of the total variability, which was mainly contributed positively by relative water content, nitrate reductase, free amino acid, proline and malondialdehyde. In drought recovery, first two components of PCA together accounted for 53.2 per cent of the total variability, which was mainly contributed positively by photosynthetic rate, stomatal conductance, transpiration rate, chlorophyll stability index, glycine betaine and malondiadehyde.

Selection index were worked out to select accession tolerant to drought stress and recovery based on biochemical parameter and chlorophyll content using first principle component as index. It was found that KFRI T55 was most tolerant and quickest to recover after reliving stress among accession.

It can be concluded from the results that physiological and biochemical variations exist in teak plus tree accessions. These results could be useful in selection of drought tolerant