

**STUDIES ON PLUS TREE SELECTION, VARIABILITY
AND SEED BIOLOGY OF TERMINALIA PANICULATA
ROTH (COMBRETACEAE) IN KERALA PART OF
PENINSULAR INDIA**



**Thesis submitted to the University of Calicut in partial
Fulfillment of the requirements for the degree of
Doctor of Philosophy in Botany**

by
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Under the guidance of
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November 2022




CERTIFICATE

This is to certify that the thesis entitled “**Studies on Plus Tree Selection, Variability and Seed Biology of *Terminalia paniculata* Roth (Combretaceae) In Kerala Part of Peninsular India**” submitted to the University of Calicut for the award of degree of Doctor of Philosophy in Botany by Mr. Sanal C Viswanath is the result of bonafide research work carried out by him under my guidance in the Department of Forest Genetics and Tree Breeding, KSCSTE Kerala Forest Research Institute, Peechi. Further, I certify that this or part thereof has not been the basis for the award of any other diploma or degree either in any institution in any institution or university.

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DECLARATION

I, Sanal C Viswanath hereby declare that the thesis entitled “**Studies on Plus Tree Selection, Variability and Seed Biology of *Terminalia paniculata* Roth (Combretaceae) In Kerala Part of Peninsular India**” embodies the results of bonafide research work done by me under the guidance of Dr. Hrideek T K, Senior Scientist, Department of Forest Genetics and Tree Breeding, KSCSTE- Kerala Forest Research Institute, Peechi. I further declare that this or part thereof has not been the basis for the award of any other diploma or degree either in any institution or university.



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Date: 01-11-2022

Place: Kozhikode

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Abbreviations

a.s.l	Above sea level	MC	Moisture content
BT	Bark thickness	NAA	Naphthalene acetic acid
CD	Crown diameter	PB	Primary branches
CL	Crown length	PC	Principle component
CPT	Candidate plus tree	PCA	Principle component analysis
CV	Coefficient of variation	PT	Plus tree
DAS	Days after sowing	PTSP	Plus tree selection program
ESR	Rooted epicormic shoots	R	Correlation coefficient
EST	Treated epicormic shoots	RP	Rooting percentage
FD	Fruit dry mass	SB	Secondary branches
FF	Fruit fresh mass	SD	Sapwood density
FR	Forest range	SD	Standard deviation
GD	Germination duration	SE	Standard error
GH	Girth at breast height	SE	Empty seeds
GP	Germination percentage	SG	Germinated seeds
HT	Total tree height	SL	Small wings length
IBA	Indole 3 butyric acid	SM	Sapwood moisture content
KFRI	Kerala Forest Research Institute	ST	Stress wave time
KFSC	Kerala Forest Seed Centre	STD	Standard
LA	Leaf area	SV	Viable seeds
LC	Leaf chlorophyll content	TP	<i>Terminalia paniculata</i>
LD	Leaf dry mass	TPP	<i>Terminalia paniculata</i> population
LF	Leaf fresh mass	WD	Wildlife division
LL	Large wing length	WS	Water soaking
LW	Large wing width	X	Mean

CHAPTER 1
INTRODUCTION

Chapter 1

Introduction

Tree improvement is a dynamic technology drawing upon a large, escalating body of scientific knowledge, technical methodology practical experiences. Tree improvement, or as it is often referred to as genetic improvement, is the process of enlightening the genetic quality of a tree species. It is the implementation of forest genetic principles within a given silvicultural system to enhance the genetic quality of the forest. Its goal is to improve the genetic value of the population while maintaining genetic diversity. Meeting this goal means that genetic improvement is intended at the population level rather than the improvement of breeds or inbred lines. Tree improvement programs deliver a known seed source, seedlings, or propagules for forest establishment. Worldwide, tree improvement programs are linked to a range of silvicultural systems, but they are most commonly unified with plantation silviculture (Zobel and Talbert 1984).

Each tree improvement program must be planned to fit the species' life history and natural range and the organization's planting schedule an annual budget harvest goals. For example, the least intensive tree improvement programs deliver a known seed source for a specific period. More specific tree improvement programs typically have sufficient profit incentives to invest in a full-scale breeding program for generations to come. Tree improvement as the application of genetic principles to silviculture is considered a 20th-century idea. Still, few realize that the idea of planting trees is ancient, harking back as far as the origins of Neolithic agriculture. The first accounts for grown tree farms date to the Ptolemy kingdom in Egypt circa the third century B.C. (Perlin 1989).

Starting with Bradley in England, they listed some thirty outstanding persons who had worked from 1717 to 1935 on different components of tree improvement such

as seed sources, inheritance, hybridization of species, vegetative propagation, reproductive biology; the establishment of seed production stands orchards a few specific characteristics such as stem straightness wood characteristics. Work on provenance variation self-fertilization of larch was initiated at the Institute of Forest Genetics in Sweden in 1902. The Eddy Tree Breeding Station was established mainly conducting pine hybridization in Placerville, California, in 1924. Succeeding World War II, systematic tree breeding programs based on a good understanding of genetic principles originated in Europe, Japan North America.

In Europe, the species were both indigenous exotic while in Japan; work was focused initially on the indigenous *Cryptomeria japonica*. In both Canada and the USA, attention was concentrated on native *Pseudotsuga*, *Pinus* and *Picea*, with some work on timber nut-bearing hardwood species. In the 1960s, tree improvement programs were started in Australia (federal state-oriented), New Zealand (Forest Research Institute), South Africa (De Wet Tree Breeding Station), and Zimbabwe (Forestry Commission); these have become exceptional examples of highly planned, efficiently conducted economically practical tree breeding. Subsequently, many other countries in the British Commonwealth started tree breeding programs with the initial guidance of the Commonwealth Forestry Institute (=Oxford Forestry Institute). Francophone countries in Africa and the Pacific region were assisted in breeding programs by the Centre Technique Forestier Tropical France.

The intentions of a tree breeding program range from yield improvement adaptation to particular conditions to pest- disease-resistance for better wood properties, etc. Currently, tree breeding is starting to take advantage of the rapid development in plant genetics genomics. The scientific principles and technical methods of tree improvement have been thoroughly illustrated (Wright 1962; 1976; Faulkner 1975; Namkoong 1979) and exhaustively reviewed (Shelbourne 1969; Van Buijtenen et al. 1971; Morgenstern 1974; Morgenstern et al. 1975; Namkoong et al. 1980; Morgenstern

1983; Morgenstern and Mullin 1987; Ondro et al. 1995; Paques 2013). Plantation tree growth rates of 25 m³per ha per yr are harvested at 6-years. With the genetic improvement of tree species tree breeding, average growth rates of 35 m³per ha per yr are expected (Kellison 2005). The average productivity of poplar under an agroforestry system is 20-25 m³/ha/yr, which is five times overhead than traditional forests throughout Punjab, Haryana, Uttar Pradesh, and some parts of Bihar, West Bengal, Assam state (Singh et al. 2001).

Tree breeding helps in the genetic adjustment of plants to the service of humans and capitalizes on the natural variability packages of the best traits (Chuntanaparb and Ranganathan 1981). Tree breeding comprises packaging the desired features into improved individuals, large-scale production of improved individuals for planting purposes developing and maintaining a genetic base population for an advanced generation. It helps increase yields, and shortened rotations, so planted forests become increasingly attractive as an investment for producing industrial wood. It can ensure supplying of fuel wood and meet other needs of rural people by introducing fast-growing multipurpose tree species for farmland planting (Shea and Carlson 1984).

Two discrete methods of tree improvement are generally used: provenance studies and tree breeding programs. Provenance studies are performed to identify the best wild populations of the selected tree species. Tree breeding programs are conducted to choose breeds from the best individuals within the best populations. After selecting suitable species, the most appropriate seed source for the planting site must be determined. This is accomplished by choosing suitable provenances. Provenance is the area on which any stand of the tree is growing. The frame may be indigenous or non-indigenous. During the early-mid-1900s, wide-range genetic surveys (i.e., provenance testing) were emphasized using "common garden" experimental principles. Ecologists' early standard garden experiments in the 1940s (earlier) served as the initial models for these investigations in forestry ecology research.

The process was relatively simple in concept. Samples are collected from natural populations of interest (i.e., provenances) and then planted in several test environments (usually within the range of the provenance collections) to test the performance of the local provenance versus others. These early studies showed that most tree species had large amounts of genetic variation within populations, but local populations were not always the best. However, in general, forest trees show patterns of adaptation that are reasonably well associated with the climate from which they originated. More importantly, there are differences among species in the general level of transformation.

Tree breeding is an essential component of tree improvement which involves the application of genetic principles for the large-scale production of seedlings with desired traits to accomplish better adaptability to the environment and higher productivity and vigorous growth rate. Early tree breeding was a short-term activity aiming to achieve the immediate need for seeds acquiring rapid genetic gains by establishing seed orchards with selected trees. More ambitious plans arose to lead the tree breeding further to advanced generations when these goals were achieved. This led to conceptual differences between the continuous breeding cycle multiplication populations (e.g., seed orchard), which aim for short-term genetic gain (Burdon and Shelbourne 1972; van Buijtenen 1975).

The current long-term breeding plans thus combine the maintenance of sufficient genetic variation essential for long-term genetic gain and high immediate gain of short-term actions, which justifies the breeding work economically (Gullberg and Kang 1985). A long-term tree breeding program consists of three steps: selection, and crossing testing, repeated in successive generations. The repetition of these phases, together with the connected populations, forms the breeding cycle, which functions to enrich the favourable alleles (Venalainen and Ruotsalainen 2002). Crossing, or more generally sexual reproduction, shuffles the trees' genes. Old gene combinations are broken into new ones, and more favourable ones are created, later to be sieved by selection testing.

Tree improvement most often has counted on traditional breeding techniques like selecting superior (plus candidate) trees for volume stem straightness, grafting these into breeding orchards producing seed orchards. When breeding orchards begin to flower, pollination of selections is artificially controlled, seeds are collected, and progeny tests are established. The best offspring are chosen for the next cycle of breeding. At the same time, selections whose offspring did not perform well in the progeny tests are removed from the production seed orchards to improve genetic quality (Tridasa et al. 1996). After selecting suitable species, the most appropriate seed source for the planting site must be chosen (Mborra et al. 2009). This is accomplished by selecting suitable provenances. Provenance is the area on which any stand of the tree is growing. The stand may be indigenous or exotic. A strong differentiation of adaptive traits can be found among tree populations growing under different ecological conditions. Such variabilities are characteristics of provenances that may be expressed by various features on the genetic, morphological, and phenotypic phenological levels, which can be obtained by submitting provenances to specific testing procedures.

Tree improvement of temperate hardwoods has been more limited than that of coniferous species hardwoods of the genus *Populus* and *Eucalyptus* (Merkle and Nairn 2005). Considerable effort has been exerted over the last 40-years in conventional tree improvement programs through breeding selection strategies for tree improvement of temperate hardwoods (Burley and Kanowski 2005). The long generation reproductive cycle, difficulty in conducting controlled pollinations, and intermittent or scarce seed crops seed recalcitrance of hardwood trees are some of the limitations imposed on conventional tree breeding programs.

People fascinated by the incredible diversity of tropical forests will be surprised to learn that robust estimates of the number of tropical trees species are lacking. We show that there are at least 40000, but possibly more than 53000, tree species in the tropics, in contrast to only 124 across temperate Europe. Almost all tropical tree species are

restricted to their respective continents. The Indo-Pacific region appears to be as species-rich as tropical America. These two regions are nearly five times richer in tree species than African tropical forests. Tropical hardwoods are predominantly found around the equator between the Tropic of Cancer, forming a belt of green life. Geographically, tropical forests are located along the southeast coast of Brazil, the Guinea coast, and the Zaire basin in eastern Madagascar in Africa. They are found in the Himalayas (north-south slope), south-eastern parts of India, Malaysia, Indonesia, Thailand, and New Guinea in Asia. They can also be seen in Australia as a narrow, discontinuous strip along the eastern coast. For instance, in Amazonian Ecuador, tree richness (diameter > 10 cm) was 473 per hectare.

Unfortunately, tropical forests are under intense use pressure. They are rapidly converted into agricultural lands or pastures. Industrialization, deforestation, changes in cultivation practices several other human activities have further enhanced the rates of loss of tropical hardwoods. They are being overexploited for wood and non-wood products. The demand for hardwood from tree plantations will continue to rise as the worldwide consumption of forest products increases. The environmental, commercial and political pressures of restricting the logging of high-quality trees from natural forests also increase (FAO 2001).

Tropical forests make up more than half of the world's forests. The productivity of these forests can be markedly increased by cultivating high-yielding varieties of native species from known or improved genetic stock. In this context, protection conservation in general tree improvement work, in particular, assumes significance. Therefore, an attempt should be made to set up an operational breeding program to obtain improved seeds for artificial reforestation. Tree improvement had a significant effect on forest management in temperate regions where the forestry is intensively practiced. It should make an even more substantial contribution in most tropical countries where forestry is now becoming more critical. The tree breeder faces many questions that must be

answered before a program can be started. He must know the market potential kind of product needed the growth potential the size of the area where the program is to be operated. Put another way, the critical requirement for a viable tree improvement program to be initiated as an active plantation program with a suitable species, that is, one that has been adequately evaluated over a full rotation (at least in experimental plots).

Obtaining regular supplies of improved seed is frequently a problem in the planting program in the tropics, as elsewhere. Fortunately, local seed-bearing stands and mature plantations available at present provide the possibility for securing early improvement:

1. Seed may be collected from the best individuals selected throughout the stand plantations.
2. Seed may be obtained from seed production areas established in plus stands or plantations from which phenotypically inferior trees will be removed. The area should be isolated to prevent an influx of foreign pollen.

An intensive program of intra-population improvement involves the following element:-

1. Selecting trees from the unimproved population.
2. Producing seed in some form, either from seedlings or vegetative propagules of these phenotypic selections.
3. Testing evaluating offspring of the first generation selection to improve the genetic quality of seed from the first-generation orchard by removing poor clones / or providing selected trees for the second generation with which to establish new orchards. Testing is also most important to validate the success of the selection program, so support must be continued.

Forest tree breeding is artificial evolution directed to improve the silvicultural success of the raw materials that forests produce. The first generation of tree breeding results

shows considerable gains in volume production and quality. Successive generations of tree breeding make much more significant improvements, especially when inexpensive methods for vegetative propagation are developed. Tree breeding programs are essential in Europe to meet the demand for wood, wood products, and a wood-based economy throughout the globe.

Here is the significance of tree breeding programs in tropical countries under high anthropogenic pressure like West, South and Southeast Asia, East and West African countries. India is one of the highly populated countries in the tropics with a high population density. Like any other country in the tropics, India needs many woods and wood products to meet the demand. India's total timber wood production was estimated to be about 70.9 M m³, whereas; the total estimated wood consumption (excluding fuel wood) in India comes to about 68.9 M m³/yr. India's demand for industrial wood has been growing steadily by an average of 0.9 M m³, reflected evidently in the increase in imports of industrial round wood, which grew from 2.55 M m³ in 2001 to 6.23 M m³ in 2014. If the imports increase at the same rate for the next 15-years, India's imports in 2020, 2025 and 2030 are projected to be 22.51, 27.01, 31.50 M m³, respectively, which would negatively impact the economy of the nation (Shrivastava and Saxena 2017).

On the other hand, the contribution of forests to the total domestic wood supply in the country is only 6.40% of the absolute domestic requirement. Hence, inadequate raw material available from the forests forced the wood-based industries to look toward the agro forestry farms for their sustenance. Since the expansion of farm areas in the country is not possible due to limiting factors of land resources, enhancing efficiency by incorporating fast-growing tree species on farmlands is a realistic approach to satisfy the demand for wood. Presently, exotic species like *Eucalyptus*, *Casuarina* and *Populus* species dominate the agroforestry farms as significant raw material sources. However, numerous problems pose a threat to the ecosystem because these crops are constrained by numerous problems.

There are two necessary conditions for the success of breeding programs:-

1. The existence of variation within the species for traits of interest.
2. Effectiveness of choosing the parents for the breeding programs.

The variation within a population is of two types, genetic or genotypic variation and phenotypic variation. Genetic variation is a fundamental requirement for the long-term stability of forest ecosystems since the amount of pattern of genetic variation determines the ability of forest tree species to adapt to the variability of environmental conditions. It is the diversity of gene frequency. Genetic diversity can be referred to as differences between individual's differences between populations. Phenotypic variation is the variability of the phenotypes of the population. Phenotypic variation is the cumulative result of a genotypic variation environment.

Terminalia paniculata Roth (Combretaceae) is one of the multipurpose tree species endemic to Peninsular India, commonly distributed in Southern Western Ghats of India (Chakrabarty and Kumar 2017). The tree grows up to 30 m in height and is more than 2.50 m in diameter at breast height distribution ranging between 800 m a.s.l. and 1200 m a.s.l. (Pillai 2017). Wood is commonly used for construction, agricultural implements, boats, plywood, and blackboard packing cases. Non-wood products from the tree are used for drug preparation, tannins, gums, oils, fodder certain organic compounds (Nazma et al. 1981; Jain and Dangwal 1985). Botanical Garden Conservation International and several other agencies listed *T. paniculata* as one of India's commercially important tree species (Nazma et al. 1981; Mark et al. 2014). But tree breeding domestication of *T. paniculata* is still in its infancy.

As mentioned above, assessing phenotypic variation in a tree species is a crucial starting point in any domestication program (Eriksson et al. 2006; Neale and Kremer 2011). Traits relating to fruit weight and size of wings are most commonly focused on in the case of winged fruits and stand-up tree characteristics. Several studies related to

population variation in terms of different fruit sizes and weights have already been conducted in other species, but no such studies have yet been carried out on *T. paniculata* (Abasse et al. 2011; Onyekwelu et al. 2014; Tsobeng et al. 2015).

Former studies confirmed that *T. paniculata* is one of the tropical trees with high seed emptiness and poor germinability. Even though the tree shows high seed emptiness, which results in poor germinability, it maintains its natural population by producing many fruits annually. It is essential to study the variation in fruit traits of targeted species to observe superior fruit traits. Quantitative characters such as yield and its determinants exhibit substantial interaction with the environment. Thus it is imperative to analyze the variability present in the germplasm and partition it into genotypic phenotypic nature. Release of high-yielding cultivars is impossible without ascertaining the magnitude of variation present in the available germplasm interdependence of growth pattern with yield. Selection for improved biomass is best estimated by growth attributes that should be made during the first 2-years, especially in trees and shrubs of perennial nature. Subsequently, environmental effects will have caused too much variation to distinguish effectively between genotypes (Dierig et al. 2001).

If a tree planter would like to develop a business plan based on tree planting and tree traits (e.g., total height, circumference at breast height, crown diameter and crown length), the fruit traits must also be known. This present study aims to perform the tree improvement program for the tropical timber tree species, *T. paniculata*, with the following objectives:-

1. Plus tree selection of *Terminalia paniculata* in different populations of Kerala.
2. To study the variations of *Terminalia paniculata* in different populations of Kerala.
3. To study the seed characteristics and seed handling techniques.
4. Development of a protocol for propagation in *Terminalia paniculata*.

CHAPTER 2
REVIEW OF LITERATURE

Chapter 2

Review of Literature

2.1. Plus tree selection of *T. paniculata* in different populations of Kerala.

Tree breeding is the most advanced applied branch of forestry science, focusing on genetic improvement and management of forest trees in terms of growth, reproduction and economic principles. This branch aims to apply genetic principles and practices in the progress of trees, varieties and populations to fulfill human needs. Long-term tree breeding aims to yield improvement, adaptation to unfavorable conditions, pest and disease resistance, wood quality and other properties. Tree breeding projects are active in all countries that depend on forest trees for various purposes, mainly for timber products. Even though the forest trees are genetically close to their natural range, significant variation exhibits between and within the populations in terms of tree growth pattern and wood quality. Tree breeding Programs were very active in cold countries due to reproductive constraints, anthropogenic pressure, extreme tree harvesting, and agricultural activities, especially in Europe. India is one of the largest countries in the world in terms of administrative area and human population. Among the total administrative area, 23% is notified as forest land by the Government of India (FSI 2019).

According to Forest Research Institute Dehra Dun, the total forest cover of India is divided into 5 different groups: moist tropical, dry tropical, montane temperate, montane sub-tropical and alpine forests. Forest areas account for 75% of the gross primary production and provide valuable ecosystem goods and services to humanity, including food, fiber, timber, medicine, pure water, aesthetic and spiritual values and climate moderation. According to the Food and Agriculture Organization, India's total

forest land area grew at 0.20% from 1990 to 2000 and 0.70% annually from 2000 to 2010 (FAO 2010).

Compared to general plant or crop breeding, tree breeding shows significant differences due to the long rotation nature, need of many years to attain reproductive maturity, the requirement of wild strands to begin, challenging hybridization features, etc. Tree breeding programs are based on conventional followed by modern tree breeding techniques. Tree breeding techniques begin with plus tree selection, provenance test, hybridization and accelerated breeding, followed by seed orchards (CSO: clonal seed orchard and SSO: seedling seed orchard), micro-propagation (somatic embryogenesis and organogenesis) and genetic engineering (gene transfer). Both conventional and modern tree breeding techniques like molecular breeding are very active. The objectives of tree breeding programs are economic characteristics (productivity and quality), adaptability (level of genetic diversity) and gene conservation (maintenance of the natural level of diversity). The specific conditions faced by any tree breeding program include biological, economic, institutional and socio-political factors (Thakur and Schemerbeck 2014; Berlin et al. 2012).

Plus tree selection is the beginning of a tree breeding program, including a population survey, candidate plus tree selection, elite tree selection, and province test. Generally, there are 3 methods of plus tree selection from candidate trees, such as comparison tree selection, baseline selection and regression method (Rudolf 1956; Jansons et al. 2009; Kim et al. 2020). The comparison tree selection method is suitable for even-aged tree strands, and the superiority of candidates over the average comparison tree is worked out for each trait. A candidate plus tree is designated as a plus tree if it proves superior to comparison trees; otherwise rejected. In the baseline method, the selection is made simultaneously for all characters but leaves all individuals who fail to meet the minimum selection standard for any trait. It is helpful for uneven-aged strands

when adjacent candidates plus trees are unavailable. The regression method is suitable for all aged tree strands, and candidates plus trees chosen by the breeder are subjected to regression analysis. Characters are plotted on the regression graph in order, the candidate plus trees placed above the regression line are accepted, and those set below the line are rejected.

Plus trees are phenotypically superior in exceptional growth rate, desirable growth habit, high wood quality, and exceptional resistance to diseases and pest attacks. They are identified from candidate plus trees without any progeny testing, from a population used for tree breeding programs like genetic improvement and management. Candidate plus trees have been selected for tree breeding programs to identify plus trees after comparing and testing. Plus trees have been proven to be genetically superior using progeny testing. Many scholars defined plus trees as a phenotype judged (but not confirmed by the test) to be unusually superior in some quality or quantity, e.g., exceptional growth rate, desirable growth habit, high wood density, exceptional apparent resistance to disease, and insect attack, or other adverse environmental factors or appearing distinctly superior to the average (Rudolf 1956; Clark and Wilson 2005).

The importance of plus tree selection as the first step of the tree improvement program, is to achieve enhanced timber productivity and quality. There are attributes to be considered during field assessment of plus trees and considerations for the selection, i.e., stem straightness, timber height, diameter, forking, branch angle, branch thickness, self-pruning, crown dimensions, fluting, straight grain, disease, and epicormics. A study detailed the attribute considerations in selecting plus trees of British hardwoods with the help of an ash tree datasheet, including details such as estate identification number, tree location, stand and site characters and tree characters. They also familiarized database development for digitalizing datasheet details for further research with the help of the

National School of Forestry, University of Central Lancashire, England (Clark and Wilson 2005).

The American forester Paul O Rudolf put forward the first reliable method of phenotypic selection of superior trees from a population (Rudolf, 1956). He prepared a guide for selecting prominent forest trees and stands in the Lake States. Rudolf was also interested in hybrid poplar planting in the Lake States, seed production areas, seed storage for western conifers, and the silvicultural aspects of Jack Pine (*Pinus banksiana*) and Red Pine (*Pinus resinosa*). He formulated the method for identifying phenotypically superior tree selection from a population stand based on morphological characteristics such as tree height, diameter at breast height, crown diameter and crown length. He also categorized the characters into four: characters easily observed or measured at any season; characters discernible only at certain times; characters not readily observable, and characters not directly concerned with wood production. The first category includes growth rate, crown development and stem form, and the tree selection formula is based entirely on the first character category (Rudolf 1956).

An experiment was conducted to test open-pollinated progenies from plus trees on average performing trees of Norway spruce (*Picea abies*) for the timing of growth bud set and autumn frost hardiness during the first growing season in each of five northern natural strands in Norway. Tests showed no difference between natural strand progenies from plus trees and control stand trees but showed significant differences in plus tree progenies from their seed orchard half-sibs. Test results were compared with non-native southern seed orchards. Environmental effects on the progenies showed higher phenotypic performance than expected in Southern Norway (Johnson and Ostreng 1993)

Ireland Forest Research Institute conducted a detailed survey to identify superior populations and individuals of Downy Birch (*Betula pubescens*) and Silver Birch (*Betula pendula*). They noted the taxonomy, distribution, morphology, genetics, seed production

and germination, seedling growth, timber properties, niche markets for birch timber products and research on birch. Tree breeding programs on genus *Betula* included population selection (individual selection, birch database formation, identification of distribution, location of birch, documentation of managed and planted birch sites and growth observations) followed by seed collection, grafting, studies on the timing of bud break, female flower production, pollen release, the study of catkin development, seed production from controlled crosses, planting material production, germination of fresh seed, seedling production and clone production (O'Dowd 2005).

Assessment of tree breeding and tree improvement programs on Sitka Spruce (*Picea sitchensis*) in Great Britain resulted in advanced silvicultural practices, most suitable for different planting programs. Silvicultural practices improved growing stock material for growers to get maximum return according to their objectives. The current phase of tree breeding and improvement on *P. sitchensis* needs a combined approach to maximize the benefits through the best silvicultural packages and genetically improved planting materials. The assessment suggested that tree selection and progeny testing enhance the quality of planting materials (Hubert and Lee 2005).

The Latvia Forest Research Institute conducted tree breeding programs on Scots Pine (*Pinus sylvestris*), the only Pine native to northern Europe to meet the timber demand. Scots Pine is categorized by relatively short, blue-green leaves with orange-red bark. For the study, data collected from ten open-pollinated progeny trials between 14-year to 33-year-old plants were used and estimated characteristics such as tree height, clear bole length, diameter at breast height, slenderness, the diameter of the thickest branches, number of branches, crown length, stem volume, etc. The study concluded that branch thickness and clear bole length had narrow-sense heritability. Low heritability was recorded for slenderness and proportional length of the green crown and branches per whorl. Correlation analysis revealed that tree height was weakly correlated with

branch thickness and strongly correlated with clear bole length. To increase the precise bole length by decreasing branch thickness, it is sufficient to include the diameter of the thickest branch in the first 2-meter in the selection index (Jansons et al. 2009).

Scots Pine (*Pinus sylvestris*) is a timber tree endemic to Spain, later introduced to Portugal to fulfill the demand for timber and timber products. Detailed tree breeding programs conducted in Southern Portugal to increase the production of Scots Pine kernel resulted in sixty-four phenotypically superior plus trees. Morphological and reproductive characters (number of cones, cone crop weight and relative production capacity) were used to identify the plus trees for other breeding programs. Cluster analysis was also carried out based on diameter at breast height, crown height, crown diameter, crown area, basal area and height diameter ratio to classify Thousand two-hundred and thirty-seven Pinus trees in Southern Portugal (Carrasquinho et al. 2010).

Guide for selecting forest trees and silviculture in Ireland detailed the plant species and forest planning, site productivity, species selection guidelines, mixed species plantations, covering species such as broadleaves, conifers, native trees and shrubs. The study also listed the common and botanical names of trees and vegetation, common and scientific names of pests and diseases, soil sampling guidelines, and foliar sampling guidelines (Horgan et al. 2003).

Northern Europe, including Scandinavia, is the central point globally tree breeding activities. Tree breeding activities in Northern Europe included breeding principles, molecular and biotech methods, inbreeding, breeding in connection to climate change, mass propagation (seed orchards, vegetative propagation, provenances and genetic diversity of the regeneration material), legislation, breeding status, potential increases in growth through breeding and economic gain. The tree breeding programs in Northern Europe are more focused on *Piceaabies* and *Pinus sylvestris* (Haapanen et al. 2015).

In 2013, the result of the Seventh Framework Programs of European Community Supported Research updated the best practices for European tree breeding. The published work communicates about the breeding strategies and evaluation of alternatives in the case of Scots Pine (*Pinus sylvestris*), and Maritime Pine (*Pinus pinaster*) and extension of best practices for other trees authored by the famous European tree breeders from Spain, Sweden, France, United Kingdom, and Finland. The work detailed about objectives of breeding programs, consideration and constraints of tree breeding, general strategies for deployment, breeding, testing, selection, comparing alternative methods, species-wise tree breeding programs, and status in the case of significant trees in Europe and other countries outside Europe. The study has discussed biological, economic, institutional, socio-political and breeding strategies such as population management, mating, phenotype assessment, genotype assessment and selection (Rosvall and Mullin 2013).

A multi-locational study of Monterey Pine (*Pinus radiata*) was conducted in drought-prone sites on sandy soil in Mediterranean dry lands in Central Chile to evaluate the field performance. The study assessed phenotypic variability of growth rates, survival, and six eco-physiological characters (root collar diameter, height, volume, survival, net photosynthesis, transpiration, stomatal conductance, intrinsic water use efficiency and predawn water potential) in thirty-open and control-pollinated families from two-breeding families and three-breeding generations in a 3-year-old field trial. The study showed no difference between the populations in terms of regional origin by analyzing the characters. However, a significant difference was established by the effect of breeding generations in terms of tree height and diameter. The study concluded that drought is an essential environmental factor in Mediterranean ecosystems that affects seedling survival and productivity (Espinoza et al. 2017).

In Canada, tree breeding programs focus more on *Picea*, *Pinus*, *Pseudotsuga* and *Abies*. Among the above species, except Norway Spruce, *Picea abies* (European endemic)

and Scots Pine, *Pinus sylvestris* (Eastern Siberia endemic), all others are endemic to Canadian provinces. The study explained seed collection; provenance and tree breeding research of the species mentioned above in Canada and updated the then status of provenance trials (Holst 1962).

Plus tree selection program of Coast Douglas-fir was conducted in British Columbian coastal land approximately half a century ago. There are three-varieties of Douglas-fir native to Northern America such as *Pseudotsuga meinziesii* var. *meinziesii*, *P. meinziesii* var. *glauca* and *P. meinziesii* var. *lindleyana*. Except for *P. meinziesii* var. *lindleyana*, the other two varieties are endemic to British Columbia. *P. meinziesii* var. *lindleyana* plus tree selection program covers a detailed study regarding the selection of area, selection of the strands, selection of tree within strands, registration and records, general considerations of the selection, criteria developed for the selection, criteria for subsequent selection, a summary of cruising for plus trees, scion material collection and propagation, wood quality and present status of the programs. Selection criteria include tree height, diameter, disease and pest resistance, narrow wedge-shaped crown, double leaders, straightness, clear bole, light and short branches, inter-nodal branches, thin bark and cone production, etc. (Heaman 1967).

Another tree improvement program through clone production was carried out through vegetative propagation from phenotypically superior trees in terms of height, diameter at breast height and branching habit of Black Spruce (*Picea mariana*) and White Spruce (*Picea glauca*) in Central Canadian Province, Ontario. Both the species were endemic to Canada and well known for their timber products. Materials for vegetative propagation were selected from 6-local nurseries; only white spruce was chosen from the three southern nurseries. Propagation yielded information regarding the effectiveness of selection, the clonal repeatability and total genetic variation of certain characters for quality planting material production (Rauter 1973).

A study on endemic Black Spruce (*Picea mariana*) was conducted in Quebec, Canada, to understand the productivity by site tree selection and site index determination of Black Spruce stands. A sum of thirty-six plots from six-sites was selected for the study. Measures such as stem density, total basal area, total volume, age at one-meter height, and top size of individuals were recorded from each plot to analyze productivity. To standardize the site determination index, variables such as dominant height, age, site index, rotation age and volume at rotation age were calculated using different methods of site index determination (Maily et al. 2004)

Japanese Larch (*Larix kaempferi*) tree breeding programs in Japan focused on seed orchards, progeny testing; wood quality improvement (improvement in spiral grain and assessment of wood quality as sawn timber), resistance breeding and hybrid larch were started in the 1960s. Resistance breeding programs were conducted to overcome needle cast disease during the 1960s. 50-year-long tree breeding programs resulted in several registered varieties. In addition to that, several hybrids of Japanese Larch were developed with an efficient seedling production system. The development of combinations of approximately twenty-meter-long trees changed its nature from forestry species into a favorite bonsai tree in Western Europe (Susumu 2005).

New Zealand Forest Research Institute conducted a plus tree selection program on Swamp Gum (*Eucalyptus regnans*) using provenance tests between Australia and New Zealand populations. Provenance tests were conducted on height growth, *Mycosphaerella* leaf blotch disease resistance, branching quality, and stem straightness. Correlation analysis resulted in a significant correlation between the characteristics such as fast growth, good frost tolerance and disease resistance, and improved branching quality and stem straightness. Based on the results, a clonal seed orchard was developed from eight of the fifty five original New Zealand plus-trees and thirty-four new second-generation

plus-trees and seedling seed orchards from the seeds collected from twenty-three New Zealand and seven Australian provenances (Wilcox 1982).

Eucalypt tree selection programs have also been conducted in Brazil to understand the efficiency of early selection for cloning using sixty-one hybrid progenies. The study was born in the north, northwest and central state of Minas Gerais in Brazil. Circumference at breast height was measured for evaluation and assessed by change in coincidence index, genetic correlation, phenotypic correlation, Spearman rank correlation, correlated response, gain per year and repeatability of phenotypic values. The programs concluded that early selection was effective for cloning in the case of trees and parents (Lima et al. 2011).

A similar selection program on River Red Gum (*Eucalyptus camaldulensis*) was conducted in Iraq on growth performance and fiber morphology for different propagation techniques in Northern Iraq. A total of sixty trees were selected for study, which belonged to two different categories, 10-years and 5-years old. Parameters such as height, number of branches, diameter and straightness were measured and analyzed. The study resulted in the identification of 5-year (three numbers) and 10-year (four numbers) Old River red gum trees from the study area as plus trees (Qader et al. 2014).

The selection of superior provenance and plus trees of Himalayan Birch (*Betula alnoides*) was conducted in Southern Fujian, China-based on tree height, diameter at breast height, individual volume, stem form index, height to crown base, crown diameter, and crown form, and branchiness index. Among the twenty-five provinces used for the study, Provenance G is the best followed by Provenance L and suggested these two provenances for the collection of seeds for nursery. A total of hundred and eighty-two plus trees were screened out and could be applied for vegetative propagation practices (Chen et al. 2020).

Plus tree selection of *Pongamia pinnata* was conducted in Bali province of Indonesia based on growth parameters such as total height; clear bole height, diameter at breast height, canopy width and oil content through the comparison tree method. Hundred and twenty-six mature trees located on the coastal beach of Bali and eight-plus trees of *P. pinnata* were identified using the comparison tree method. The study suggested that *P. pinnata* is concentrated in the Buleleng district of Bali, but the number of individuals is significantly less and thus needs further action for conservation (Arpiwi et al. 2018).

Teak (*Tectona grandis*) tree selection was conducted based on densitometry and wood characters using six hundred and sixty-nine trees in Togo and ninety trees in Benin. Characters such as diameter at breast height, total height, bole height and wood density, percentage of heartwood and color of heartwood were analyzed. Natural heartwood durability, fiber saturation point, modulus of elasticity and shrinkage were analyzed using infrared spectroscopy models. The study resulted in identifying thirty-three plus trees from six hundred and sixty-nine individuals (Kokutse et al. 2016).

Quercus salicina and *Q. glauca* plus tree selection were conducted in Korea based on characteristics such as growth (superiority in growth form and superiority in tree form), adaptability (to disturbance and environment) and seed production (distinction in seed production and potential of seed production). The study identified fifteen population strands (8 *Q. salicina*, three *Q. glauca* and four both) and eighty-five plus trees (fourty-five *Q. salicina* and fourty-one *Q. glauca*) of Oaks from Korea. The study suggested these plus trees for different oak breeding programs in Korea (Kim et al. 2020).

Institute of Forest Genetics and Tree Breeding Coimbatore conducted clonal multiplication programs on Teak (*Tectona grandis*). The tree breeding programs followed a preliminary assessment of the clones from superior quality trees from Kerala. They selected forty-one plus trees located in Nilambur and Thrissur forest regions of Kerala

with ages ranging from 51 years to 64 years, of which thirteen trees were superior in height (30 m to 35 m) and circumference at breast height (151 cm to 220 cm). Among the clones, seven trees showed a 30% increment in size, and fourteen trees showed a 30% increment in girth at breast height compared to plantation trees. The increment indicated the possibility of increasing productivity through clonal propagation. Vegetative propagation using coppice shoots resulted in clones between 45% and 100% rooting. Compared to all the forty-one clones, clones from Panayangode (Nilambur) exhibited maximum height (35 m) and circumference at breast height (220 cm) with 100% rooting competency, and this could be the best clone among the whole. The study suggests clones from Panayangode for *ex-situ* conservation and preservation of superior *T. grandis* genotypes for further tree breeding Programs (Palanisamy et al. 2009).

Forest Research Institute Dehradun and Kerala Forest Research Institute conducted many genetic improvement programs on Teak (*Tectona grandis*) focused on selection and breeding, vegetative and macro-propagation, germinability of seeds, radio-sensitivity of seeds, phyllotaxy variation, geographic variation, genetic characters, genetic variance, etc (Kedharnath and Mathews 1962; Sharma and Rawat 1998; Goh et al. 2005).

Pongamia pinnata breeding was conducted to identify candidates plus trees and evaluate variability in seed source. Twenty plus trees were selected based on age, tree height, canopy diameter, number of pods in 1-meter, seeds per kilogram, and oil content. Variability estimates were made based on pod characters such as length, thickness and weight, volume and seed characters such as width, thickness and volume. The study confirms the presence of significant variation in terms of morphological characters and oil content. Oil content in twenty plus trees varied between 31% and 42% (Raut et al. 2010).

Plus tree selection of Ghaf (*Prosopis cineraria*) was conducted in Haryana, India, based on age, total height; clear bole height, circumference at breast height, crown height, crown spread, and straightness. Correlation analysis showed that inter-correlation between the characters was significant. Juvenile growth of seedlings of all plus trees was recorded, and it concluded that the Bawal population is the most outstanding, followed by the Fatehabad population (Singh et al. 2019).

A study was conducted in Kerala to determine the genetic diversity and select candidates plus trees of *Ailanthus triphysa*. Based on the regression selection method, thirty candidates plus trees were identified in Kerala. Tree height (20 m to 37 m), girth (0.69 m to 2.11 m), crown diameter (3.5 m to 9.75 m) and clear bole height (7 m to 24 m) of candidate plus trees were recorded (Lalnunpuia et al. 2021).

Plus tree selection of Blue Pine (*Pinus wallichiana*) in Kashmir Himalaya was conducted based on characters such as total height, clear bole height, crown length, girth at breast height, diameter at breast height and bole volume using the scoring method. Plus, tree selection was followed by estimating seed germination of Blue Pine. The study suggested plus tree number fifteen as the best among the selected plus trees based on seed germination behavior and growth characters (Aslam et al. 2017).

A study was conducted on the genetic divergence of plus trees of Blue Pine (*Pinus wallichiana*) in the Kashmir valley of Himalaya. Eighty-eight plus trees belonging to ten clusters were analyzed during the study in various forest sections of Kashmir Himalaya. Experimental results showed that among the ten clusters and cluster eight is the best in seedling height, collar diameter, needle length, needle diameter and the number of needles per seedling (Aslam et al. 2011).

In *Melia dubia*, candidate plus tree selection was conducted based on qualitative and quantitative characteristics such as stem straightness, roundness, and tree height;

clear bole height, circumference, and disease resistance. Selection resulted in the identification of twenty candidates plus trees of *M. dubia* (Chauhan et al. 2018).

Melia azedarach plus tree selection programs were conducted based on stem straightness, self-pruning ability, clear bole height, low branching habit, and disease resistance. Characters such as field emergence, seedling height, shoot length, root length, basal diameter, number of branches per seedling and seedling dry weight were measured for progeny analysis. The selection programs identified twenty plus trees of *M. azedarach* (Daneva et al. 2018).

A tree breeding program was conducted to get plus trees of Sujan Gum (*Acacia nilotica*) and analyze their genetic diversity. Sixty candidates plus trees belonging to seven clusters were identified from the study area. Characters such as stem straightness, self-pruning ability, clear bole height, low branching habit and disease resistance were used for the selection. Seed characters were measured and analyzed for the genetic diversity among the seven clusters (Singhdoha et al. 2017).

Plus tree selection programs in European Beech (*Fagus sylvatica*) resulted in the identification of seven provinces. The comparison tree method was used to identify sixty plus trees from candidate trees based on phenotypical characters such as total tree height, girth, clear bole, branch angle, branch diameter, health, apical dominance, forking, etc. Population variation between provinces was calculated based on tree height, girth and precise bole length to find out the best among them. Variation analysis showed significant variation between populations in the native range (Kumar et al. 2003).

Tree variation analysis of European Beech (*Fagus sylvatica*) was conducted by examining two plantations. Both plantations were made up of seedlings from different provinces in India's northeastern, northern, central, and southern parts. Tree variations

with particular reference to tree forms in terms of clear bole, tapering, the persistence of axis, straightness, branch thickness and mode of branching were studied (Indira 2006).

Indian rosewood (*Dalbergia sissoo*) tree selection programs identified eighty-three candidates plus trees based on seed weight, germinability, and collar diameter; clear bole height, collar diameter, etc. All candidates plus trees showed significant differences in size, diameter and clear bole height among the individuals (Yadav et al. 2005).

Poplar breeding is developed in India with eight exotics, thirteen legitimate cultivars and more than three hundred imported cultivars. Instead of eight exotics, there are six endemic poplars present in India. Many legitimate cultivars and three hundred imported cultivars show the importance of poplar tree breeding in India (Khurana 2012).

Candidate plus trees selection was conducted to identify plus trees of *Salix alba* from twenty locations by examining the clonal trail laid out in the garden of JKAU. Morphological characters such as tree height, diameter, volume, clear bole length, and crown diameter were used for selection. Hundred candidates plus trees were marked from twenty locations based on phenotypical superiority with a height of more than 20%, a diameter of more than 35% and a volume of more than 150%. Correlation analysis showed a strong and positive correlation between most growth, biomass and leaf characters with collar diameter and volume index (Paray et al. 2017).

Fagus and *Picea* selection for professional and non-professional purposes depends on the tree breeder's behavior. A study on human behavior confirms that tree breeders have wrong perceptions about their tree selection styles and marking behavior. Changing the behavior of the tree breeder also influences the selection of the same tree repeatedly. The behavior of 5 tree breeders of their dependence on tree diameter and other characteristics was evaluated during the study (Pommenering et al. 2015).

A tree breeding program was conducted in *Terminalia chebula* and *T. bellerica* to confirm tree selection parameters. The study revealed that crown volume and precise bole length could be used as selection parameters in *T. chebula* while crown diameter, girth and fruit weight could be used in *T. bellerica* (Khobragade 2013).

Plus tree selection program on *Prosopis juliflora* was conducted in Uttar Pradesh, India, based on phenotypic superiority in growth, tree form and disease resistance, resulting in twenty-one plus trees. Progeny testing of plus trees showed that six of them outperformed than others, and among them, three-plus trees were outstanding in-field performance. The study suggested these three plus trees for other elite tree selection (Goel et al. 1997).

The genetic variability of *Terminalia chebula* was analyzed as part of tree breeding programs by using fortygenotypes in 2008. Each genotype was characterized by six tree morphological features (height, canopy diameter, diameter at the base, diameter at the breast, girth at bottom and perimeter at the breast) and twelve physical fruit parameters (fresh weight, diameter, length, dry weight, pulp weight, seed weight, moisture, total soluble solids, acidity, total sugar, reducing sugar and non-reducing sugar). Maximum variability observed was with morphological characters (diameter at the base, diameter at the breast, girth at bottom and rim at the breast). For selecting candidates plus trees, preference should be given to physical characters. Based on characters, four candidates plus trees of *T. chebula* were chosen from the study area (Navhale et al. 2011).

2.2.To study the variations of *T. paniculata* in different populations of Kerala.

Natural plant populations are influenced by their breeding system, gene flow, genetic drift, natural selection, and environmental and geological changes. Population genetic structure of adult plants shows the ecological and evolutionary processes that occurred in the past; meanwhile, the genetic structure of juvenile plants reveals the

current environmental methods. Natural populations vary from one-geographical site to another depending on plant phenotypic characteristics. The genetic component of individuals depends on the effects of the environment. For determining the provenance effects (variation among populations of different geographical origins), diverse phenotypic characters based on the studies were measured and analyzed by scholars. The objectives of a tree breeding program are mainly divided into economic characteristics (productivity and quality), adaptability (level of genetic diversity) and gene conservation (maintenance of natural levels of diversity). The primary deciding factors of the programs include biological (biology of the target species and ecologic conditions), economic (maximizing profit), institutional (planning and implementing) and socio-political (management activities) factors (Rosvall and Mullin 2013).

Phenotypic variation between the twelve populations of *Yucca capensis* in Baja California Sur of Mexico was analyzed in flowering and vegetative morphological characteristics; they included total length, stem length, stem circumference, rosette length, rosette diameter, leaf length and leaf width. The results showed a high coefficient of variation with plant length, stem length and rosette length and a low coefficient of variation with leaf length and leaf width. Also, the study revealed that all the vegetative traits had a positive relation with annual precipitation. The study confirms that *Y. capensis* has sizeable phenotypic variation in the case of vegetative characters, and rainfall has a significant influence on the production of reproductive structures (Arteaga et al. 2015).

Phenotypic variation and heritability of some morphological and physiological characters in *Fagus orientalis* in three different populations (700 m, 1200 m and 1700 m a.s.l elevation gradients) of Northern forests of Iran have been studied. Characters selected for the study included leaf length, leaf width, petiole length, leaf area, dry weight, relative water content, leaf index, petiole index and distance from leaf base to maximum leaf width. Statistical analysis revealed significant differences in characters

between the populations. High heritability was observed with distance from leaf base to full leaf width (Bijarpasi et al. 2019).

A study was conducted in North-eastern China regarding the phenotypic variability and genetic diversity in *Pinus koraiensis* clonal trials by analyzing twenty-eight phenotypic characters and sixteen microsatellites. Twenty-eight phenotypic characters included tree height, stem diameter and volume, bark thickness, stem straightness, branch angle, crown height and breadth, branch number per node, wood density, fiber length and width, cone number, length, width and weight, layer number, seed number, size, width and weight, nut length, width and weight and coat thickness. All the clones showed significant differences in most of the characters. Most phenotypic characters showed substantial genetic variation, but genotypic differentiation was weak between the clones. The analysis resulted in clustering the total clones into 3 groups based on phenotypic characters (Kaviriri et al. 2020).

The study on *Santalum austrocaledonicum* analyzed the geographic and phenotypic variation among different populations in Vanuatu in terms of its heartwood and essential oil characters. For the study, they measured heartwood, the basal diameter of the stem, the color of heartwood, heartwood oil, oil concentration and the percentage of four oils (alpha-santalol, beta-santalol, cis-nuciferol and z-beta-cucumen-12-ol) from eleven different populations in seven islands of Vanuatu. Even though the heartwood color varied between the trees, there was no significant relationship between color and santalol. However, there was a weak relationship between the oil concentration and color saturation. It was confirmed that heartwood color was not a factor in oil quality. This detailed novel study helped develop highly superior cultivars to meet the demand for sandals in the international market (Page et al. 2010).

A study conducted in Cameroon for domesticating *Cordyla africana* initiated with an analysis of the phenotypic variation in fruits and kernels. Fruits (n=24 x 52) were

collected from fifty-two trees from four land uses (home gardens, cocoa farms, fallows and forests) in two populations of Cameroon. They studied characters like mass (fruit, nut, kernel, flesh and shell), length and width of fruit, flesh thickness, flesh and fruit color, shell brittleness, taste score and fibrosity score. The study revealed highly significant variation in fruit length, fruit width, flesh depth, fruit and kernel mass and shell mass between villages but not between land uses. This work quantified the variation in fruit characters in *C. africana* and the domestication potentials (Atangana et al. 2001).

A study was conducted in Malawi to understand phenotypic variation in fruits and seeds of *Adansonia digitata*. Five populations, including three populations on the banks of Malawi Lake, were selected. The study documented fruit weight, fruit length, fruit width, number of seeds, seed weight and pulp weight by analyzing twenty fruits per tree from fifty-five trees located in five different populations. The statistical analysis showed a significant difference in fruit, seed and pulp weight, fruit length and width, number of seeds and single seed weight, and seed length and width between different populations in Malawi. The study also suggested other tree breeding programs to maintain the diversity in fruits and seeds of *A. digitata* in Malawi (Munthali et al. 2012).

Phenotypic variation of agro-morphological characters of *Vitellaria paradoxa* was analyzed by sampling forty-one populations located in Mali, including ten to thirty-five adults. It calculated twelve morphological tree, leaf and fruit) characters from each individual. Statistical analysis showed a shallow genetic relation between the three characteristics related to the tree, leaf and fruit. Leaf and fruit size characters were positively and significantly correlated with rainfall; in the meantime, tree circumference was negatively associated with rains. Drier areas were noted with large *V. paradoxa* populations recorded during the study, and it provided preliminary information for other breeding programs (Sanou et al. 2006).

Morphological variation among the forty-four ethno varieties (hundred and seventy-six individuals) of *Vitellaria paradoxa* conducted in Uganda based on pulp (taste, quantity and hardness), fruit (pubescence, size, length, width and weight), nut (length and width), leaf (length and width), petiole length, lamina (base angle and apex) canopy diameter, diameter at breast height characters. The study showed high pulp weight, stem diameter, fruit weight, and canopy diameter variation. Also, they observed a strong positive correlation between pulp and fruit weight, leaf length and width, petiole length, and leaf length. Cluster analysis divided these forty-four ethno-varieties of *V. paradoxa* in Uganda into three clusters (Gwali et al. 2012).

Variation between the populations of *Pinus densata* located in China was studied by analyzing the seed germination and early seedling growth characters. Seeds were collected from twenty open-pollinated trees within eight autochthonous populations in the natural distribution area. Results showed significant differences among the populations and seed relationship with seed size and seed weight. The study concluded that more giant seeds germinated earlier and faster than tiny seeds, and seedling size significantly correlated with seed size (Xu et al. 2016).

Variation in fruit and seed morphometry of *Chrysophyllumr oxburghii* located in Kodagu district of Karnataka, India, accessed by studying the characteristics such as fruit width, fruit length, fruit weight, fruit pulp weight, seed weight and several seeds. The study revealed that the population located in Appanagala showed superiority in all fruit and seed morphometric characters than other four populations and concluded that the high rainfall nature of Appanagala is the cause for the power (Sathish et al. 2020)

Variation between the populations (North Canara, Nilambur and Konni) of *Tectona grandis* was studied in Western Ghats, India, regarding mechanical and anatomical wood properties. Characters used for variation analysis included diameter at breast height, ring width, vessel, fiber, parenchyma, cell wall, modulus of rupture,

modulus elasticity, and maximum compressive stress and air-dry density. The study concluded the importance of variation between the populations for other *T. grandis* improvement programs and utilization of Indian genetic resources (Bhat and Priya 2004).

Phenotypic variation between the natural populations of *Juglan* in Northern Albania was studied to identify promising material for conservation. Two-hundred and fifty-three trees were initially labeled, but later, sixty-five trees were sorted out from them for phenotypic variation estimation. Nut characters such as nut weight, nut length, nut width, kernel weight, fat content and fruit weight were measured to estimate the phenotypic variation between the natural populations of walnut. Statistical analysis of the characters showed considerable variation between the populations, and the study proposed seven botanical varieties of *Juglan* in Northern Albania (Zeneli et al. 2005).

Interspecies variation of genus *Terminalia* (*T. cuneata*, *T. bellerica*, *T. chebula* and *T. myriocarpa*) located in Assam, India, were estimated based on wood anatomical characters such as wood color, hardness, heaviness, grain, texture, vessel (length and diameter), fiber (length, diameter and wall thickness), ray (height and width), inter vessel pitting and fiber, vessel, parenchyma and, ray. The variance of nine characters (fiber length, fiber diameter, fiber lumen diameter, fiber wall thickness, vessel length, vessel diameter, ray height, ray width and wood density) among *Terminalias* showed significant variation. Correlation analysis showed a strong positive considerable correlation of vessel length with wood density and a strong negative correlation with fiber diameter (Singh et al. 2013).

A study was conducted to estimate the phenotypic variation of sixty-seven individuals of *Euscaphis japonica* in nine different populations located in Southern China by measuring twenty-three phenotypic characters. Phenotypic characters included compound leaf (length and color), annual branch color, leaflet (number, area,

circumference, length and width), leaf (index, margin and texture), petiole length, fruit (width, length, index and color) and pericarp thickness, irregular ribs, fruit sequence color and seed (number, length, width and index). Correlation analysis revealed a significant difference in elevation and fruit color, irregular ribs, leaf margin and texture. Q-type clustering resulted in the clustering of sixty-seven samples into four clusters. Generally, leaf and fruit characters had abundant phenotypic diversity, and the study suggested two categories as deciduous type at high altitude and evergreen type at low altitude. Also, it provided a theoretical reference for its cataloging and variety, which is helpful for other conservation practices (Sun et al. 2019).

A phenotypic variation analysis on *Sclerocarya irrea* was conducted by analyzing eighteen provenances belonging to the African continent. Fruit characters such as several fruits, fruit weight, seed weight, pulp weight, fruit length and fruit diameter estimated during the study resulted in significant phenotypic variation between the populations (Mkwezalamba et al. 2015).

Sapwood and heartwood variation in *Eucalyptus tereticornis* was analyzed by examining thirty-six progenies collected from seven locations. Tree height, wood diameter, heartwood diameter, total taper diameter, taper wood diameter, taper heartwood diameter, wood content and bark thickness were measured. The study showed that heartwood content in the tree is positively related to tree size, genotypes and factors that will result in faster tree growth (Kumar and Dhillon 2014).

Genetic diversity within the Western Ghats populations of *Terminalia paniculata* was assessed on six allozyme systems. Six enzyme systems generated fifteen loci from four individuals and were used for estimating allele frequency, polymorphic loci and heterozygosity and Shannon information index. One-population resulted in twelve rare alleles, two private alleles and a high polymorphic level (87%) (Thangaraja and Ganesan 2012; 2015).

A study on leaf variation among genus *Terminalia* (*T. paniculata*, *T. cuneata*, *T. chebula*, *T. catappa* and *T. chebula*) located in the Kolhapur district of Maharashtra, India, in the similarity between species in terms of leaf characters such as leaf length, leaf width, petiole length and lateral nerve number. Principal Component Analysis results showed that leaf width significantly correlated with leaf length and lateral nerve. Cluster analysis showed that among the *Terminalias*, *T. paniculata* had maximum dissimilarity with other *Terminalias* in terms of leaf characters (Deshmukh et al. 2013).

Genus *Terminalia* are well known for recurring fruit characters such as body (size, shape, thickness and structure), wing (number, size, shape and consistency) and pubescence. Winged fruits have commonly occurred among American *Terminalias* except a few. But most of the species possess un-winged drupaceous fruits in Madagascar, South-East Asia, Melanesia and Australia. In tropical Asia, half of the *Terminalias* are winged fruited, and 55% of the species in the world have un-winged fruits. Type species, *T. catappa* has un-winged fruits with- a rounded shape. Many scholars have studied seed characters of winged and un-winged *Terminalias* in the tropics. A study on seed size, germination and seed viability of tropical tree species also recorded the seed character of *Terminalia paniculata*. The study recorded the flowering (March to April) and fruiting period (August to December), seed weight (2 g), viability (180-days) and germination days (15-days) of *T. paniculata* (Murali 1997).

Detailed information regarding seed characters of *Terminalia paniculata* was recorded in the seed manual published by Kerala Forest Research Institute. The published work recorded the seed maturity, collection, transportation, processing, seed description, seed dimension, weight, insect infestation, fungal infection, storage physiology, viability period, germination type, period, storage, viability test, pre-sowing treatments and seedling production (Chacko et al. 2002).

Variation among natural populations of *Terminalia cuneata* located in Central India estimated leaf morphological traits such as width, length, area, fresh mass, dry mass, thickness, petiole length, the distance between veins, midrib length, number of primary veins and veins length. All the phenotypic characters show significant variation between the populations, and correlation analysis resulted in a positive correlation between the characters (Wani and Singh 2016).

Variation analysis of *Terminalia bellerica* based on morphometric characters, germination and oil content was conducted between nine provinces in Odisha, India. The study concluded that the highest fruit length (3.87 cm), fruit weight (13.40 g), seed weight (6.45 g), kernel weight (0.99 g), maximum oil yield observed with Kantamal provenance and the exact provenance also show the best germination and seedling characters (Das et al. 2020).

Effect of pre-treatments on seed germination of *Terminalia ivorensis* carried out by analyzing the germinability of Four-hundred and eighty seeds. The study suggests mechanical scarification in *T. ivorensis* is the best method for enhancing seed germination (Chika et al. 2020).

Germination studies in *Terminalia sericea* proved the best temperature regime, photoperiod and pre-treatment. Experimental conditions included different temperature regimes (10°C, 15°C, 20°C, 25°C, 30°C, 35°C and 40°C), photoperiods (4/20, 8/16, 12/12, 16/8, 24/0 and light/dark) and pre-treatments (GA₃ 50 ppm, 100 ppm, 200 ppm and 400 ppm; KNO₃ 2000 ppm, 4000 ppm, 6000 ppm and 8000 ppm). The study proved that the optimum temperature regime was 25°C while the 12/12 light/dark hour photoperiod had relatively high germination of 33%. Maximum germination (67%) was obtained with GA₃ (400 ppm) and was insignificant in germination (Amri 2010). The diversity between two-tree populations (n= 40 trees) in South-Western Nigeria of

Terminalia catappa was conducted using twenty-eight quantitative and twelve qualitative characters.

2.3. To study the seed characteristics and seed handling techniques.

A study conducted in South Western Nigeria estimated the diversity between two populations of *Terminalia catappa*. Twenty-eight quantitative and twelve qualitative characters were measured from forty trees. These two populations show variability in terms of leaf shape and ripe fruit colour. Scholars recorded four clusters through UPGMA cluster analysis based on morphological characters. The study suggests fruit size, leaf size and fruit colour are the lead characters for further tree improvement programs (Oboh et al. 2008).

Study on the effect of altitudinal range (400 m a.s.l to 1400 m a.s.l) on seed storage of Terminalias such as *Terminalia bellerica*, *T. chebula* and *T. elliptica* conducted in the Garhwal Himalaya region of India. Germination characteristics such as seed length, thickness, weight, and germination were used for the study. The study concluded that the seed weight of *T.bellerica* is inversely correlated with altitude, but *T. elliptica* is positively correlated. The seed weight of *T. bellerica* and *T. chebula* is inversely correlated with germinability and positively correlated with *T. elliptica*. The germination of *T. chebula* and *T. elliptica* is positively correlated with altitude (Chauhan et al. 2007).

In Thrissur, Kerala, an experiment was conducted to estimate the seedling characters and early growth performance of candidates plus trees of *Ailanthus triphysa*. For the study, twelve candidates plus trees were selected from three agro-ecological zones of Thrissur, Kerala. Germination experiments resulted in that maximum germinability (83.50%) and minimum germinability (67%) being observed with candidate plus trees located in coastal sandy and high range agro-ecological zones. The study suggests that

the coastal sandy agro-ecological zone is the best agro ecological zone for the growth of *A. triphysa* (Abhijith et al. 2020).

Study on seed biology of Axle wood (*Anogeissus latifolia*) resulted that the seed emptiness is 95% when collected from trees. Germination percentage is 1-2%, but the study is recorded that 50% of the seeds are sound and viable. To check the impact of insect attack on seeds, seeds were subjected to insecticide application for a period of six months in field. Germination experiments on insecticide applied and not applied seeds resulted that there is no significant variation on seed germination of applied seeds than control. Study concluded that high seed emptiness in Axle wood is due to high insect infestation and can't control by applying insecticide and they suggests more research to crack the infestation (Singh et al. 2015).

Seed collection and germination studies in *Terminalia sericea* include the analysis of pre-treatment effects conducted in Malawi. Seeds were subjected to four pre-treatments (hot water soaking, immersing in sulphuric acid, cold water soaking and fire scorching). Two types of seeds were subjected to the experiment based on the color of the seed (deep green-brown and purple-brown-pink-purple). Among the first collection (deep green to brown color seed), maximum germinability (51%) was observed with seeds treated with cold water soaking for 12 hr. Second collection (purple-brown-pink-purple), maximum germinability (14%) was marked with seeds immersed in sulphuric acid for 2 hr (Likoswe et al. 2008).

A study was conducted on seed germination and seedling development of selected tree species, including *Terminalia elliptica*, *T. bellerica* and *T. chebula*. Three seeds were chosen for the study (small, medium and large) and calculated seed weight, germinability, collar diameter, and seedling height (after 3 months and 6 months). The results indicated that large-sized seeds showed maximum germination compared to small and medium-sized seeds of all the species, such as *T. elliptica* (57%), *T. bellerica* (100%)

and *T. chebula* (47%). Large seeds showed maximum seedling growth also after three months and six months in terms of collar diameter and seedling height. The study suggested collecting large-sized seeds for multiplication and seedling establishment (Negi and Todaria, 1997).

A study on seed germination, seedling growth and biomass production of *Terminalia chebula* was conducted in Manipur, India. Under the survey, de-pulped seeds of *T. chebula* (three different sizes) were subjected to eight pre-sowing treatments, followed by seedlings' transplant (three different potting mixtures). The study concluded that maximum germinability was observed with seeds nicking at the broad end and soaking in water (36 hr). Among the potting mixtures, soil, sand and farmyard mixture (1:2:3) are best. Suggest suggests a particular pre-sowing and potting methods for other nursery practices and germination experiments (Benjamin et al. 2019).

To enhance the natural germinability of *Terminalia bellerica*, a germination experiment was conducted at Kerala Agricultural University Thrissur, India, by various pre-treatments. Pre-treatments include seeds soaked in soaking (24 hr), seeds soaked in water (48 hr), seeds soaked in soaking (72 hr), de-pulped seeds soaked in water (24 hr), alternate drying and wetting (5-days), seeds soaked in cow dung (72 hr), and seeds soaked in 10% sulphuric acid (10 min). Maximum germinability (47.50%) was recorded with de-pulped seeds soaked in water (24 hr) followed by seeds soaked in water (48 hr). The seedling growth pattern indicates that alternate drying and wetting (5-days) is the best treatment and shows better performance (Kumar 2017).

Germination studies conducted in *Terminalia bellerica* covered seed germination and seedling establishment based on growing media, nursery beds and containers. Effect of manures (bio-organic soil enriched, farmyard manure, goat and poultry manure mixed with soil), nursery beds (hollow, flat and rose), containers and soil (silt loam, sandy loam and sandy) were examined during the study. The experiment resulted in maximum

germinability in farmyard manure, sunken beds, 4000 ml plastic pots, a minimum in poultry manure, 350 ml root containers, and 1600 ml polybags. Seedling survival was the highest in farmyard manure, sunken beds and silt loam soil and 4000 ml plastic pots compared to other treatments and reported the combination as the best germination and seedling establishment. Survival was the highest in farmyard manure and bio-organic soil enriched under field conditions, but seedlings raised in plastic pots recorded maximum height and collar diameter (Bali et al. 2013).

A study was conducted in *Terminalia chebula* to examine the effect of germination and seedling growth in the nursery. Seeds subjected to different pre-treatments such as control, de-pulped, de-pulped seeds soaked in cold water (12 hr), de-pulped seeds soaked in cold water (24 hr), de-pulped seeds soaked in cold water (48 hr) and de-pulped seeds soaked in hot water (2 min). Germination was observed between 29-days to 86-days, and 67% was the highest. The highest germinability (67%) was marked with de-pulped seeds soaked in cold water (48 hr), followed by de-pulped seeds soaked in cold water (24 hr) (60%), and the lowest germinability was observed with control seeds (49%). The result showed an increase of 18% in seed germination with pre-treatment. The highest germination value (4%), germination energy (59%) and vigor index (5291) were also obtained with de-pulped seeds soaked in cold water (48 hr) (Hossain et al. 2005).

A study was conducted in *Terminalia chebula* to understand the effect of de-pulping and water soaking of seeds for enhancing seed germination and growth characteristics. Thousand and two-hundred seeds were subjected to various pre-treatments under two categories: pulped seeds and de-pulped seeds six-hundred each. Seeds were subjected to pre-treatments soaked in water for 24 hr, 48 hr and 72 hr, respectively. High germination (73.80%) was observed with de-pulped seeds soaked in cold water for 48 hr, and low germinability was marked with pulp seeds. Growth characters such as shoot length, root length, total height, leaf number, leaf area, collar

diameter, dry mass and vigor index were measured to analyze the effect of pre-treatments on growth. The same treatment mentioned above shows maximum and significantly higher than other treatments. Thus the study suggests de-pulped seeds soaked in cold water for 48 hr for further seed germination experiments and nursery practices (Hossain et al. 2013).

Former studies recorded the seed germination of *Terminalia paniculata*, and the experiment was conducted in Kerala and Tamil Nadu states of Peninsular India. Murali (1997) recorded 15% seed germination, and a study conducted by the Tamil Nadu Forest Department supports the former by registering seed germination of 15% and germination duration of 25 days (TNFD 2005). Experiments conducted in Kerala recorded very low germination of *T. paniculata* seeds, ranging between 0.75% and 2% (Pillai and Chandrasekhara 2011; Chacko et al. 2002).

A study conducted under nursery conditions to estimate the effect of seed size on seedling emergence of *Terminalia bellerica* resulted in three grades (large: 1.18 g, medium: 0.95 g and small-sized: 0.76 g) of seeds show significant variation in terms of seedling emergence. Seed weight shows a positive correlation with seedling emergence in *T. bellerica*. The study suggests that selecting large-sized seeds for mass propagation is advisable (Kuniyal et al. 2013).

To break the dormancy and determine the storage behavior of *Terminalia laxiflora* studied, the seeds were subjected to four pre-treatments and germination trials before and after 2-years of storage (-10°C). Seed pre-treatments include control, scarification, and soaking in cold water, hot water and high temperature (78°C) treatments (5 min, 10 min and 15 min). Germinability of *T. laxiflora* before storage results that maximum germinability (80%) observed with high-temperature treatment (10 min). While control, scarification and cold water soaking treatments resulted in zero germinability. After 2-years, the germinability of stored seeds resulted in maximum germinability (78°C)

observed with high-temperature treatment (15 min). The study suggests high temperature (10 min) treatment for initial germination trials and high temperature (15 min) treatment for stored seeds germination (Mewded et al. 2018).

Effect of pre-treatments on seed germination and juvenile growth of *Terminalia bellerica* analyzed by the seeds collected from plus trees located on Bangladesh Forest Research Institute campus. To check the impact of seed pre-treatments, both pulped and de-pulped seeds were subjected to cold water soaking for 24 hr, 48 hr and 72 hr. Maximum germination (93%) resulted in de-pulped seeds soaked in cold water for 48 hours, followed by de-pulped seeds soaked for 24 hours. Pulped seeds subjected to water soaking (24 hr, 48 hr and 72 hr) show approximately 50% reduced germinability than de-pulped seeds. The seedling growth pattern is also higher in the same treatment mentioned above, and this study suggests this pre-treatment for further germination experiments and nursery practices (Hossain et al. 2014).

Effect of pre-treatments on seed germination of *Terminalia cuneata* seeds estimated by 3 categories (small, medium and large-sized) of seeds collected on the campus of SKUAST Jammu. Three varieties of seeds were subjected to four pre-treatments, such as water soaking (36 hr), hot water soaking (6 hr), concentrated sulphuric acid soaking (10 min) and control. Study results showed that seeds treated with concentrated sulphuric acid and large-sized seeds perform better than other treatments and seed categories (Zazai et al. 2018).

A study was conducted to understand the effect of pre-treatments on the germination of *Terminalia chebula* seeds without endocarp. Five pre-treatments excluding control were applied during the research, and seeds were collected from five provenances of Karnataka, India. Maximum germination occurred with 500 ppm GA₃ treatment with seeds collected from four provinces viz., Arasikere (74%), BR Hills (80%), Madhugiri (62%) and Shimoga (42%) and 200 ppm GA₃ treatment with seeds collected from

Channapattana (76%). Maximum seedling growth in height occurred with 500 ppm GA₃ in three provinces and with 200 ppm GA₃ in two provinces. The study revealed that GA₃ treatment is the best for the germination and growth of *T. chebula* seeds without endocarp than control and water soaking treatments, and 500 ppm GA₃ was the best among them (Raju et al. 2014).

Seed germination studies in *Terminalia paniculata*, *T. bellerica*, *T. chebula* and *T. catappa* have been conducted. The study examined the effect of synthetic hormone (IAA, GA₃ and 2, 4-D) on the germinability of Terminalias. The study revealed that plant growth hormones had a role in seed germination results positively or adversely. Authors suggested that synthetic hormone could be used for germination enhancement in selected *Terminalia* at different concentrations (Kadam et al. 2015).

Variation between 5 tree populations (n=5 x 20) of *Terminalia chebula* located in Himachal Pradesh, India, estimated fruit and pulp characters, including fresh fruit weight, dry fruit weight, fruit length, fruit breadth, new pulp weight and dry pulp weight. Analysis of variance resulted in fruit and pulp characters showing a significant correlation between the populations. Praur (Kangra district, Himachal Pradesh) population shows superiority in all characters except fruit length and kernel length. The study shows the significant-high tree to tree variation of *T. chebula* in fruit and pulp characters (Sharma et al. 2015).

Geographical variation in seed morphological characters of *Terminalia cuneata* has been analyzed. Seeds were collected from thirty different provinces, and morphological characters (age, girth, height, length and width of the fruit) were recorded. Seed morphological and germination characteristics such as length, width, hundred-seed weight, germinability, mean daily germination, peak value, germination value and germination speed were evaluated and recorded. The study suggested medium and large-

sized seeds for growing because of their higher germinability and seedling growth (Kumar et al. 2017).

Seed production potential of *Terminalia alata* has been analyzed by comparing trees from three different forest types viz., moist deciduous, dry deciduous, and semi-evergreen and three other DBH classes viz., 10 cm to 40 cm, 40 cm to 80 cm and above 80 cm. Fruit yield characteristics include the number of primary branches per tree, number of secondary branches in the main branch, number of fruits per tree, and weight per tree. The maximum number and weight of fruits per tree occurred in moist deciduous forest type (DBH>80 cm) followed by wet deciduous forest type (DBH 40 cm to 80 cm) (Shivaprasad and Channabasappa 2011).

Seed regeneration of *Terminalia elliptica*, *T. paniculata* and *T. travancorensis* in natural forest land was observed to analyze the regeneration, phenology and seed biology and develop nursery packages. Among them, *T. paniculata* was observed in hundred and sixty eight plots, *T. elliptica* in hundred and one plots, and *T. travancorensis* in five zones (northern, eastern, central, high range and southern), showing that *T.paniculata* is more or less stable in Kerala. The regeneration pattern showed 46% of unestablished seedlings (<3 cm circumference), 24% established seedlings (3 cm to 9.99 cm circumference) and 30% advanced category (10 cm to 30 cm circumference). *T. elliptica* showed 56% seedlings, 26% saplings and 18% poles, and *T. travancorensis* showed a negligible regeneration rate. The study revealed that the germinability of *T. paniculata* was very low due to infertility and heavy pest infestation (Pillai and Chandrasekhara 2011; Pillai 2017).

Terminalia brownii is an African multipurpose tree species widely distributed in Kenya and whose potential is underutilized due to poor seed germination. To study the dormancy and germination of *T.brownii*, a sample of hundred fruits and extracts were collected from different sites. These seeds subjected to different pretreatments (control,

de-winged fruits, fruits nipped at the distal and proximal ends and extracted seeds) and conducted germination trials. Extracted *T. brownii* seeds show maximum germinability (76%) and fruits nipped at the distal and proximal ends shows minimum (13%). The study concluded that physiological seed dormancy is due to hard samara fruit and suggests seed extraction is the best method to overcome this (Okeyo et al. 2020).

2.4. Development of a protocol for propagation in *Terminalia paniculata*.

In India, 16 species of *Terminalia* are indigenous. Even though these species produce many fruits, *Terminalia*'s are characterized by poor seed germination and low natural regeneration status. Researchers all over India strive to develop propagation protocols for these species to meet the demand. A study conducted to estimate the effect of seed size on seedling germination of *Terminalia bellerica* indicated that seed size significantly influenced germination. Seed weight had a positive correlation with seedling emergence. The study suggests that selecting large-sized seeds for propagation is advisable (Kuniyal et al. 2013).

A study conducted in Ethiopia aimed to breaking the seed dormancy and to determine storage behavior of *Terminalia laxiflora*. Seeds subjected to four pretreatments (soaking, scarification, high temperature and control), done before storage and storage after 2-years (dry storage at -10°C). Before storage, seeds treated with high temperature (78°C for 10 min) show maximum germinability (80%). After storage, seeds treated with high temperature (78°C for 10 min) show maximum germinability (75%). The study suggests high temperature treatment is the best pretreatment to break dormancy of *T. laxiflora* seeds (Mewded et al. 2018).

Effect of pre-treatments on seed germination and juvenile growth of *Terminalia bellerica* by subjecting pulped and de-pulped seeds to cold water soaking for 24 hr, 48 hr and 72 hr. Maximum germination (93%) was observed in de-pulped seeds soaked in cold

water for 48 hours, followed by de-pulped seeds soaked for 24 hours. Pulped seeds subjected to water soaking (24 hr, 48 hr and 72 hr) had approximately 50% reduced germinability than de-pulped seeds. The seedling growth was also higher in de-pulped seeds soaked in old water for 48 hr (Hossain et al. 2014).

Effect of pre-treatments on seed germination of *Terminalia cuneata* seeds estimated pre-treating three categories (small, medium and large-sized) of seeds to water soaking (36 hr), hot water soaking (6 hr), concentrated sulphuric acid soaking (10 min) and control. Large-sized seeds and treatment with concentrated sulphuric acid were better for germination (Zazai et al. 2018).

Effect of pre-treatments on seed germination of *Terminalia chebula* collected from 5 provenances (Madhugiri, Arasikere, BR Hills, Shimoga and Channapattana) of Karnataka, India was studied. It was observed that seeds of Madhugiri provenance treated with 100 ppm and 200 ppm IBA (1 hr) took minimum days (9.5-days) to complete germination. Seeds of BR Hills and Madhugiri provenances treated with GA₃ 500 ppm had maximum germination (80%) and shoot length (21.3 cm), respectively (Raju et al. 2014)

A study was conducted to understand the effect of pre-treatments on the germination of *Terminalia chebula* seeds collected from 5 provenances in Karnataka. 5 pre-treatments excluding control were applied on seeds without endocarp. Maximum germination occurred with 500 ppm GA₃ treatment results with four provinces viz., Arasikere (74%), BR Hills (80%), Madhugiri (62%) and Shimoga (42%) and 200 ppm GA₃ for seeds collected from Channapattana (76%). Maximum seedling height growth occurred with 500 ppm GA₃ in three provinces and with 200 ppm GA₃ in two regions. The study revealed that GA₃ treatment is the best for the germination and growth of *T. chebula* seeds, and 500 ppm GA₃ was the best among them (Raju et al. 2014).

A study examined the effect of synthetic hormone (IAA, GA₃ and 2,4-D) on the germinability of *Terminalia paniculata*, *T. bellerica*, *T. chebula* and *T. catappa*. The study revealed that plant growth hormones had a role in seed germination. The authors suggested that synthetic hormone at carrying concentrations could be used for germination enhancement in Terminalias (Kadam et al. 2015).

Variation in fruit and seed characters of five tree populations (n= 5 x 20) of *Terminalia chebula* in fruit and pulp characters, including fresh fruit weight, dry fruit weight, fruit length, and breadth of new fruit pulp weight and dry pulp weight, were studied. Fruit and pulp characters showed a significant difference among the populations. The study indicated significant-high tree to tree variation of *T. chebula* in fruit and pulp characters (Sharma et al. 2015).

Geographical variation in seed morphological characters of *Terminalia cuneata* was analyzed. Seeds were collected from thirty different provinces. Seed morphological (age, girth, height, length and width of the fruit) and germination characters (length, width, hundred-seed weight, germinability, mean daily germination, peak value, germination value and germination speed) were studied. The study suggested medium and large-sized seeds had higher germinability and seedling growth (Kumar et al. 2017).

Seed production potential of *Terminalia alata* was analyzed by comparing trees from three different forest types viz., moist deciduous, dry deciduous and semi-evergreen and three other DBH classes viz., 10 cm to 40 cm, 40 cm to 80 cm and above 80 cm. The maximum number and weight of fruits per tree were observed in trees of moist deciduous forest type with DBH>80 cm followed by DBH 40 to 80 cm (Shivaprasad and Channabasappa, 2011).

Seed regeneration of *Terminalia elliptica*, *T. paniculata* and *T. travancorensis* were studied to analyze the regeneration, phenology and seed biology and develop nursery

packages. Among them, *T. paniculata* was observed in hundred and sixty-eight plots, *T. elliptica* in hundred and one plots, and *T. travancorensis* in five plots (northern, eastern, central, high range and southern), showing that *T. paniculata* is more or less well distributed in Kerala. The regeneration pattern of *T. paniculata* showed 46% of unestablished seedlings (<3 cm girth), 24% established seedlings (3 cm to 9.99 cm girth) and 30% advanced category (10 cm to 30 cm girth). *T. elliptica* regeneration had 56% seedlings, 26% saplings and 18% poles, while *T. travancorensis* showed a very nominal regeneration rate. The study revealed that the germinability of *T. paniculata* was very low due to infertility and heavy pest infestation (Pillai and Chandrasekhara 2011; Pillai 2017).

Clonal multiplication through vegetative propagation is a comparatively more straightforward method for producing a maximum number of copies within a short period. Clonal multiplication through vegetative propagation is very common in the case of tree species. Several types of vegetative propagation methods and protocols have already been developed all around the globe. Vegetative propagation by artificial means includes cutting, grafting, layering, suckering, tissue culture, etc. Clonal multiplication through rooted cuttings is effortful in the case of hardwoods trees and scholars have developed protocols for clonal multiplication in several trees like *Tectonagrandis*, *Swietenia macrophylla*, *Eucalyptus*, *Gmelina arborea*, *Meliadubia*, *Dalbergia*, *Santalum album* Uniyal et al. 1985; Gurumurthi et al. 1988; Surendran 2014; Uniyal et al. 1993; Gupta et al. 1993; Hossain et al. 2004; Yasodha et al. 2005; Amri et al. 2010; Singh et al. 2012; Azad and Matin 2015; Azad et al. 2016; Geetha et al. 2018; Borpuzari and Kachari 2019; Nhung et al. 2019), etc.

Propagation techniques of forest trees in the Western Ghats region of India were assessed by Bhat et al. (2001) as the reestablishment of indigenous species instead of plantations of exotic species. They assessed both trees and shrubs in the Western Ghats,

especially the genus *Terminalia* (*T. bellerica*, *T. paniculata* and *T. tomentosa*). They grouped the plants based on propagations methods such as propagating through seeds and vegetative parts, seeds only and vegetative parts only and tried propagation. They listed *T. paniculata* in the first (propagated through seeds and vegetative parts) category.

After the propagation trials, scholars listed *T. paniculata* as problem species because of the failure in propagation along with *Alseodaphnae semicarpifolia*, *Celtis cinnamomea*, *Dillenia pentagyna*, *Ficus asperrima*, *Ficus asperrima*, *Flacourtia montana*, *Lagerstroemia microcarpa*, *Mallotus phillippensis*, *Randia spinosa*, *Schleichera oleosa*, *Strychnos nux-vomica* and *Vitex altissima*. Like *T. paniculata*, *L. microcarpa* is also a well-known timber species among the problem species. They suggest that the failure in propagation may be due to the short viability of the seeds, lack of specific treatment, effects of the season of the planting, size and age of cuttings, type of branch or internal factors.

The effect of the hormone on the rooting of *Swietenia macrophylla* cuttings was estimated by collecting stem cuttings from phenotypically superior trees. Stem cuttings treated with 0.40% IBA had 97% rooting followed by treatment with 0.20% IBA and control. The number of roots produced per cutting was maximum with cuttings treated with 0.40% IBA followed by 0.20% IBA (Hossain et al. 2004).

In a study on the effect of different potting media (sand, farmyard manure, clay soil, loamy soil, vermiculite and coco-peat) on the growth of *Terminalia bellerica* seedlings, it was observed that a mixture of sand, farmyard manure and loamy soil (1:2:1) as the potting media gave the best growth in seedlings (Patel et al. 2013).

Clonal propagation protocols for a few *Terminalia* viz., *T. bellerica*, *T. catappa* and *T. chebula* have been reported (Bhardwaj et al. 1993; Jose and Thomas 1998; Sharma and Thakur 2002; Shidiki et al. 2013; Kusum et al. 2017; Babu et al. 2018; 2019). Vegetative propagation protocols have been developed in *T. cuneata* using 1-year-old branch cuttings

by treating with synthetic hormone (IBA and NAA) in different concentrations (500 ppm, 1000 ppm, 1500 ppm and 2000 ppm). The experimental result showed that IBA was more effective than NAA. Of different concentrations, 2000 ppm IBA concentration was the best and resulted in more than 75% rooting in cuttings (Kusum et al. 2017).

Vegetative propagation studies in *Terminalia bellerica* and *T. chebula* from stem cuttings. 20 cm to 25 cm long stem cuttings with 1 node to 2 nodes were selected from 1-year-old healthy shoots of mother plants. Cuttings were collected from the terminal, middle and basal portions of the stem, treated with three concentrations of IBA (2000 ppm, 4000 ppm and 6000 ppm), and planted in October and March. *T. chebula* cuttings were planted in March sprouted and rooted poorly. The middle portion of the treated cuttings (IBA 4000 ppm) gave the best shoot and root growth. Maximum sprouting obtained in the shoot was collected from the middle without any treatment (38%), followed by base cuttings treated with IBA 4000 ppm (33%). Maximum rooting was obtained in the case of shoot collected from the middle treated with IBA 4000 ppm (38%), followed by shoot collected from the terminal part treated with IBA 4000 ppm (30%). The experiment showed that IBA 4000 ppm was the best concentration for stem cutting, sprouting and rooting in *T. chebula* (Bhardwaj et al. 1993).

Chemical examination indicate that the bark of *Terminalia paniculata* contain extractives (27%), carbohydrate (28%), lignin (33%) and ash (9.60%). No alkaloids present in the bark, but it contain tannin or tannic acid (14%). Tannin has a pyrogallol nucleus in the molecule rather than catechol nucleus along with the gallic acid (Beri and Karnik 1965)

A vegetative propagation experiment was conducted in *Terminalia chebula* to evaluate the nature of stem cuttings (tender, semi-hard and hard), seasons (in summer and monsoon) and synthetic hormone treatment (IAA, IBA and NAA) at concentrations of 500 ppm, 1000 ppm, 2000 ppm and 3000 ppm). A trial was conducted in a mist

chamber using juvenile apical stem cuttings (5 cm to 6 cm long with 1-node to 2-nodes and 2-nodes to 3-nodes) collected from 2-year to 3-year-old seedlings in the nursery. The experiment failed to root in cuttings from the old tree. But 100% of the juvenile stem cuttings are rooted before 30-days without any treatments. The study concluded that vegetative propagation in *T. chebula* is successful by rooting juvenile apical stem cuttings (Jose and Thomas 1998).

Rooting behavior of *Terminalia chebula* stem cuttings treated with synthetic hormone (IBA and IAA) in different concentrations (500 ppm, 1000 ppm, 1500 ppm and 2000 ppm) showed that IBA was more effective than IAA, and 2000 ppm was the most efficient concentration among them, with 70% rooting (Babu et al. 2018).

A study conducted in Sri Lanka recorded propagation techniques on *Terminalia chebula*. The study recorded tree characters (height, diameter at base, diameter at breast height, girth at base and girth at breast height) and fruit characters (shape, apex shape, number of ridges, diameter, length, weight, dry weight, seed weight and moisture) of *T. chebula* trees located in Galle and Matara districts of Sri Lanka. In addition, the study recorded 4 vegetative propagation (wedge grafting: *T. catappa* and *T. chebula*; patch budding: *T. catappa* and *T. chebula*; wedge grafting: *T. bellerica* and *T. chebula*; patch budding: *T. bellerica* and *T. chebula*) protocols for *T. chebula*. Air layering experiments with or without hormone treatment are not practical in *T. chebula* (Nakandalage et al. 2021).

A study was conducted in Edinburgh to analyse the effect of leaf area on the rooting of *Terminalia spinosa* in a non-mist propagation system (Newton et al. 1992). For the rooting experiment, leaves were trimmed at various levels (0, 7.5 cm², 15 cm² and 30 cm² before cuttings were severed from stock plants). 5 cm cuttings from the lateral shoots of already pruned stock plants grown in the glasshouse. The study noticed that removal of the entire leaf area prevents rooting; cuttings (with leaf area trimmed 7.5 cm², 15 cm²,

30 cm²) show 80% rooting after 3 weeks; an increase in the leaf area supports an increased rate of rooting and length of longest roots. Measures such as photosynthetic rate, stomatal conductance, water potential and relative water content of the cuttings were taken at regular intervals. The study concluded that cuttings with a 30 cm² trimmed leaf had lower relative water contents, stomatal conductance and photosynthetic rate than those of a 7.5 cm² and 15 cm² trimmed leaf.

CHAPTER 3
METHODOLOGY

Chapter 3

Methodology

3.1.Plus tree selection of *T. paniculata* in different populations of Kerala.

Plus trees are phenotypically superior in terms of growth rate, desirable growth habits, high wood quality, and resistance to diseases and pest attacks. Plus trees are identified from a population of trees using appropriate methods. Plus trees are graded against selected candidate trees, but their genetic worth has not been tested through any program like progeny testing. Before selecting plus trees, the researcher must identify the populations of the species in the study area and, from there, spot candidates for inclusion in the tree breeding program. An extensive ground survey is the best method to understand populations and population-rich regions of tree species. Regional flora, scientific reports, research publications and regional digitalized checklists are used to find the presence of the species and population-rich regions within the study area.

3.1.1. Selection of base population

The regeneration study of selected *Terminalia* in Kerala published by the Kerala Forest Research Institute gives comprehensive information regarding the *T. paniculata* population in Kerala (Chandrasekhara 2007; Pillai and Chandrasekhara 2011). Based on the literature survey, a detailed population survey was conducted throughout Kerala state.

Sl. No.	Attributes of selection	Considerations in selection	Aim
1.	Stem straightness	Little taper	Maximum volume; Minimises waste in timber production
2.	Timber height	Maximum height	Maximises potential for long lengthy timber
3.	Diameter	Little stem taper; Superior volume increment	Increase productivity and growth rate
4.	Forking	Minimum frost damage; insect; pathogen attack	Minimises waste in timber production
5.	Branch angle	Close to horizontal orientation	Less knot wood
6.	Branch thickness	Light branching	Less knot wood
7.	Self-pruning	Abscission of lower branches	Reduce the risk of stem infection; Increase timber quality
8.	Crown dimension	Large healthy crown	Maximum photosynthetic potential
9.	Fluting	Regular stem form at the base	Minimises waste in timber production
10.	Straight grain	Minimum orientation of bark	Minimise difficulty to machine work
11.	Disease	Maximum resistance	Increased productivity
12.	Epicormics	Free from epicormics	Uniform light level in the crown

Table.1. Plus tree selection attribute, considerations and aim.

Even though *T. paniculata* is a dominant commercial timber tree species, no plantations of this species have been developed so far. The forest area in the study region is administratively divided into circles and protected areas, further subdivided into forest divisions. Each forest division is further divided into forest ranges, sections, and forest

beats. The forest circles in Kerala are Northern, Eastern, Central, High range and Southern circles. Each circle is further divided into forest divisions and wildlife divisions.

In the Kerala part of Peninsular India, *T. paniculata* populations were reported from many protected areas belonging to Nilgiris and Agasthyamala biosphere reserves. All the forest regions were surveyed manually with the help of local forest staff, including Range/Deputy Range Forest Officer, Section and Beat forest officer, and Reserve and Tribal watcher. During the population survey, selectively logged forests were avoided to eliminate poor genetic materials.

3.1.2. Selection criteria and methods

Selection criteria for screening of trees included vigorous and healthy growth, a minimum height of 20 meters, straight stem, light and spreading branches, natural pruning, dense mass foliage and good flowering and fruiting, as detailed in **Table.1**. Identification of natural population stand is the first stage of the plus tree selection program through extensive population surveys throughout the study area. The survey was conducted from May 2016 to December 2017. In the field survey conducted in each forest division, populations were recorded and retagged. Identified populations were screened per the criteria proposed by Clark and Wilson (2005) and Kim et al. (2020) to select the candidate trees (**Table.2**).

Sl. No.	Selection attributes	Details of attributes
1)	Stem straightness	Stem straightness provides maximum volume for the trunk and minimizes wastage of timber during timber production. Straightness is important in the case of rural as well as in urban landscapes like fencing, wooden houses, posts, supports, utility poles, etc. Straight clear bole is most demandable as well as easy for timber production. Trees with a straight stem will be considered for the plus tree selection.
2)	Clean bole height	Long timber poles are an important preference in the commercial timber industry. Based on the length, timber is majorly divided into 3 classes such as short length logs, lengthy logs and long lengthy logs. Also, the standard of timber based on length classifies the timber into different classes in connection with the quality of logs, length of poles, etc. The maximum height provides maximum volume for the trunk and supports reaching different strata (emergent and canopy layer) to collect maximum sunlight. Since selecting the candidate plus trees, trees with maximum clear bole will be considered for the selection procedure.
3)	Diameter	Diameter is the major attribute that increases tree volume and related to productivity and growth pattern. The candidate tree shows superiority in diameter. It also minimises waste in timer production. Based on diameter, timber is classified into different classes for export quality. Terminalias normally have more than a 2-meter diameter. Since considered trees with minimum 2-meter diameter for the plus tree selection.
4)	Forking	Damage or breakage of the leading shoot results in forking and it may be due to the attack of pests or pathogen or may be due to continuous rainfall. Forking minimizes tree volume, stem straightness and maximizes waste in timber production as well as growth rate. Forking is not a desired character for plus trees, so eliminated such individuals from the selection program.

Sl. No.	Selection attributes	Details of attributes
5)	Branch angle and thickness	The branch angle is the angle formed between the main stem and the primary branch. The large branch angle high knot wood per unit length of the stem. If the branch angle is less, that will minimize the stem quality. Large branches reduce stem straightness and wood quality which also maximizes wastage during timber production. A tree with thick branches reduces demand in the timber industry. The thickness of the branch also reduces the strength of the main stem. Trees with minimum or appropriate branch angles were considered for the plus tree selection.
6)	Self-pruning	Self-pruning is a characterization process involving the detaching of shaded, diseased, or infested branches that may become a burden for the tree. Early self-pruning eases the risk of infection and increases timber quality. Hence, selected trees that show self-pruning ability.
7)	Crown dimension	The crown is the top portion of the tree including the branches from the main trunk and supports the leaves for photosynthesis. Each tree species shows different crown patterns depends on the age, height, several primary and secondary branches, etc and a healthy increase of crown length and diameter maximizes the photosynthetic activity. The trees with the healthy crown are the most desirable trees for the plus tree selection program and such trees are selected.
8)	Fluting	Fluting is the longitudinal groove in the trunk of the tree in the base, which is a type of irregular type of stem. Any kind of change in regular stem character is undesirable and reduces wood quality. Fluting severity increased with age and which limits the concentric cross-section. Since fluting is not a desirable character, trees with fluting will be eliminated from the selection.

Sl. No.	Selection attributes	Details of attributes
9)	Straight grain	Wood grains are classified into different types based on physical properties as well as aesthetic characters. Based on physical properties, wood grains are divided into many groups such as straight, spiral, interlocked, wavy and irregular, etc. Based on aesthetic properties, wood grains are divided into flat, edge, end, etc. Based on working easiness, wood with a straight grain is the best for timber production because the grain is unidirectional and parallel to the axis of the stem. Unidirectional grain increases wood quality and wood demand.
10)	Disease	The disease is the condition in which the tree shows the deviation in normal functioning. The plant shows different types of mechanisms to resist the disease and which is different from one species to another and also the nature of the disease differs. The development of a plant disease results in growth retardation and breakage of physiological activities resulting from the loss of wood quality. Superior tree individuals show maximum resistance to diseases and pests and such trees are selected.
11)	Epicormic	Epicormic shoots are the dormant buds that originated from the bark because as a result of different kinds of stress such as sudden environmental alteration, crown damage, thinning and hard pruning, root death, change in water content, cold and warm climate. The formation of epicormic shoots shows receptivity of the individual and reveals the unhealthy status of the individuals and such trees were not considered for breeding programs.

Table.2. Selection attributes and details of attributes

While comparing plus tree selection methods, Comparison Tree and Baseline methods are not suitable for populations of unknown age. Hence regression selection method is to be followed, which is selected for the identification of plus trees from a

minimum of five candidate plus trees. Based on attributes of tree selection, a minimum of five trees with maximum height and circumference at breast height were selected from each population as candidates plus trees.

So a candidate plus tree is a tree satisfying all tree selection attributes and the superior tree of the population in terms of total height and circumference at breast height. These trees selected for further tree selection and breeding programs were marked with serial numbers at breast height. The selection attributes for selecting candidates plus trees from a population are directly connected with timber quality, physiological efficiency and the healthy nature of the individual. Selection attributes are more specific to timber quantity and quality because of the economic importance of *T. paniculata*. These selection attributes will change from one species to another based on the economic significance of the species.

Measurement of selection variables

Only four tree characters were required for the selection of plus trees of a timber species from candidate plus trees, such as total tree height (H), girth at breast height (G), crown diameter (D) and crown length (L). All four were 100% quantitative characters, reducing human behaviour's effect on plus tree selection. A Haga altimeter and electronic TruPulse 200 Laser Rangefinder were used to measuring the total tree height and crown diameter. The flexible tape was used for measuring crown length and girth at breast height. The number of primary and secondary branches was determined by counting, and Garmin GPS eTex 10 was used for recording the geo-location (Latitude, Longitude and Altitude). Photographs of all plus trees were also recorded during the field study after tree selection.

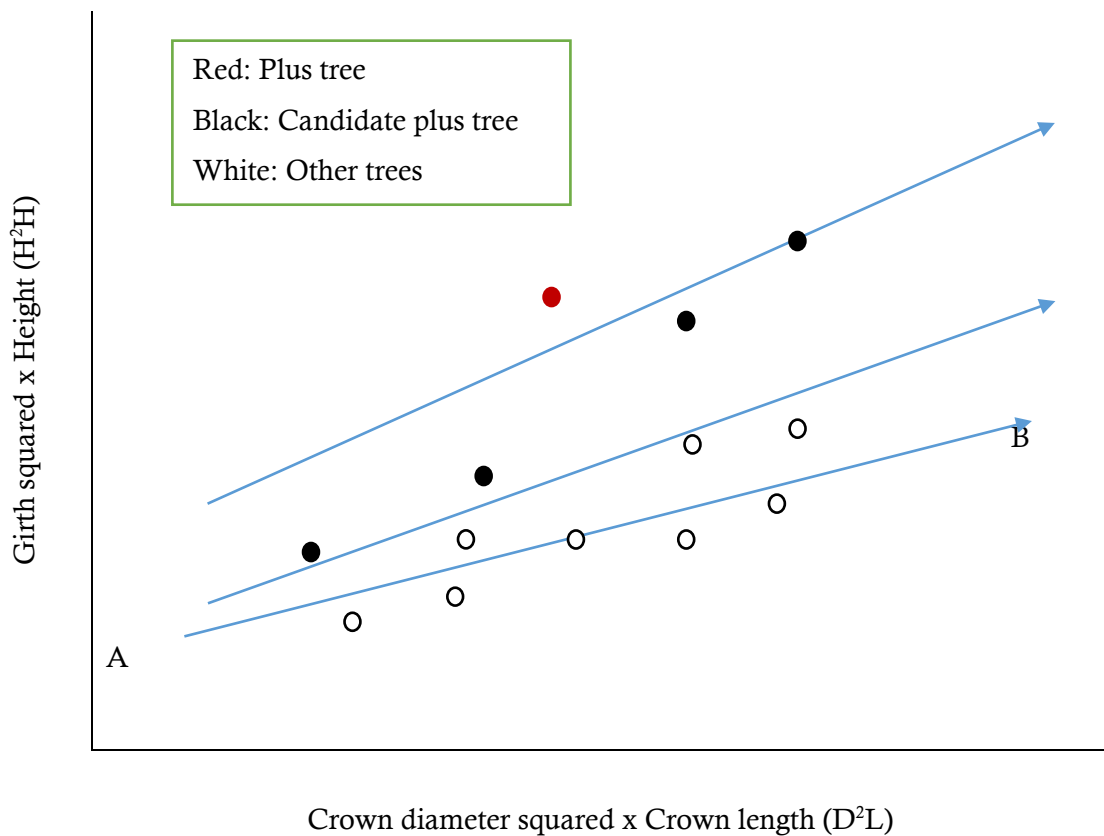


Figure.1. Regression selection method of plus tree selection

3.1.3. Regression selection method

Regression analysis was done using squared girth at breast height x total height (G^2H) as dependent and squared crown diameter x crown length (D^2L) as the independent variable. Paleontological Statistics Software (PAST) was used for carrying out linear regression analysis. Trees falling above the regression line were selected as the plus tree of that particular population. If more than one tree fell above the regression line, the tree placed far away from the line was selected as a plus tree (**Figure.1**). Regression analysis resulted in a coefficient of correlation (r) and coefficient of determination (r^2), but the method was used here to find the superior among the candidates. Therefore the analysis never recommends the documentation of coefficient of correlation (r) and coefficient of determination (r^2) for further studies.

3.2. To study the variations of *T. paniculata* in different populations of Kerala.

Most tree characters are connected with their heredity to a certain degree, while environmental factors highly modify some. The growth of trees in a particular geographical area depends on genetic and environmental factors. Several methods are available to estimate the variation between the populations of a tree species. For this study, variation between populations was assessed by analyzing phenotypic characters. Based on the nature of phenotypic characters, characters used for variation studies were categorized into four groups.

Eighteen phenotypic characters belonging to three groups were characters selected based on the literature survey for estimating the variation between the populations (Al-Sagheer and Prasad 2010; Capuzzo et al. 2012; Dangi et al. 2012; Diaz et al. 2015; Guo et al. 2017; Ashwath et al. 2020). It included characteristics that can be easily observed and measured at any season of the year independent of different phenophases (girth at breast height, total tree height, crown length, crown diameter, primary branches and secondary branches), characters that can be understood and measured only by tests (bark thickness, sapwood moisture content, sapwood density, leaf chlorophyll content, leaf area, leaf fresh mass and leaf dry mass) and characters that are not directly connected with timber production (fruit large wing length, fruit large wing width, fruit small wings width, fruit fresh mass and fruit dry mass).

Data were collected from sixteen populations of *T. paniculata* selected during the plus tree selection program. A minimum of six individuals were selected from each population including four candidates plus trees, one plus three and one mature tree and 96 (n=16 x 6) individuals for data collection. Standard protocols were followed for the selection of characters and measurement with the help of conventional instruments and tools (Wight et al. 2004; Diaz et al. 2015; Perez-Harguindeguy et al. 2016; Xu et al. 2016) (**Table.3-4; Figure.2**). The measurement of each tree character is as follows:-

Sl. No.	Name of character	Measurement
1.	Girth at breast height (GH)	Measurement of the distance around the stem measured upright to the stem axis at breast height (1.37 m). The measuring region of the stem should be free from damages and bumps, flutes and free from climbers and close contact with other trees. GH was measured at defect-free regions of the tree.
2.	Tree height (HT)	Shortest distance between the upper boundaries of the main photosynthetic tissues (excluding inflorescences) on a tree and the ground level, expressed in meters. HT was measured by moving away from the tree to a distance from where the tree tip is seen. The horizontal distance from the base of the tree was measured. The appropriate distance scale was set up in the instrument using the rotating rod. The pointer of the instrument was released by pressing the side button. The required point on the tree was sighted and the trigger was pulled after the pointer was settled. HT was directly read from the appropriate scale meters. The base of the tree was then sighted and measured. HT was measured by combining both measurements.
3.	Crown length (CL) and diameter (CD)	The crown is the top canopy part of the tree, which is characterized by branches that grow out from the main stem and this portion supports photosynthesis. CL is the vertical length of the tree excluding the clear bole. CL of the tree was measured by subtracting the clear bole length from the total height of the tree. CD is also a measure of the crown portion of the tree. For measuring CD, distance from the base of the tree up to the tip of branches in four directions (south, north, east, west) was measured. The average of these measurements was taken as the CD.
4.	Primary (PB) and secondary branches (SB)	The number of branches included both PB and SB. PB originated from the tree trunk and SB originated from PB. Both PB and SB were counted manually with the help of binoculars.

Sl. No.	Name of character	Measurement
5.	Stress wave time (ST)	Time taken for stress wave to travel in a unit distance (here 1 m) through wood is the ST. Tree Sonic Microsecond Timer is the instrument used for estimating the ST. The instrument consists of two sensors connected to a timer and the sensors fixed on the wood in a unit distance at 45 ⁰ angles to measure the ST. Stress is induced by hitting the top of the start sensor by a hammer. Stress wave formed in start sensor will be collected by the stop sensor in the other end. ST will appear on the timer screen.
6.	Bark thickness (BT)	The bark is the outermost layer of the stem which is composed of outside dead tissues and inside live tissues (vascular cambium). BT was measured at breast height and a portion of the bark was removed from the stem using mechanical tools in square shape and thickness was calculated.
7.	Sapwood moisture content (SM) and density (SD)	Sapwood is the outermost portion of the woody stem composed of living cells and is different from heartwood by colour. Sapwood portion of the stem in square shape was collected after removing the bark using mechanical tools (chisel and hammer). The fresh and dry mass of sapwood was estimated by using the weighting balance. A hot air oven was used for drying the sample. SM was calculated as the difference between the fresh weight and oven dry weight. Sapwood volume was estimated by the water displacement method and the density of the sapwood was calculated by dividing the dried mass of the sapwood by the volume of the sapwood.
8.	Leaf area (LA)	The area of a leaf is the most common metric for leaf size and is defined as the one-sided or projected area of an individual leaf. LA was determined using a LI-3100C leaf area meter.
9.	Leaf chlorophyll content (LC)	LC was measured using an instant SPAD (Soil-Plant Analysis Development) 502 plus chlorophyll meter. Before measuring the LC, the instrument was calibrated. Measurements were taken from the leaf lamina where leaf veins are not prominent. Six

Sl. No.	Name of character	Measurement
		measurements were taken from a single leaf and the average value was taken as the LC.
10.	Leaf (LF, LD) and fruit mass (FF, FD)	For estimating the leaf fresh (LF) and dried mass (LD), leaves are directly collected from plants without the loss of moisture content and brought to the laboratory. Fresh and oven dried mass of the leaf and estimated using weighting balance. Fruit fresh (FF) and dried mass (FD) were also determined similarly.
11.	Fruit wings (LL, LW, SL)	Fruit characters such as fruit large wing length (LL), large wing width (LW) and small wings length (SL) are other important characters of tree species especially in the case of winged fruited species. These measurements were taken using a using Vernier Calliper.

Table.3. Measurement details of tree characters of *T. paniculata* in Kerala.



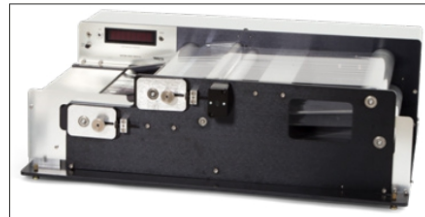
A



B



C



D



E

Figure.2. Instruments used for measuring tree characters
(A: Rangefinder; B: Altimeter; C: Chlorophyll Meter; D: Area meter; E: TreeSonic Timer).

Sl. No.	Name of character	Instruments/ Tools	
		Sample collection	Measuring
1.	GH		Measuring tape
2.	CD		
3.	HT		TruPulse 200 Laser Rangefinder;
4.	CL		Haga altimeter
5.	PB		
6.	SB		Binocular
7.	ST		TreeSonic Microsecond Timer
8.	BT	Chisel; Hammer	Mitutoyo 530-118 Vernier callipers
9.	SM	Chisel; Hammer	Kern Analytical balance ABT 100-5NM; BSSSCO Hot air oven 24 x 18 x 18 S.S. chamber with digital temperature controller
10.	SD	Chisel; Hammer	BSSSCO hot air oven 24 x 18 x 18 S.S. chamber with digital temperature controller
11.	LC		SPAD 502 Plus Chlorophyll meter
12.	LA		LI-3100C Area meter
13.	LF		Kern Analytical balance ABT 100-5NM;
14.	LD	Polythene bags	BSSSCO Hot air oven 24 x 18 x 18 S.S. chamber with digital temperature controller
15.	LL		
16.	LW		Mitutoyo 530-118 Vernier callipers
17.	SL		
18.	FF		Kern Analytical balance ABT 100-5NM;
19.	FD	Polythene bags	BSSSCO Hot air oven 24 x 18 x 18 S.S. chamber with digital temperature controller

Table.4. Instruments and tools used for the collection and measurement.

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass).

3.2.1. Univariate summary statistics

Univariate summary statistics were done to identify basic statistical information about the data, such as minimum, maximum, mean, standard error, standard deviation, skewness, kurtosis, geometric mean and coefficient variance. Basic statistical information about the data helps to understand the measure of dispersion, the extent of the shape of the distribution and an estimate of statistical dependence. PAST software is used for doing Univariate summary statistics (Hammer et al. 2001).

3.2.2. Analysis of Variance (ANOVA)

To estimate significant variation within morphological characters, the statistical model ANOVA was carried out using IBM SPSS statistics software (version 26: 2018) (IBM 2019). Data regarding morphological tree characters help to confirm the existence of variation between populations. Tree phenotypic characters are subjected to ANOVA.

3.2.3. Correlation Analysis

Correlation analysis was carried out using IBM SPSS statistics software (version 26: 2018) to estimate the relationship and strength between the characters used for variation studies. Correlation analysis helps state negative or positive, strong or weak, significant or non-significant relationships between the characters. Here correlation analysis resulted in a correlation coefficient, which indicates the impact of variation of a character on another.

3.2.4. Principal Component Analysis (PCA)

Principal component analysis (PCA) was carried out using Minitab software to reduce the complexity of high-dimensional data without the loss of trends and patterns of data. PCA helps to summarise the high-dimensional data into a compressed form. Also, principal component analysis results in the list of main characters among the total characters used for the study. Principal characters are the tangible assets to studying

variation between populations (Abdi & William 2010). The principal component analysis is one of the statistical methods to determine the variation in data by defining the minimum number of principal components. Three methods are used to assess variation using principal component analysis such as eigenvalue, proportion and scree plot. Eigenvalue explain how many principal components determine the interpretation, and a principal component with an eigenvalue of more than one (>1) is the real asset of the variability.

Proportion indicates how much a principal component accounts for the variation in data. The ratio of variance components helps determine the level of conflict in data depending on the application. For the descriptive purpose, 80% of the variance explained is needed. For further data analysis needs, 90% of the conflict. A Scree plot is a simple graphical representation that helps to determine how many principal components have eigenvalue above 1, accurately describing the variability.

3.2.5. Cluster Analysis

Cluster analysis is a pattern of grouping the whole data into different clusters based on similarities within the data (Plotkin et al. 2002; Suranto 2002; Li-Hammed et al. 2015; Abozeid et al. 2017). In the present study, all the selected populations were grouped into clusters based on morphological tree characters. Cluster analysis of tree populations helps to indicate the similarity between the populations and find out the most similar populations on the whole. Dendrogram, a type of hierarchical clustering (paired group UPGMA algorithm; Euclidean similarity index) constructed using PAST software.

3.3. To study the seed characteristics and seed handling techniques.

3.3.1. Fruit production and maturity index

The study was confined to trees from the Peechi Wildlife Division. As part of the plus tree selection program, two large populations of trees were located in the Vallikkayam forest section of the Peechi Forest Range, Peechi Wildlife Division. Five mature trees were selected for the study. Twelve inflorescences from each tree were marked; thus, sixty inflorescences were marked and labelled during flowering initiation. Periodical weekly observations were done from flower initiation to fruit maturation, covering the entire floral phenophases. Maturity evaluation of fruit observations started from the beginning of fruit formation, and each stage of the phenophases was recorded regularly (Srimathi et al. 2013; Barroso et al. 2016).

Fruits were collected during different stages of fruiting phenophases. The Munsell colour chart determined the colour of fruits and seeds (Dranski et al. 2010; Reeder et al. 2014). The collection of fruits from each tree was done manually. Fruits (n= 1000 x 8) collected during different stages of maturity were subjected to a germination test. Seed emptiness and germinability were estimated and recorded. Germination trials were conducted in germination trays (LBH= 30 cm x 40.50 cm x 7 cm) filled with vermiculite in laboratory conditions at room temperature.

3.3.2. Seed Characteristics Experiments

According to the world agroforestry centre (ICRA), there are five seed source classes: natural forests, plantations, farmland seed sources, seed orchards and vegetative propagules. In Kerala, *T. paniculata* populations were restricted to natural forests only, and there are no plantations or seed orchards of this species. During the plus tree selection program, a maximum number of plus tree populations (6/16; 37.50%) were identified from Karulai Forest Range, Nilambur South Forest Division. Samples for the solo study about the timber characteristics of *T. paniculata* were collected from Karulai

Forest Range by Forest Research Institute Dehra Dun (Jain and Dangwal 1985; Muthukrishnan and Swaminath 1999).

Karulai Forest Range (265.608 sq. km) was the source for seed characters study of *T. paniculata*, the border with North Nilambur Forest Division in the north, Kalikavu Forest Range and Silent Valley National Park in the west and Mukurthi National Park, Tamil Nadu in the south and eastern side (Latitude 11° 17'; Longitude 76° 21').

3.3.3. Seed Collection and Cleaning

Seeds were collected from six plus trees located at Karulai Forest Range for seed germination experiments (**Figure.3**). The reproductive phase in this species starts during the last week of September and ends during March. Fruiting begins in the previous week of December and ends during March. Mature seeds were collected from selected plus trees from January 2017 to February 2017. Seeds were manually collected from the trees from each tree from all four directions (north, east, west and south). Six separate wet white cotton cloth bags for each tree were used for collecting and transporting seeds to the Kerala Forest Seed Centre. Following observations were recorded for each bag.

- Name of collector
- Type of Source
- Place of Collection
- Collection method
- Altitude in meter
- Collection bag
- Period of collection
- Type of seed
- Number of bags
- Distance from the collection site to the laboratory

Infected and damaged seeds were removed and cleaned adequately at Kerala Forest Seed Centre. Seed adjuncts, broken, rotten seeds and other materials were removed during cleaning. Cleaning includes aspiration, scalping and grading. The entire cleansing operation was done manually. Cleaned seeds were stored in air-tight thick plastic containers.

3.3.4. Seed Morphometry and Moisture Content

Morphometry of cleaned seeds was recorded using Vernier callipers and electronic weighing machines. Morphometry of seeds includes the length of wings (large middle 1 and 2 small side wings), the width of the large middle wing and length of seeds, the fresh mass of seeds, and the dry mass of seeds and seeds per kilogram. The gravimetric method using drying in hot air drying was used to estimate seed moisture content. Seeds were dried in a hot-air oven for 1 hr at 130°C. Moisture content (MC; in %) was calculated as follows:-

$$MC = (M2 - M3) \times 100 / (M2 - M1)$$

Where

M1 is the weight of the container with a cover (g), *M2* is the weight of the container with its cover and seed before drying (g) and *M3* is the weight of the container with its cover and seed after drying (g).

3.3.5. Seed Viability

Seed viability is the measure of live seeds which can develop into plants. A germination test and cutting test were done to test viability. As per International Seed Testing Association (ISTA), 800 (n=100 x 8) is the standard sample for germination tests of small size seeds. But because of high seed emptiness, 8000 (n=1000 x 8) seeds were used for the experiments (Ribeiro-Oliveira et al. 2016). A manual cutting machine is used for seed cutting. The presence and absence of seeds within the fruits were recorded per standard methods. For germination experiments, seeds were sown in plastic trays (LBH=

30 cm x 40.5 cm x 7 cm) filled with vermiculite and maintained in the laboratory at room temperature (25°C). Data were recorded from the start of germination to culmination as per standard methods.



Figure.3. Collection of seeds of *T. paniculata* from Karulai Forest Range

3.3.6. Germination Testing

Randomly selected cleaned seeds ($n=1000 \times 8$) were sown in plastic trays (LBH= 30 cm x 40.5 cm x 7 cm) filled with vermiculite and kept in laboratory condition at room temperature. Data were recorded from commencement of germination to culmination, such as germination percentage, initial germination, final germination, and germination duration as per standard methods (Xu et al. 2016).

- $GI = DGI - DSS$
- $GF = DGC - DSS$
- $GD = GF - GI$
- $GP = \frac{TS}{TSS} \times 100$
- $MDG = \frac{GP}{GD}$

Where

- GI is the initial germination,
- DGI is the initial germination day,
- DSS is the seed sowing day,
- GF is the final germination,
- DGC is the germination culmination day,
- GD is the germination duration,
- GP is the germination percentage,

TS is the total number of germinants,
TSS is the total number of seeds sown and
MGD is the mean daily germination.

3.3.7. Pre-sowing Treatments

Even though the fruit of *T. paniculata* is very small, the fruit is winged like most Terminalias such as *T. cuneata*, *T. elliptica*, *T. myriocarpa*, etc. Due to the small size and high- emptiness, selected seeds (n=1000 x 8) were collected during the 16th-week of fruit development for germination experiments. Different pre-treatments used for the study were as follows (**Table.5**):-

Treatment	T1	T2	T3	T4	T5	T6	T7
Pre-sowing treatment	Control			Water soaking			
Duration (in hr)	Nil	12	24	48	12	24	48
Seed sowing medium	Vermiculite						
Seed type	Winged			De-winged			

Table.5. Pre-sowing treatments on *T. paniculata* seeds

De-winging fruits were done using regular scissors, and tap water was used to soak the seeds. Treated seeds were sown in vermiculite-filled plastic trays (LBH= 30 cm x 40.5 cm x 7 cm), kept under laboratory conditions, and sprayed with the tap water daily. Germination patterns were recorded from the germination commencement to culmination and determined the initial day of germination, germination duration and percentage as per standard methods (Xu et al. 2016).

Treatment	T 01	T 02	T 03	T 04	T 05	T 06	T 07	T 08	T 09	T 10	T 11	T 12	T 13	T 14	T1 5
Temperature (°C)	04	16	25	04	16	25	04	16	25	04	16	25	04	16	25
Storage container	Jute sack			Polythene cover			Cotton sack			Aluminium container			Thick plastic container		

Table.6. Seed storage experiments in *T. paniculate*

3.3.8. Seed Longevity and Storage

For seed longevity and storage experiments, cleaned seeds of *T. paniculata* were stored under cold conditions (4°C, 16°C and 25°C) in an air-tight container (jute sack, polythene cover, cotton sack, aluminium metal container and thick plastic container). A total of fifteen-treatments were used to study seed longevity (**Table.6**). Seed longevity was tested at monthly intervals from the second month after seed collection. The germinability of seeds (n=1000 x 8) was measured by sowing in plastic trays filled with vermiculite kept in laboratory condition at room temperature. Tap water was used to water the trays daily to keep the vermiculite wet.

3.4. Development of a protocol for propagation in *T. paniculata*.

3.4.1. Propagation using seeds

Seeds collected from six populations of Karulai Forest Range (TPP3, TPP4, TPP5, TPP6, TPP7 and TPP8) of Nilambur South forest divisions of Kerala during 2017-2018 were used for the study. Seeds were collected during the 16th week of fruit development for germination trials. Fruits during the 16th week of fruit development were identified with the help of the Munsell soil colour chart, and the collection was done directly from trees manually. Seeds were packed in separate wet cotton bags to minimize moisture loss and transported to Kerala Forest Seed Centre. The seeds were cleaned manually and kept in plastic containers.

Cleaned seeds (winged and de-winged) were subjected to germination trials using different mediums (soil, sand, vermiculite, soil-sand mixture, soil-vermiculite mixture and sand-vermiculite mixture). Plastic trays (LBH= 30 cm x 40.5 cm x 7 cm) filled with different sowing media were used in the study. Tap water was used for watering the trays. The germinability of winged or wingless seeds sown in the various mediums was recorded daily. Germination percentage, duration and mean daily germination was calculated using the standard formula (Xu et al. 2016).

- $GI = DGI - DSS$
- $GF = DGC - DSS$
- $GD = GF - GI$
- $GP = \frac{TS \times 100}{TSS}$
- $MDG = GP / GD$

Where

GI is the initial germination,

DGI is the initial germination day,

DSS is the seed sowing day,

GF is the final germination,

DGC is the germination culmination day,

GD is the germination duration,

GP is the germination percentage,

TS is the total number of germinants,

TSS is the total number of seeds sown,

MGD is the mean daily germination.

3.4.2. Propagation using vegetative cuttings

A vegetative propagation experiment was conducted at the mist chamber of Kerala Forest Research Institute. Cuttings were taken from the matured trees located at the campus. The leafless cuttings about 15 ± 2 cm in length and 1.5 ± 0.5 cm in diameter, having 4 to 5 buds, were used for the experiment. The basal ends of the cuttings were kept in 0.20% carbendazim 50% WP (Bavistin), an anti-fungal solution, for two minutes to prevent fungal infection. Cuttings were treated with synthetic rooting hormone (IBA) in different concentrations (500 ppm, 1000 ppm, 1500 ppm and 2000 ppm) via two methods (**Table.7**).

The dipping method was done by dipping the cuttings in synthetic hormones after soaking in an anti-fungal solution. The soaking process was done by soaking the basal end of the cuttings in synthetic hormones solutions for 12 hr. The cuttings after each hormone treatment were planted in the polythene bags (LB= 25 cm x 15 cm) filled with wood charcoal mixed vermiculite. One-third of the stem cuttings were inserted in the rooting medium and maintained in a controlled mist propagation unit. The experiment was laid out as three factors nested effect factorial experiment in randomized block design with synthetic hormone as treatment factor, concentration as a nested factor into treatment factor and species as the third factor. Each treatment had 30 (n= 10 x 3) cuttings and a total of 480 (n= 10 x 3 x 16) cuttings were planted in the experiment.

Hormonetreatment	Dipping method				Soaking method			
Treatment and Conc. of IBA (ppm)	T1 500	T2 1000	T3 1500	T4 2000	T5 500	T6 1000	T7 1500	T8 2000
Treatment and Conc. of NAA ppm)	T9 500	T10 1000	T11 1500	T12 2000	T13 500	T14 1000	T15 1500	T16 2000

Table.7.Rooting experiments of stem cuttings of *T. paniculata*

Treatment	T01	T02	T03	T04	T05	T06	T07	T08	T09	T10	T11	T12
Method of IBA application	Dipping (powder)				Dipping (liquid)				Soaking overnight (liquid)			
IBA (ppm) Conc.	500	1000	1500	2000	500	1000	1500	2000	500	1000	1500	2000

Table.8. Rooting experiments of *T. paniculata* epicormics treated with IBA.

3.4.3. Propagation using epicormic shoots

A vegetative propagation experiment was conducted at the mist chamber of Kerala Forest Research Institute. Shoots-free stump cuttings (1 m long) were collected from matured trees of the Kerala Forest Research Institute campus and kept under a mist chamber in polythene bags filled with wood charcoal mixed vermiculite. Tap water was sprayed daily to keep the vermiculite moist. Sprouting occurred after fourteen-days, and epicormic shoots at the four-leaf stage were collected using scissors. These epicormic shoots were soaked in an anti-fungal solution (50% WP carbendazim) for two minutes and treated with IBA (500 ppm, 1000 ppm, 1500 ppm and 2000 ppm). IBA treatment was applied using three different methods (dipping method in powder), soaking in a liquid formulation for two minutes and soaking in liquid formulation overnight) (Table.8).

3.4.4. Propagation through air-layering.

Air layering was tried from six mature trees on the Kerala Forest Research Institute campus. Four branches from four directions from each tree were selected for air layering. Forty-eight ($n= 4 \times 4 \times 6$) branches were subjected to a layering experiment. A terminal branch with a 1 cm to 2 cm diameter was selected from each branch for air layering. Using secateurs and a penknife, the bark was removed without any damage to the stem and the coir pith-sand-cow dung mixture was pasted over the bark removed from the branch. The entire portion was covered with polythene tapes and tied with nylon thread to hold the mix over the stem and reduce contamination. This portion was sprayed daily using tap water to minimize evaporation and keep it wet until root hairs emerged.

CHAPTER 4
RESULTS AND DISCUSSION

Chapter 4

Results and Discussion

4.1. Plus tree selection of *T. paniculata* in different populations of Kerala.

Based on a review of the literature (research reports, academic works, etc.), an extensive population survey was carried out from Kasargode to Thiruvananthapuram districts of Kerala, including territorial forest circles (Southern, High Range, Central, Eastern and Northern) and wildlife circles (Agasthyavanam Biological Park and Palakkad). As the result of population analysis, thirty locations were selected across Kerala based on the number and size (total tree height and girth at breast height) of trees within an area of 100 meters (**Table 9**).

Selected populations were highly variable regarding the number of individuals and the size of individuals from Kasargode to Trivandrum. For a tree breeding program, the phenotype of the individuals is a significant factor. Based on tree selection parameters such as stem straightness, clear bole length, crown diameter, forking, branch angle and thickness, self-pruning, crown dimension, fluting, pests and diseases and epicormics, 16 populations were sorted out with five mature individuals (**Table 10**). Even though *T. paniculata* is one of the most dominant tree species in the state with wide distribution, the ideal population of the species limited to seven Forest Ranges of the state belonging to six forest divisions.

Sl. No.	Forest Division	Forest Range	Locations
1.	Kasargode	Kasargode	1. Parappa
2.	Kannur	Kannavam	2. Kannavam
3.	Calicut	Thamarassery	3. Thamarassery Ghats
4.	Calicut	Malabar	4. Peruvannamuzhi
5.	Nilambur South	Karulai	5. Nedumkayam 6. Mundankadavu Colony 7. Mundankadavu Timber Depot 8. Chakkikuzhi 9. Pattakarimba Junction 10. Pattakarimba Colony
6.	Mannarkkad	Mannarkkad	11. Anamuli
7.	Palakkad	Walayar	12. Kava 13. Vattappara 14. Malampuzha 15. Puthussery
8.	Parambikulam	Parambikulam	16. Kallippara 17. AlmaramVayal 18. Almaram Junction 19. Parambikulam Tunnel
9.	Parambikulam	Sungam	20. Thunakadavu 21. Anappadi Bit 1 Vayal 22. Anappadi Watchtower 23. Thellickal
10.	Thrissur	Pattikkad	24. KFRI Campus
11.	Peechi	Peechi	25. Jandamukku 26. Olakara
12.	Kottayam	Erumely	27. Kombukuthi
13.	Thenmala	Aryankavu	28. Aryankavu
14.	Trivandrum	Neyyar	29. Cherupunnathodu
15.	Trivandrum	ABP	30. Kottur

Table 9. Healthy populations of *T. paniculata* in Kerala.

These 16 populations belonged to Karulai (Nilambur South Forest Division), Walayar (Palakkad Forest Division), Sungam and Parambikulam (Parambikulam Wildlife Division), Peechi (Peechi Wildlife Division), Erumely (Kottayam Forest Division) and Aryankavu (Thenmala Forest Division) Forest Ranges of Kerala. Except for the population in the Aryankavu Forest Range, all other populations are located in remote areas, and few are far from non-tribal settlements. These populations belonged to moist deciduous (Walayar, Parambikulam and Sungam), evergreen (Karulai and Aryankavu) and semi-evergreen (Peechi and Erumely) forest types. Among 16 populations, two (Peechi Forest Range) were very close to the Kerala Forest Research Institute campus. Code (TPP: *T. paniculata* population) was given to each population in alphabetical order of the Forest Ranges as TPP01 for Aryankavu Forest Range and TPP16 for the Walayar Forest Range.

Sl. No.	Population	Range	Location	Latitude	Longitude	Altitude (in m a.sl)
1.	TPP01	Aryankavu	Aryankavu	08.97283	77.15061	240
2.	TPP02	Erumely	Kombukuthi	09.48811	76.95854	414
3.	TPP03	Karulai	Nedumkayam	11.29777	76.33330	068
4.	TPP04	Karulai	Mundankadavu	11.30805	76.37500	073
5.	TPP05	Karulai	Mundankadavu	11.30800	76.37305	073
6.	TPP06	Karulai	Pattakarimba	11.27500	76.38638	084
7.	TPP07	Karulai	Pattakarimba	11.27500	76.38472	084
8.	TPP08	Karulai	Chakkikuzhi	11.26833	76.35888	068
9.	TPP09	Parambikulam	Almaram	10.41213	76.80645	660
10.	TPP10	Parambikulam	Almaram	10.40935	76.81005	693
11.	TPP11	Parambikulam	Kallippara	10.41869	76.80121	692
12.	TPP12	Peechi	Jandamukku	10.51666	76.37391	096
13.	TPP13	Peechi	Olakara	10.51444	76.43944	143
14.	TPP14	Sungam	Thellickal	10.45083	76.74444	530
15.	TPP15	Sungam	Anappadi	10.41764	76.79482	570
16.	TPP16	Walayar	Puthussery	10.86087	76.79849	236

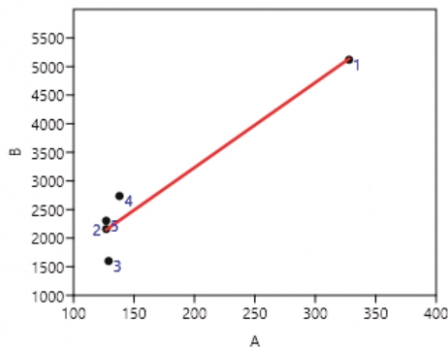
Table 10. Details of selected populations of *T. paniculata* in Kerala.

Tree No.	GH (in m)	TH (in m)	CD (in m)	CL (in m)	PB (in nos.)	SB (in nos.)
TP01	3.20	32	16	20	04	11
TP02	2.30	24	12	15	06	10
TP03	2.42	22	10	16	04	08
TP04	2.40	24	12	19	06	10
TP05	2.40	22	12	16	04	08
TP06	3.30	33	22	12	03	08
TP07	2.45	28	16	16	05	10
TP08	2.30	29	10	18	04	08
TP09	2.60	27	14	16	05	10
TP10	3.15	36	22	14	05	12
TP11	3.65	32	24	12	06	14
TP12	3.37	28	20	13	07	10
TP13	2.98	24	22	13	15	08
TP14	2.98	22	20	11	05	12
TP15	3.10	24	18	14	06	14
TP16	3.59	30	21	12	04	17
TP17	3.28	32	20	11	07	19
TP18	3.18	32	18	12	05	11
TP19	3.26	31	20	12	07	18
TP20	2.90	30	14	12	06	16
TP21	3.38	34	18	12	05	12
TP22	2.53	32	16	14	04	11
TP23	2.51	33	12	12	04	09
TP24	3.58	32	16	12	05	14
TP25	3.06	32	20	13	07	13
TP26	3.64	34	22	12	04	06
TP27	3.59	30	20	10	06	18
TP28	3.55	31	26	10	06	14
TP29	2.35	23	21	10	04	08
TP30	2.94	26	18	14	03	08
TP31	3.31	31	26	10	07	14
TP32	2.63	26	16	10	02	08
TP33	3.68	33	22	13	04	14
TP34	2.88	26	24	10	04	10
TP35	3.00	29	20	16	04	10
TP36	2.60	27	12	17	03	08
TP37	2.88	29	20	15	04	08
TP38	3.67	34	20	14	04	12
TP39	3.12	30	20	13	04	12
TP40	3.31	33	22	13	06	12
TP41	3.25	33	17	13	07	14
TP42	2.30	34	18	12	06	12
TP43	2.60	32	16	16	07	14
TP44	2.10	31	16	12	07	16
TP45	2.75	31	16	14	06	12
TP46	3.10	32	24	11	08	16
TP47	2.25	33	18	18	06	11
TP48	2.80	32	16	17	07	14

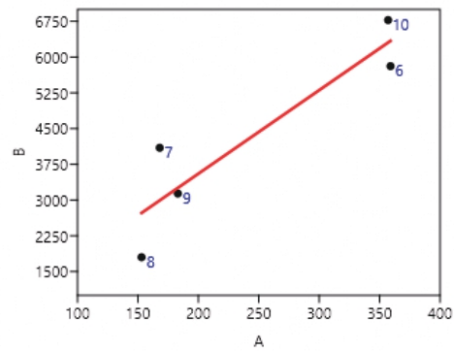
Tree No.	GH (in m)	TH (in m)	CD (in m)	CL (in m)	PB (in nos.)	SB (in nos.)
TP49	2.50	32	14	18	06	12
TP50	2.80	30	16	17	06	12
TP51	2.70	28	18	10	06	12
TP52	2.30	28	14	16	05	10
TP53	3.20	28	15	17	05	10
TP54	3.04	31	14	18	05	10
TP55	2.78	28	13	18	06	11
TP56	3.35	32	16	17	10	15
TP57	2.45	25	16	17	04	08
TP58	2.94	31	20	10	05	10
TP59	2.40	24	14	15	04	11
TP60	2.46	26	16	10	05	11
TP61	3.30	32	24	14	06	12
TP62	3.10	32	24	12	04	10
TP63	2.50	32	24	10	09	16
TP64	2.80	26	22	14	06	14
TP65	3.20	30	22	08	05	10
TP66	2.63	33	16	16	06	12
TP67	2.65	27	13	13	04	07
TP68	1.80	26	12	10	04	08
TP69	1.72	24	12	13	05	10
TP70	2.80	31	14	15	06	12
TP71	3.20	32	23	15	07	14
TP72	3.10	31	22	16	06	16
TP73	2.60	27	24	11	06	15
TP74	2.69	30	14	11	05	12
TP75	3.10	31	18	11	06	12
TP76	2.80	26	16	14	06	14
TP77	2.70	24	14	12	07	14
TP78	2.30	24	13	14	04	08
TP79	2.15	23	14	12	04	08
TP80	2.30	22	16	10	05	10
Mean	2.90	29.10	17.70	13.50	5.50	11.60
SD	0.50	3.60	4.10	2.70	1.80	2.90

Table.11. Characteristics of *T. paniculata* candidate plus trees in Kerala
(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, SD: Standard deviation).

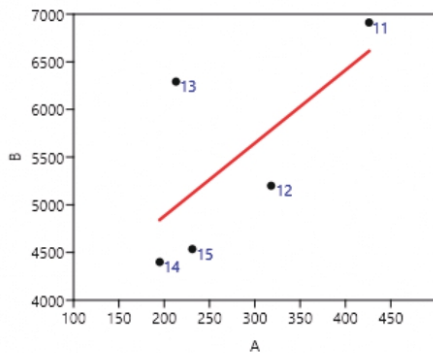
According to the Soil Health Information System of Kerala, the entire state is classified into five agro-ecological zones: the coastal plain, midland laterites, foothills, high hills and Palakkad plains and each zone are divided into several units. Above mentioned ideal populations were located in two agro-ecological zones viz Foot Hills (Walayar and Erumely) and High Hills (Parambikulam, Sungam, Karulai, Peechi and Aryankavu). These seven Forest Ranges belonged to the four agro-ecological units of Northern High Hills (Karulai and Peechi), Northern Foot Hills (Walayar), and Southern High Hills (Parambikulam, Sungam and Aryankavu) and Southern and Central Foot Hills (Erumely).



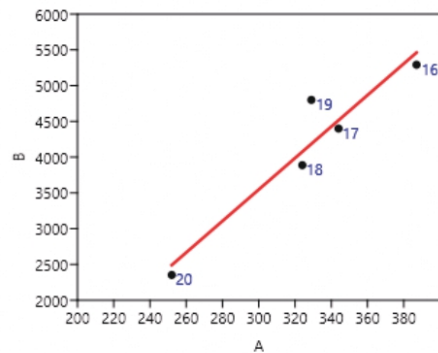
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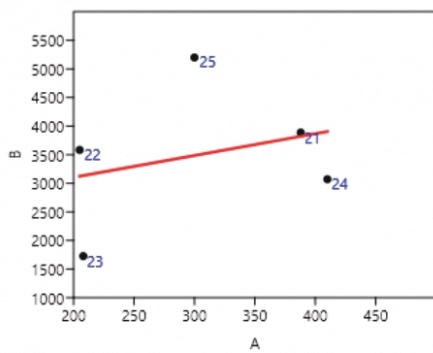
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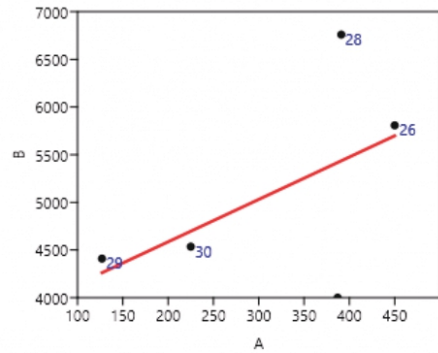
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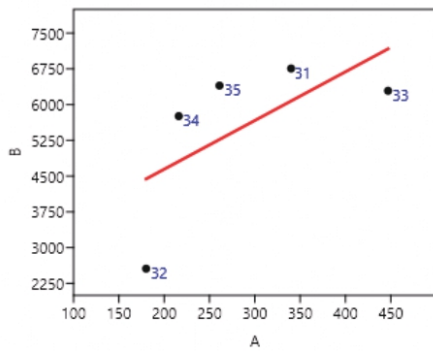
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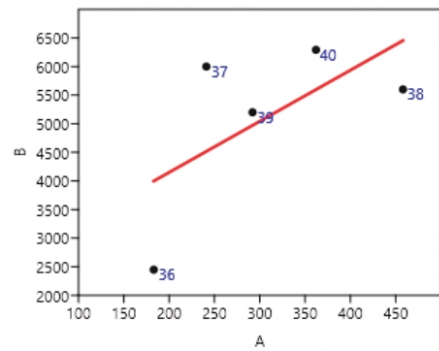
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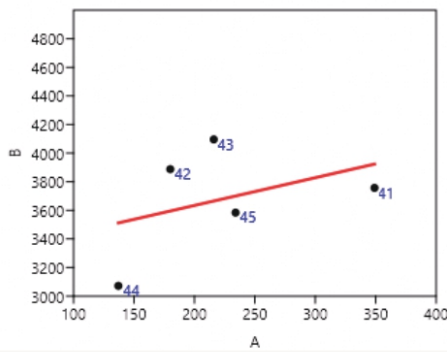
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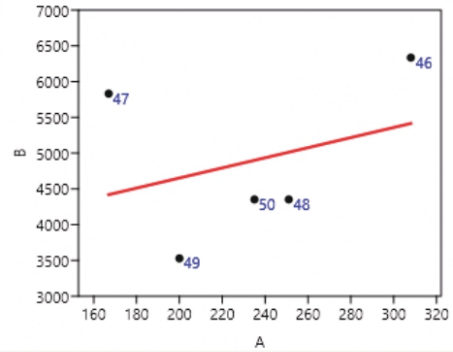
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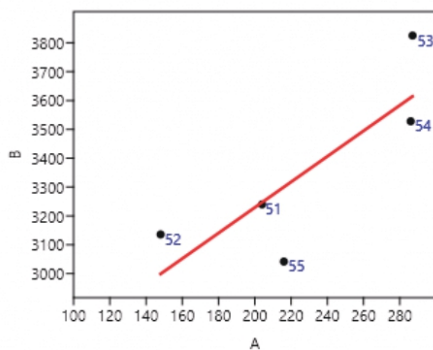
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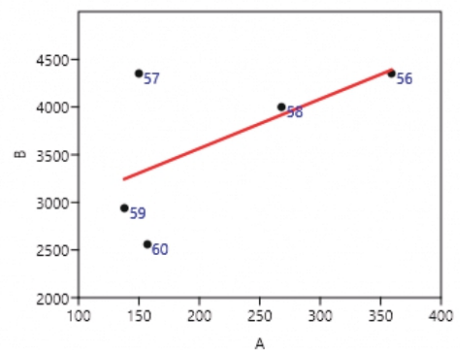
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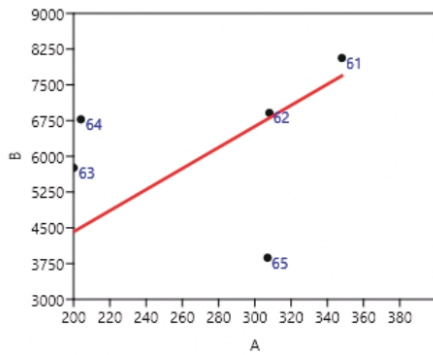
TPP 10



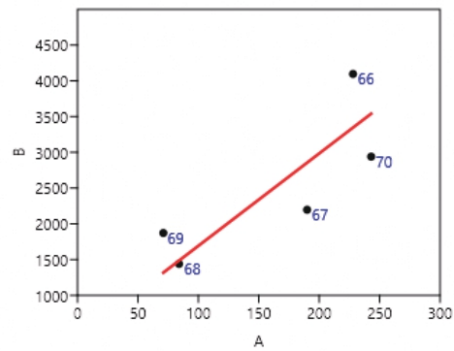
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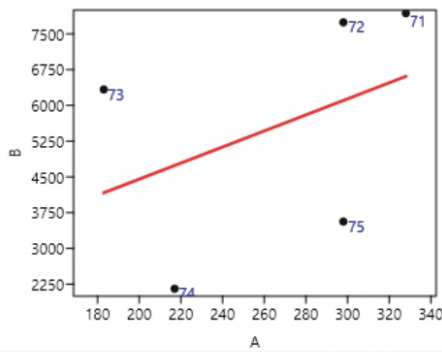
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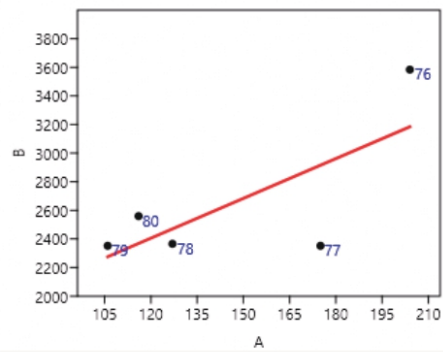
TPP 13



TPP 14



TPP 15



TPP 16

Figure.4. Regression method of plus tree selection in *T. paniculata* (A: G2H; B: D2L)

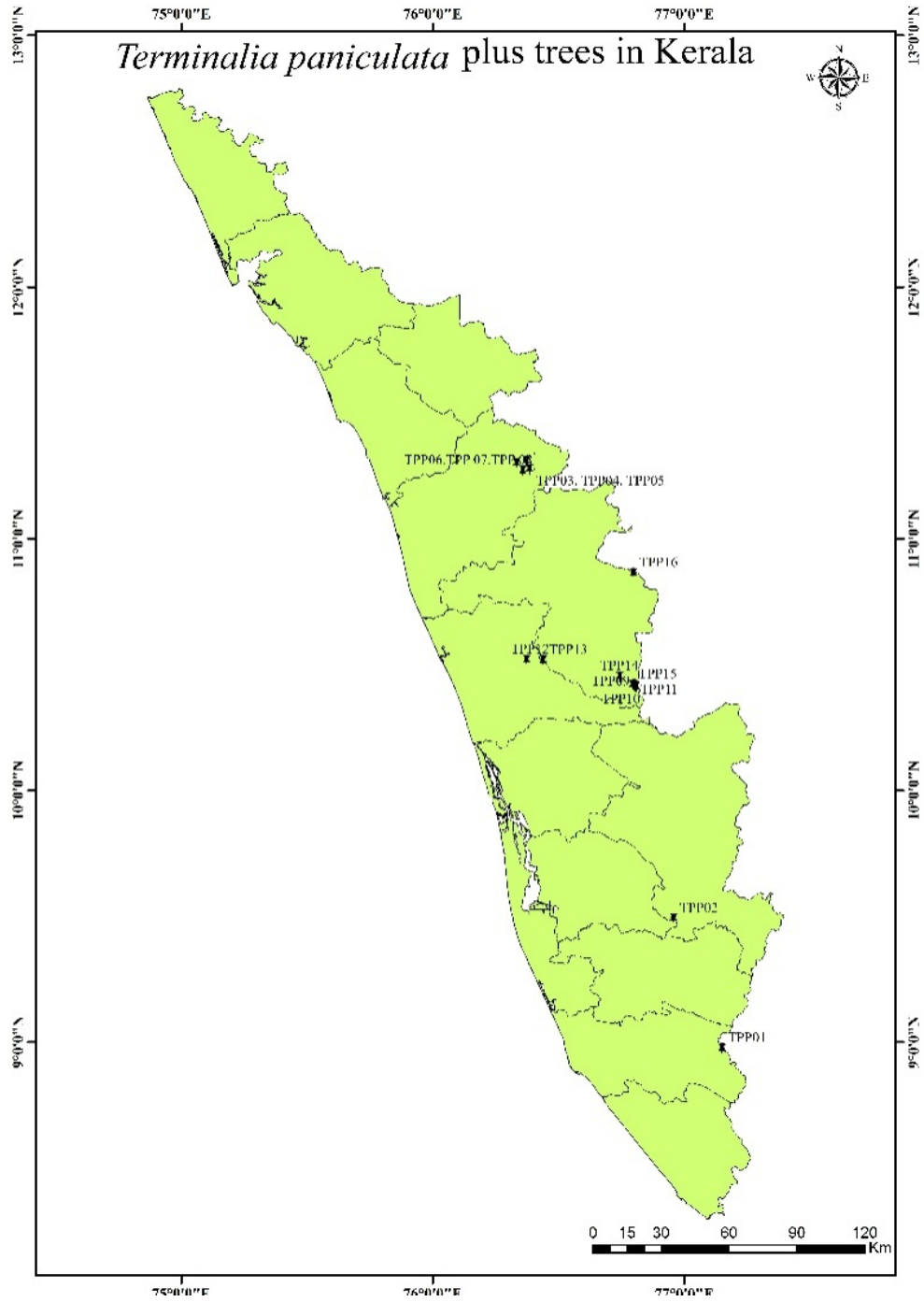


Figure.5. Map of plus tree locations of *T. paniculata* in Kerala



Figure 6. Plus trees located in Aryankavu (TP4), Erumely (TP7) and Karulai Forest Ranges (TP13 and TP19) of Kerala

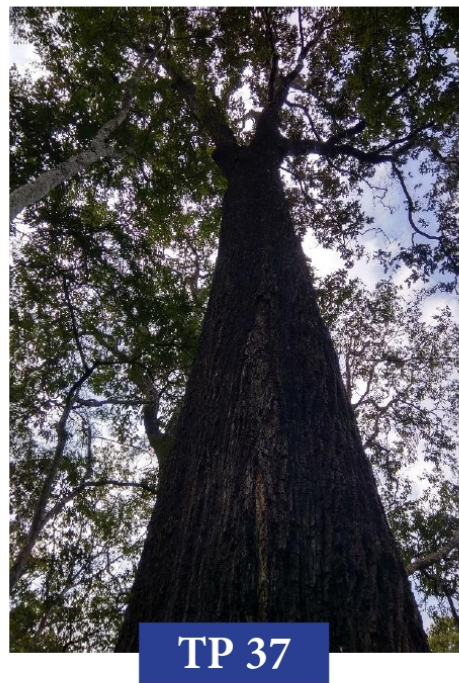
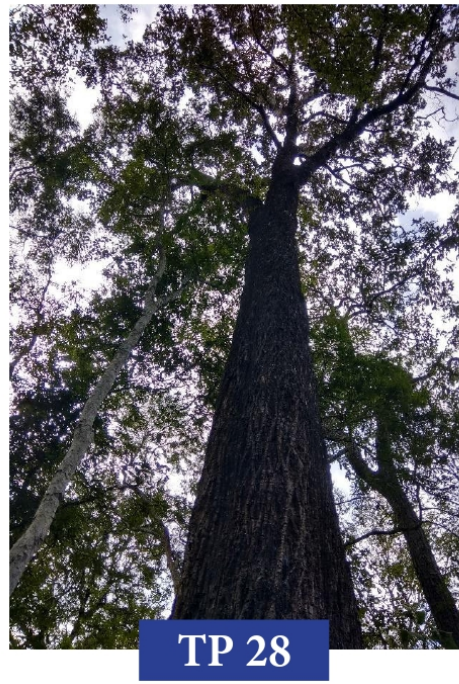


Figure 7. Plus tree located at Karulai Forest Range (TP25, TP28, TP35 and TP37) of Kerala



Figure 8. Plus trees located in Parambikulam (TP43, TP47 and TP53) and Peechi Forest Ranges (TP57) of Kerala.

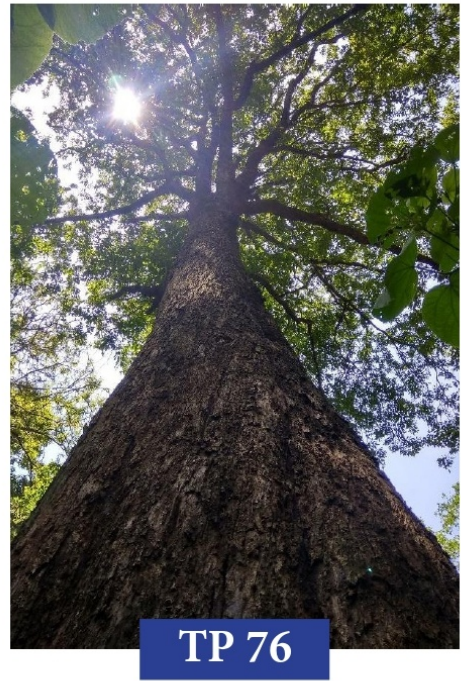


Figure 9. Plus trees located at Peechi (TP64), Sungam (TP66 and TP73) and Walayar (TP76) forest ranges of Kerala.

Sl. No.	Tree No.	Girth at breast (in m)	Tree girth (in m)	Crown diameter (in m)	Crown length (in m)	Primary branches (in no.s)	Secondary branches (in no.s)
1.	TP04	2.98	24	22	13	15	08
2.	TP07	2.88	29	20	15	4	08
3.	TP13	2.45	25	16	17	4	08
4.	TP19	2.40	24	12	19	6	10
5.	TP25	2.45	28	16	16	5	10
6.	TP28	3.00	29	20	16	4	10
7.	TP35	3.20	28	15	17	5	10
8.	TP37	2.25	33	18	18	6	11
9.	TP43	2.63	33	16	16	6	12
10.	TP47	3.06	32	20	13	7	13
11.	TP53	3.55	31	26	10	6	14
12.	TP57	2.60	32	16	16	7	14
13.	TP64	2.80	26	22	14	6	14
14.	TP66	2.80	26	16	14	6	14
15.	TP73	2.60	27	24	11	6	15
16.	TP76	3.26	31	20	12	7	18
	Mean	2.80	28.60	18.70	14.80	06.30	11.80
	S.D.	0.40	03.10	03.70	02.50	02.50	02.90

Table.12. Characteristics of *T. paniculata* plus trees located in Kerala

Each population selected a minimum of five morphologically superior candidates plus trees for other plus tree selection programs. Each CPT was coded (TP: *Terminalia paniculata*) concerning its Forest Range and numbered from 1 to 5 for the five CPT of each population. Hence, the first five (TP1 to TP5) candidates plus trees belong to Aryankavu Forest Range (TPP1), and the final five (TP76 to TP80) CPTs belong to Walayar Forest Range (TPP16).

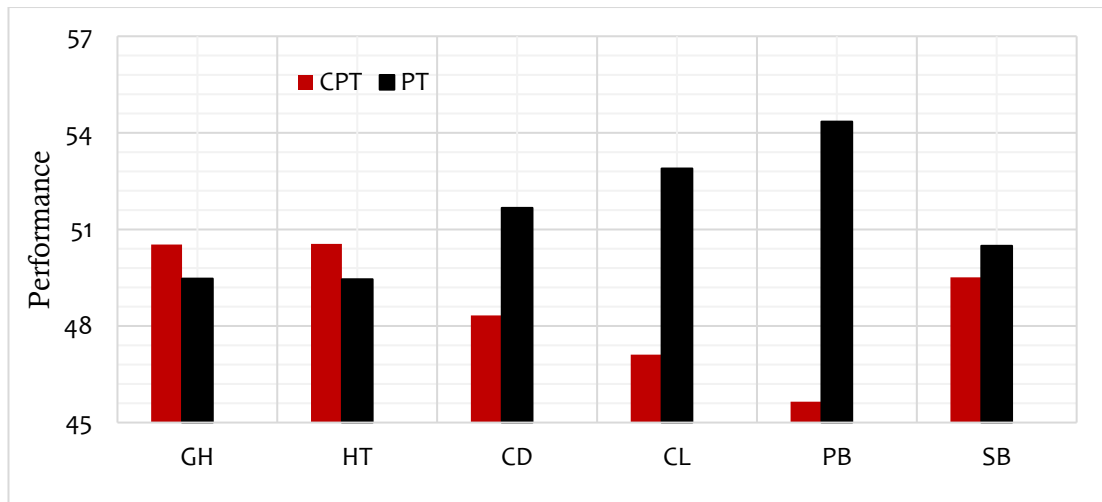


Figure.10. Characteristics of *T. paniculata* CPT and PT located in Kerala (CPT: candidate plus tree; PT: plus tree; GH: girth at breast height; HT: total height; CD: crown diameter; CL: crown length; PB: no. of primary branches; SB: no. of secondary branches)

A total of 80 candidates plus trees (TP1 to TP80) designated from 16 (TPP1 to TPP16) selected populations were measured, and the tree characters were recorded (Table.11). Candidate plus trees characteristics such as tree height (between 22 m and 36 m; $\bar{x}=29.14\pm3.59$ m; $p=0.003$), girth at breast height (between 1.72 m and 3.68 m; $\bar{x}=2.86\pm0.45$ m; $p=0.001$), crown diameter (between 10 m and 26 m; $\bar{x}= 17.73\pm4.05$ m; $p=0.001$), crown length (between 8 m and 20 m; $\bar{x}= 13.51\pm2.68$ m; $p=0.001$), primary (between 2 and 15; $\bar{x}= 5.45\pm1.76$; $p=0.046$) and secondary branches (between 6 and 19; $\bar{x}= 11.63\pm2.86$; $p=0.004$) were recorded.

Regression selection was conducted to determine the best phenotypically superior individuals from the respective populations by comparing the candidate plus trees using statistical techniques (Figure.4). Regression selection of candidates plus trees resulted in the identification of 16 plus trees from 16 locations (Figure.5-9). The regression selection selected one superior individual from each population. Among the 16 plus trees, the tree girth ranged between 2.25 m and 3.55 m ($\bar{x}= 2.81\pm0.36$ m), and total tree height ranged between 24 m and 34 m ($\bar{x}= 28.63\pm3.12$ m). The crown diameter and crown length of the plus trees were varied between 12 m and 26 m ($\bar{x}= 18.69\pm3.70$ m) and between 10 m and

19 m ($\bar{x}= 14.81\pm 2.54$ m), respectively. The number of primary and secondary branches of *T. paniculata* plus trees ranged between 4 and 15 ($\bar{x}= 6.25\pm 2.54$) and between 8 and 18 ($\bar{x}= 11.81\pm 2.90$), respectively (**Table.12**).

While comparing plus trees and candidate plus trees of *T. paniculata* in Kerala, candidate plus trees showed supremacy over girth at breast height and total tree height (**Figure.10**). Girth at breast height of CPTs was 1.10%, and total tree height is 1.10% higher than plus trees. While in the case of other characters such as crown diameter, crown length, number of primary and number of secondary branches, plus tree were superlative over candidate plus trees. The plus tree's crown diameter and length were 3.30% and 5.80% higher than candidate the plus trees. Similarly, the number of primary and secondary branches of the plus tree was 8.90% and 0.90% higher than candidate plus trees.

Advantages of tree improvement are the stable genetic makeup of the trees, more effortless vegetative propagation, and a wide range of environmental adaptability. Disadvantages of tree improvement programs are the requirement of large land area for testing genetic material, large-sized tree individuals throughout the breeding activities, great effort for variety release, and significant period for documentation of tree breeding program. Even though tree improvement has many advantages, it has a few unique difficulties. The particular problems of tree improvement are the long-time to take for the initiation of seed production, examining the genetic character of trees only through progeny testing, the uncertainty of the existence of desirable characters in progenies, requirement of a continuous experiment to confirm the transfer of genetic material from mother trees to progenies, the need of a long period for a study regarding reproductive characteristics, inadequate of knowledge about forest trees, etc.

Tree improvement program includes:-

1. Selection of species, or geographic source of the species within a specific region.
2. Assessment of variation within the species within a specific region.
3. Development of packages for improved individuals with desirable characters.
4. Production of improved individuals for large scale plantation practises.
5. Conservation and maintenance of base population for further tree improvement programs.

4.1.1. Species Selection

Forest tree breeding is evolving very fast due to the need to rejuvenate the degraded forest lands and meet the challenges of extensive deforestation and habitat loss. Tree breeders have long realized that a large amount of genetic variation in the essentially wild tree species they work with presents them with selection opportunities not shared by most crop and animal breeders (Lindquist 1948). *T. paniculata* is the species selected for the tree improvement/ breeding program and the study covers species selection, provenance selection, plus tree selection, variation assessment, seed biology and vegetative propagation. Like all other tree breeding programs, the program's purpose is to meet the demand for timber of *T. paniculata* through tree improvement approaches. Tree improvement of *T. paniculata* is significant because of the endemism, distributional range, high seed emptiness, production of many seeds, winged fruit character, large tree form, multipurpose usage, dominant nature in geographic range, etc.

4.1.2. Provenance Selection

T. paniculata is endemic to Peninsular India, recorded from Maharashtra, Karnataka, Kerala and Tamil Nadu states. These regions are categorized as West Coast Plains and Hills agro-ecological zone. But the species is commonly distributed in Kerala and Karnataka parts of Peninsular India. Geographic distribution points out that the species is dominant in the Kerala part of Peninsular India, so Kerala is selected as the

provenance for tree improvement/ breeding program. Compared to Tamil Nadu, Karnataka and Maharashtra states of Peninsular India, *T. paniculata* is dominant in all districts of Kerala. The topography of Kerala indicates that most of the land area is between 0 and 650 m a.s.l, and the natural habitat is composed of evergreen, semi-evergreen and deciduous forest lands. The southwest and northeast monsoon highly influences the climatic condition of Kerala state. Due to high anthropogenic pressure, the natural populations of *T. paniculata* in home gardens and agricultural lands drastically declined. Therefore populations of *T. paniculata* are restricted to evergreen, semi-evergreen and deciduous forest lands.

4.1.3. Mass Selection (Plus Tree Selection)

Plus tree selection is the beginning of tree breeding, and different methods scholars adopt for selecting plus trees. The Source of individuals may be wild population strands, plantations, home gardens, etc., based on the phenotypic assessment of characters of economic interest. Selection of plus phenotype based on desired characters is the preliminary step for a mass scale multiplication program (Clark and Wilson 2005). Productive stands of *T. paniculata* are mostly uneven-aged and distributed on a wide range of trees throughout the south-western part of Peninsular India. Determining the best selection techniques depends on several factors, including species characters, stand characteristics, history, and variability and inheritance pattern of essential aspects and objectives of the selection program (Chauhan et al. 2018). Selection of the best genotypes with the ideal combination of characters can give a sustained gain in terms of yield per hectare per year and return of money. Therefore, it is essential to study the relationship of different contributing characters to the main character and look for the ideal combination of characters in the tree (Singh and Choudhary 1985).

Major tree breeding program of forestry trees, including genus *Terminalia* conducted in India, is done through the comparison tree method. The comparison tree

method is based on many variables depending on the tree species' significance and importance. This method is mainly followed in even-aged stands of a single dominant species growing under comparatively uniform cultural and site conditions (Sidhu 1993; 1996). Criteria of selection and the number of variables are contingent on the ecological critical of the species and the behaviour of the tree breeder. Since the choice of candidate plus tree is made from existing uneven-aged populations of *T. paniculata* growing in scattered forms in the forest area of Kerala, candidate plus tree selection was done according to the criteria proposed by Clark and Wilson (2005). This study uses tree selection through the regression method proposed by Rudolf (1956). Compared to the comparison tree method, the regression method of plus tree selection is purely determined by four variables: tree height, girth, crown diameter and length. Also, regression analysis is suitable for both aged-known and aged-unknown tree individuals and is independent of human behaviour.

Currently, these enormous forest lands transformed into fragmented forest lands and crop plantations due to anthropogenic activities. Hence tree breeding programs are essential in a highly populated country like India to meet timber demand in the household, industrial and tertiary sectors. Ongoing tree breeding programs in India focused on economically significant trees such as *Ailanthus*, *Azadirachta*, *Dalbergia*, *Eucalyptus*, *Gmelina*, *Melia*, *Pinus*, *Pongamia*, *Salix*, *Tectona*, *Terminalia*, etc. Most of the tree breeding programs were conducted in India and completed through the comparison tree method. Regression selection and baseline method are not carried out yet in India (Wilcox 1982; Gupta 1999; Sharma et al. 2001; Kumar et al. 2003; Raut et al. 2005; Yadav et al. 2005; Carrasquinhio et al. 2010; Qader et al. 2014; Chauhan et al. 2018; Daneva et al. 2018).

Plus tree selection program on *T. chebula* and *T. bellerica* was conducted in India based on morphological and biochemical characteristics such as tree height, canopy

diameter, base diameter, breast height diameter, fresh fruit weight, fruit diameter, fruit length, dry fruit weight, fruit pulp weight, seed weight, seed moisture, total soluble solids, acidity, total sugar, reducing sugar and non-reducing sugar (Cheturvedi and Khanna 1982; Navhale et al. 2011; Khobragade et al. 2013). The classical formula proposed by Rudolf (1956) was used for the same in the present study.

Regression selection method opted for the *T. paniculata* tree selection because:-

1. No plantations are developed yet and age of individuals is unknown.
2. Difficult to collect large number of variables from extreme geographic range in forest lands within limited period of time from large number of individuals.
3. Regression analysis is based on statistical backgrounds and only four variables are required for the selection program
4. Independent from the behaviour of tree breeder or geneticist.

In Spain, Sierra proposed an idea for the selection of plus trees of pine, in which diameter at breast height and the age of the tree were considered. In Pine (*Pinus* sp.), branch characters are an essential factor affecting the grade and value of the sawn wood, which was also considered as the selection criteria (Jansons 2009). Characters like height, height up to first living branch, diameter at breast height, slenderness, the diameter of the thickest branch up to 2-m height, length of the living crown, average branch diameter and stem volume were used in the study. Among these characters, height, diameter at breast height and length of the live crown were used in the research and found to be important characters in the regression analysis. The selection of characters may differ mainly depending upon the end use of the species in question.

The selection of economically important tree species like Malabar Neem (*Melia dubia*) characteristics like good stem form, good growth characters, well-formed crown and natural pruning ability and free from pests and diseases were used. The individuals having diseases, dead branches, or attacked by any pathogen and pests were rejected in

the initial selection stage. The same method was adopted to leave trees during candidate plus trees. Major economic characteristics for the candidate plus tree selection were the stem straightness, cylindrical clear bole, girth at breast height; tree height and branch angle (Chauhan et al. 2018). Similarly, the characters based on apparent growth, crown length, stem straightness, and the feelings of priority were selected in different tree species. Neem (*Azadirachta Indica*), Northern Rosewood (*Dalbergia sissoo*), Indian Beach (*Pongamia pinnata*) (Yadav et al. 2005).

However, to make the selection efficient, Hazel and Lush (1942) emphasized consideration of the 1) extent of genetic variation present in the population (relative economic value of character), 2) heritability of the characters, and 3) genetic and environmental correlation of each character with the other. Considerable variability in growth characteristics like total tree height, circumference at breast height, crown diameter, crown length, number of primary branches and number of secondary branches was observed in the scattered population, reflected evidently in the growth data of selected trees. The heritability of those selection characters is the second important consideration while deciding the selection criteria, as suggested by Hazel and Lush (1942). Stem straightness and roundness are known to directly relate to the wood quality and easy handling in processing.

Even a simple selection of tree forms can improve the quality and quantity of products (Shelbourne 1969; Zobel and Talbert 1984). Hence, selecting candidates plus trees was preliminary based on stem form. Tree improvement following selection is dependent on the method of selection used, initial heritability, selected proportion, family number and size (Wei and Lindgren 1991). So, the characters selected and the methodology adopted for plus tree selection plays a vital role in the tree breeding program. Since the characters chosen for the candidate plus tree selection show a high degree of heritability, the regression analysis method is most suitable for selecting plus trees from this candidate plus tree.

The present study identified 16 plus tree populations of *T. paniculata* from Kerala. It makes sense that even though the species is dominant throughout Kerala, only 16 plus tree populations have been recorded from the state. Among the 16 plus tree populations, six belong to Karulai Forest Range, and five belong to Parambikulam Wildlife Division. Similarly plus tree selection program in *Tectona grandis* indicates that plus tree populations were limited to narrow provinces of Nilambur (five sites) and Thrissur (one site) only. Similarly plus tree selection program on *Ailanthus excelsa* in the entire Haryana, Rajasthan and Gujarat confirmed that twenty-one plus tree populations were restricted to three provinces (Old Hisar, Haryana; Sikar, Rajasthan and Surat, Gujarat) only.

Indian rosewood (*Dalbergia sissoo*) plus tree selection program confirmed that plus tree populations were restricted to the Northern part of Himachal Pradesh and Uttar Pradesh. All these studies reveal that plus tree population is particular to certain pockets in the whole distributional range. Plus tree selection programs point out that whether the species is dominant or not in provenance, populations for tree breeding were restricted to limited provinces only. These plus tree populations can be kept as resource material for other tree breeding programs. It is helpful for variation analysis between the populations, propagation protocol developments and other studies (reproductive biology, wood technology, physiology, biotechnology, conservation biology and growth monitoring, etc.) to get authentic information regarding the species and its environs.

As mentioned earlier, this selection ends with documentation of the provenance, distributional status, plus trees and corresponding populations, morphometric measurements of the species, suitable agro-ecological zone for the growth, locations for further provenance trials, etc. This particular tree selection program is a unique plus tree selection program on *T. paniculata* worldwide, more precisely in Peninsular India, through the regression selection method.

4.2. To study the variations of *T. paniculata* in different populations of Kerala

4.2.1. Tree Characters

Variation studies are diverse in trees according to the characteristics (nature, number, dependant on a particular season, measured by tests, qualitative, quantitative and direct relationship with timber production) selected for the study, methods (biochemical, morphological, molecular, anatomical and physiological), the objective of the study, the geography of the study area, number of samples taken, period of the study, climatic factors, the behaviour of the scholar, etc. The present study estimated the variation between populations of *T. paniculata* according to morphological characters. Morphological characters used here for variation estimation included tree form, wood, leaf and fruit characters.

Variation between the populations throughout the world is analyzed by studying the phenotypic and molecular characteristics. For variation analysis using phenotypic characteristics, different types of characters were selected by scholars over some time. Generally, stem, leaf, fruit and seed characteristics were taken by the scholars to estimate the variation between the populations (Bhat and Priya 2004; Indira 2006; Li et al. 2007; Al-Sagheer and Prasad 2010; Raut et al. 2010; Shankar and Synrem 2012; Kumar and Dhillon 2014; Paray et al. 2017; Li et al. 2018; Ashwath et al. 2020; Dolley et al. 2020), etc. The present study included variation analysis between the population in terms of morphological characteristics such as total tree height, girth at breast height, crown length, crown diameter, number of primary branches, number of secondary branches, bark thickness, stress wave time, sapwood moisture content, sapwood density, leaf chlorophyll content, leaf area, leaf fresh mass, dry leaf mass, fruit large wing length, fruit large wing width, fruit small wings length, fruit fresh mass and fruit dry mass.

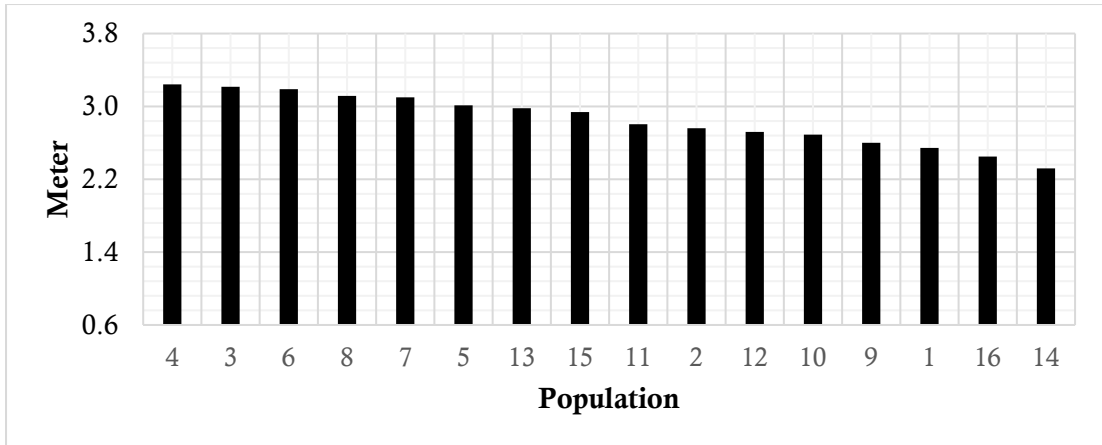


Figure 11. Girth at breast height of *T. paniculata* candidate plus trees in Kerala

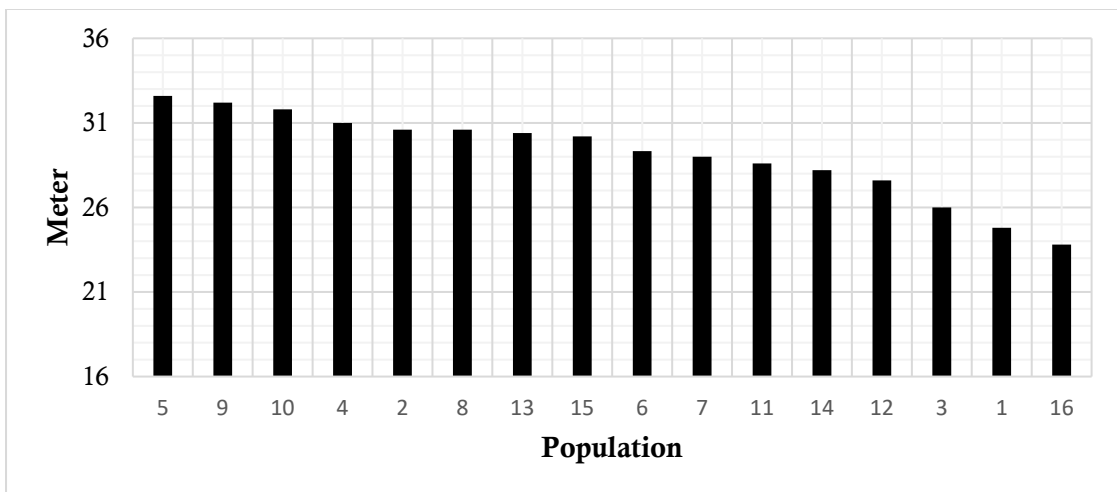


Figure 12. Total tree height of *T. paniculata* candidate plus trees in Kerala

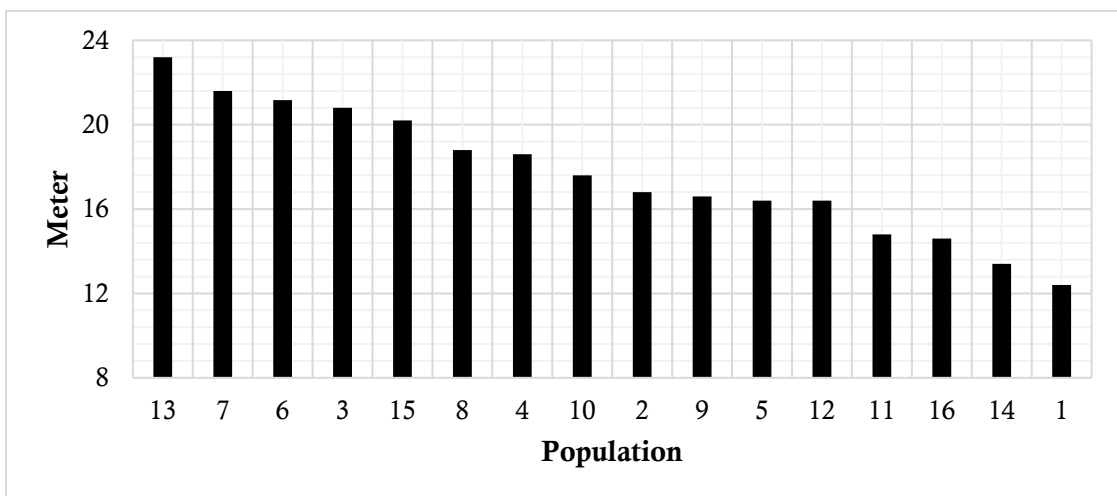


Figure 13. Crown diameter of *T. paniculata* candidate plus trees in Kerala

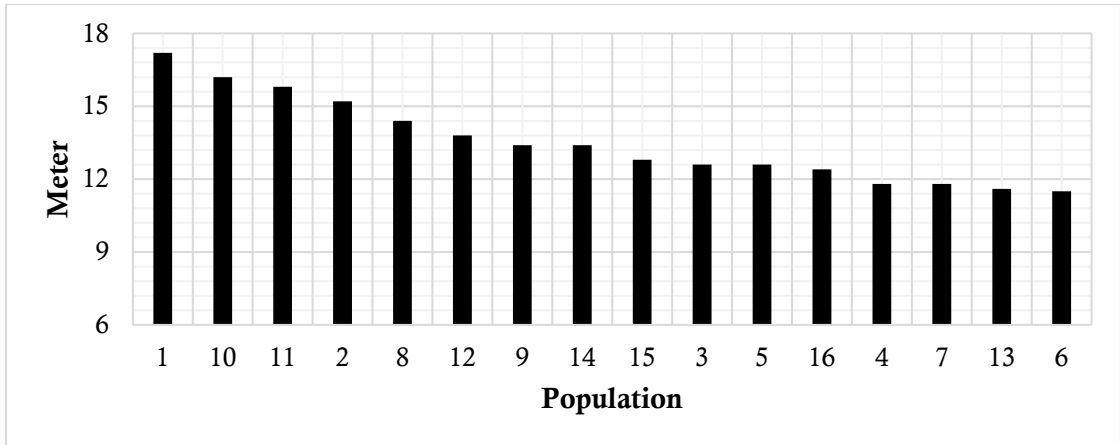


Figure 14. Crown length of *T. paniculata* candidate plus trees in Kerala

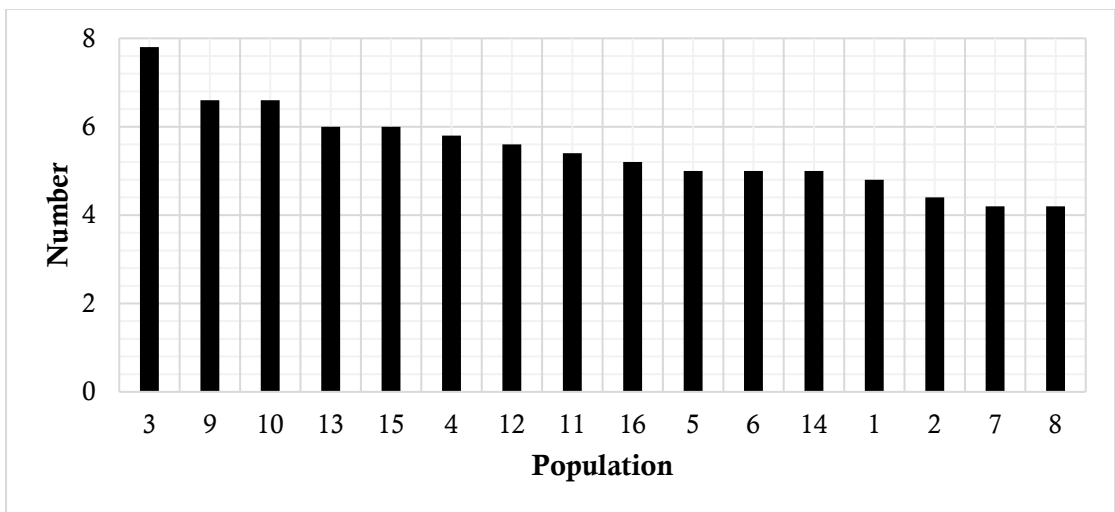


Figure 15. Number of primary branches of *T. paniculata* trees in Kerala

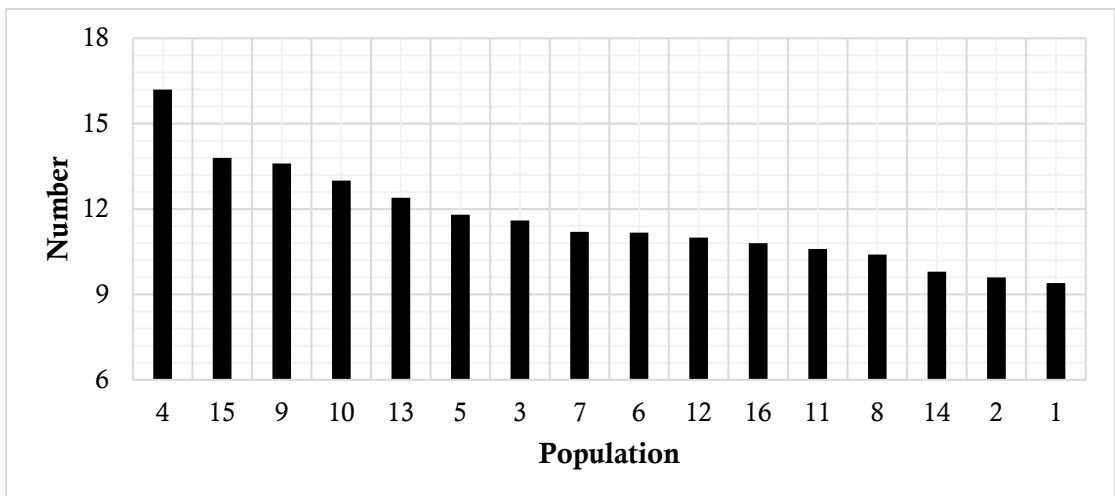


Figure 16. Number of secondary branches of *T. paniculata* trees in Kerala

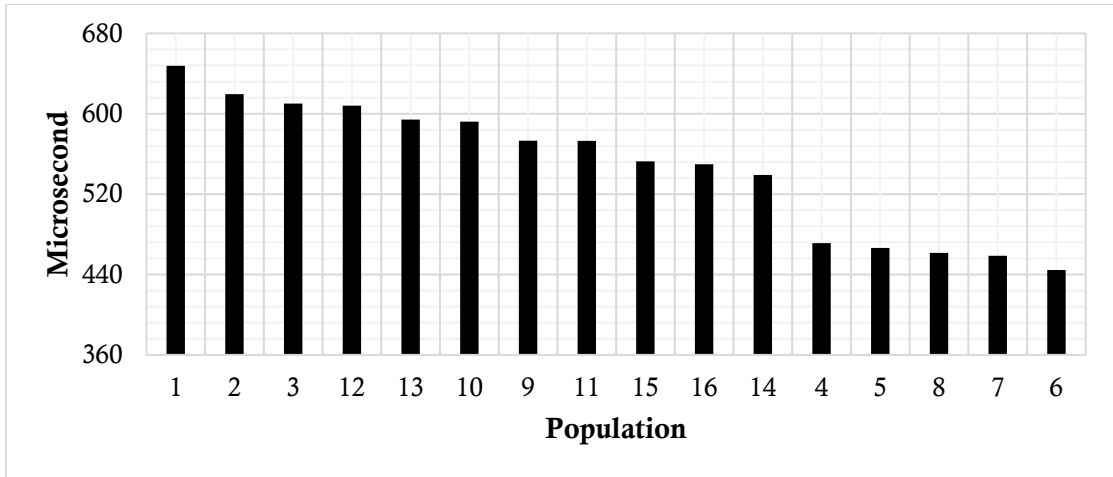


Figure 17. Stress wave time of *Terminalia paniculata* trees in Kerala

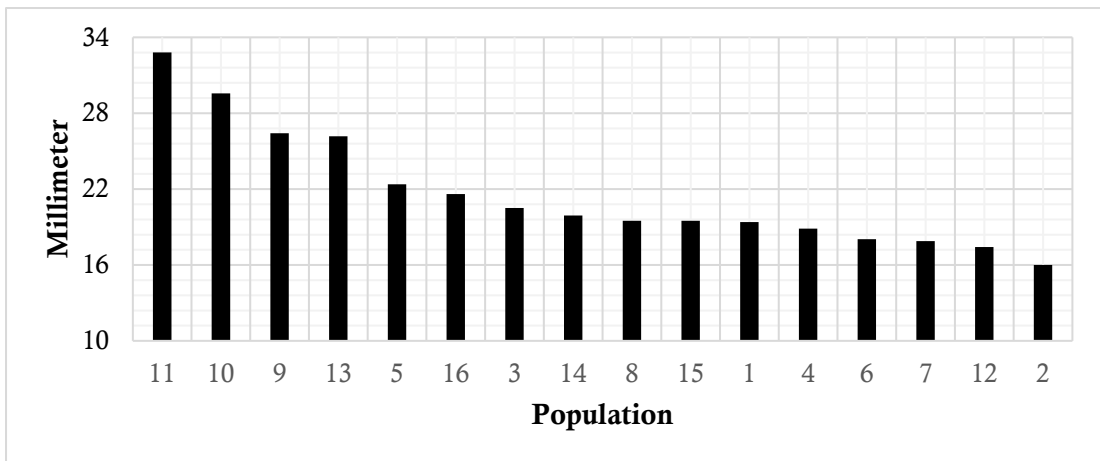


Figure 18. Bark thickness of *T. paniculata* candidate plus trees in Kerala

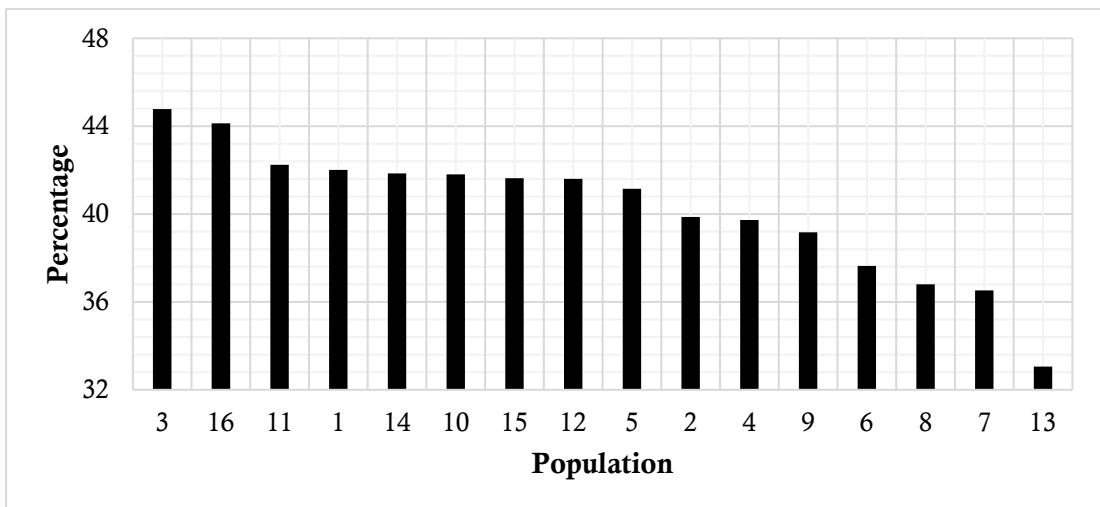


Figure 19. Sapwood moisture content of *T. paniculata* candidate plus trees in Kerala

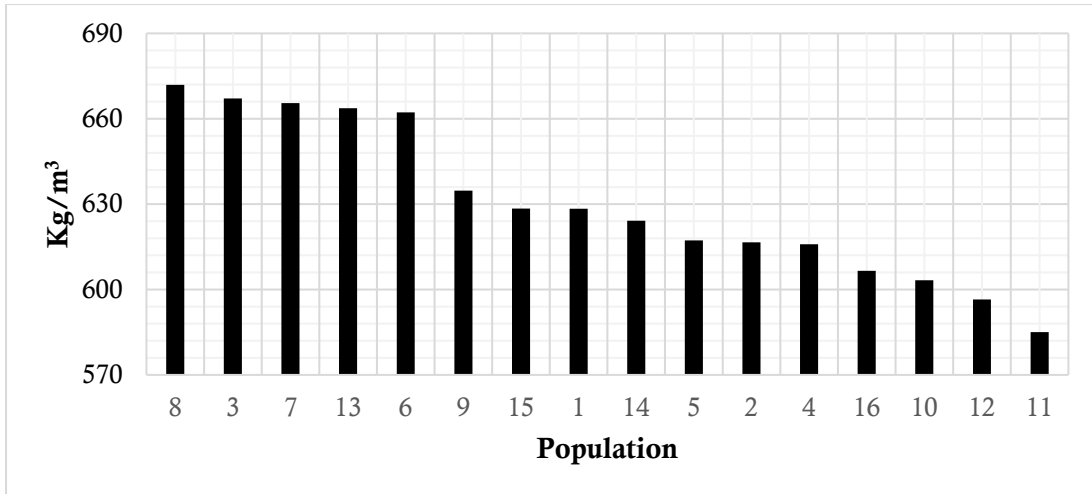


Figure 20. Sapwood density of *T. paniculata* candidate plus trees in Kerala

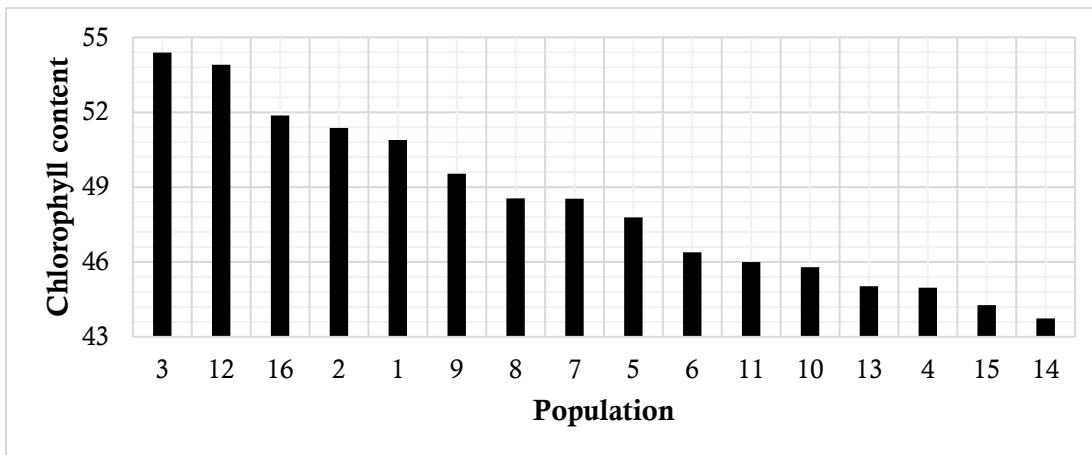


Figure 21. Chlorophyll content of *T. paniculata* candidate plus trees in Kerala

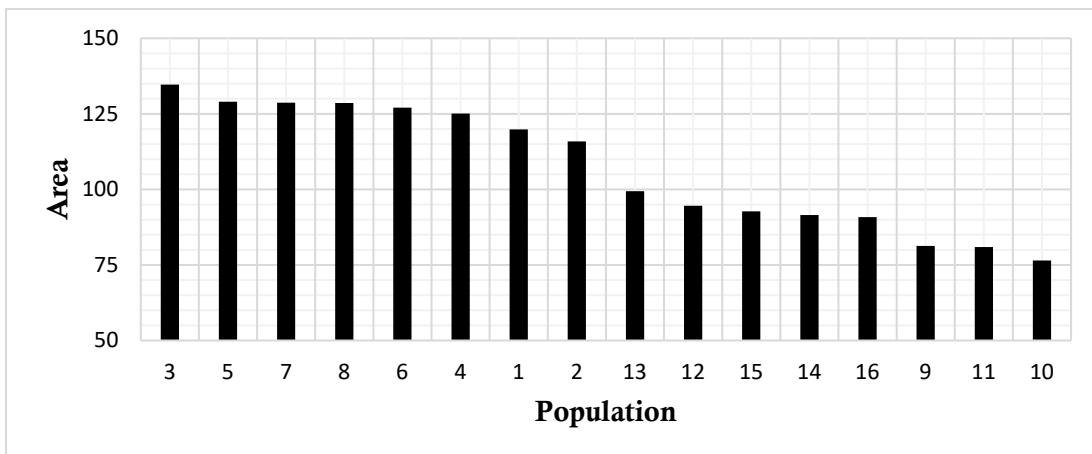


Figure 22. Leaf area of *T. paniculata* candidate plus trees in Kerala

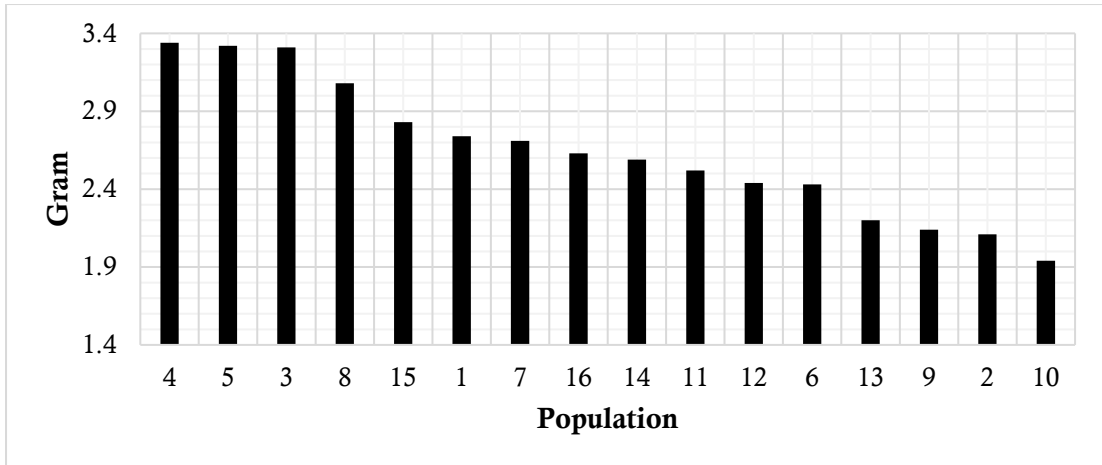


Figure 23. Leaf fresh mass of *T. paniculata* candidate plus trees in Kerala

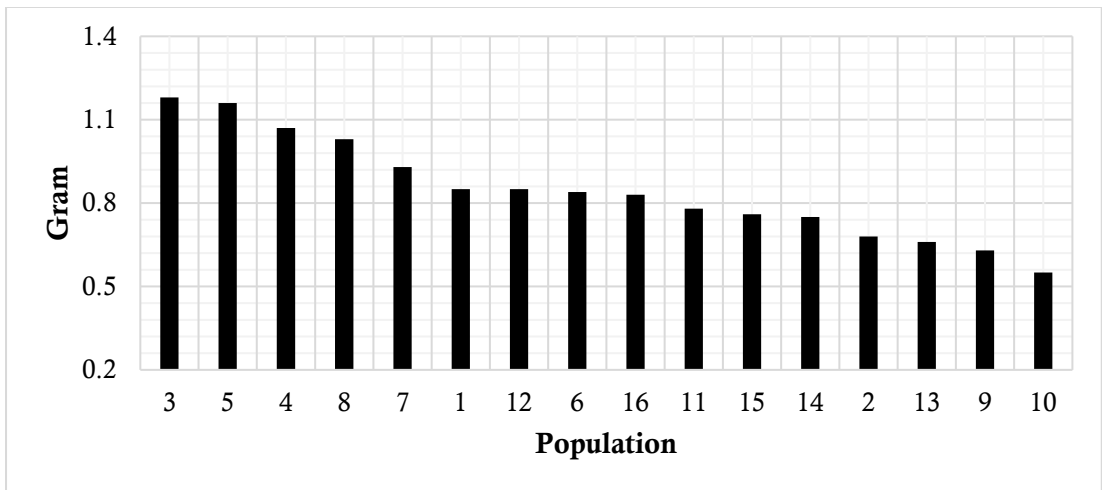


Figure 24. Leaf dry mass of *T. paniculata* candidate plus trees in Kerala

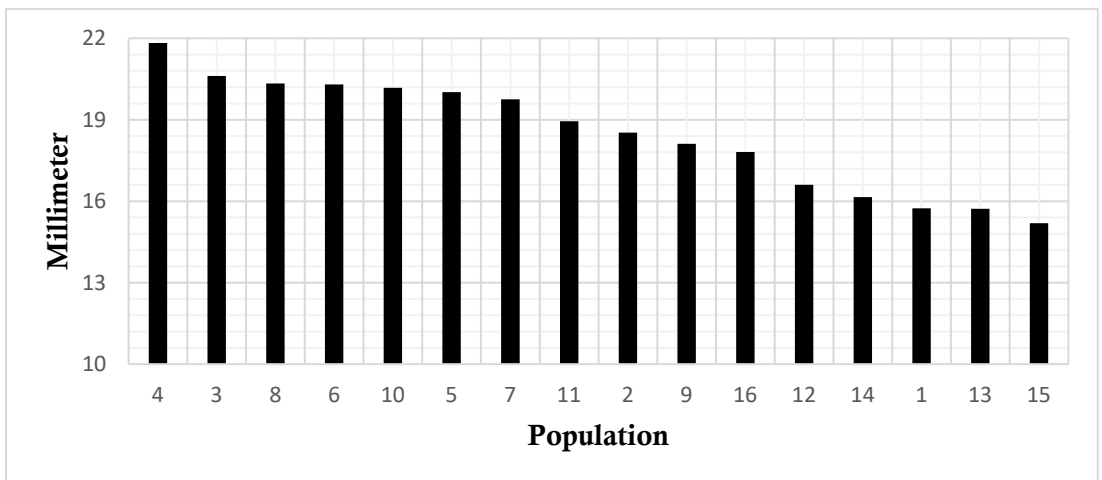


Figure 25. Fruit large wing length of *T. paniculata* candidate plus trees in Kerala

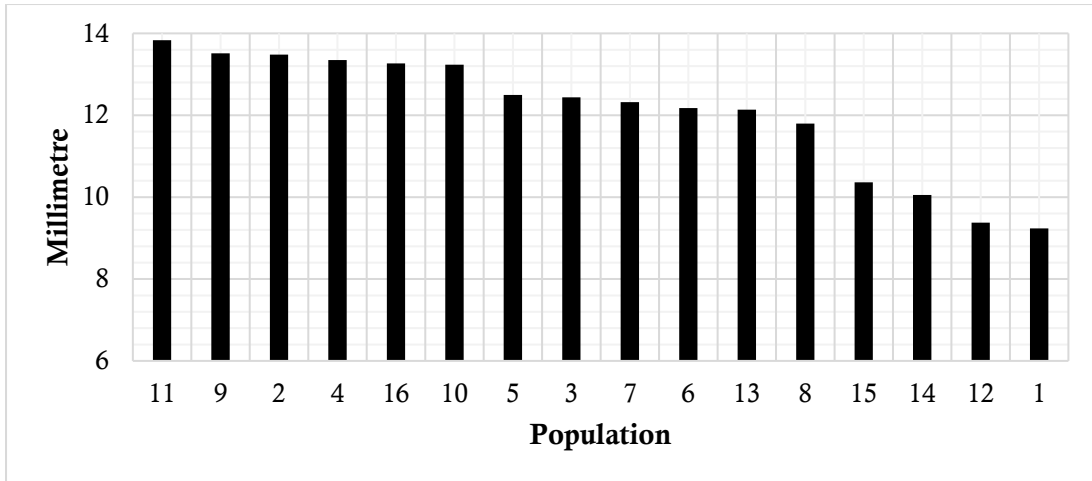


Figure 26. Fruit large wing width of *T. paniculata* candidate plus trees in Kerala

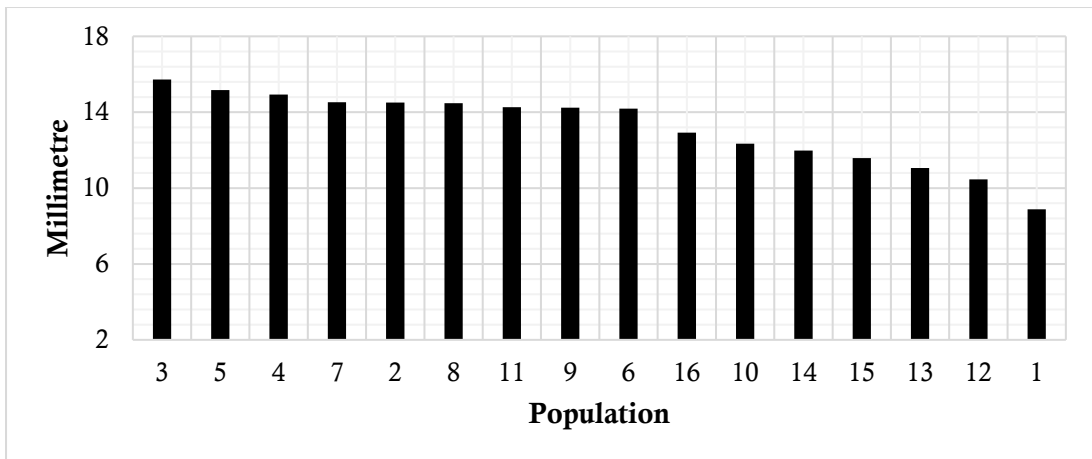


Figure 27. Fruit small wings length of *T. paniculata* candidate plus trees in Kerala

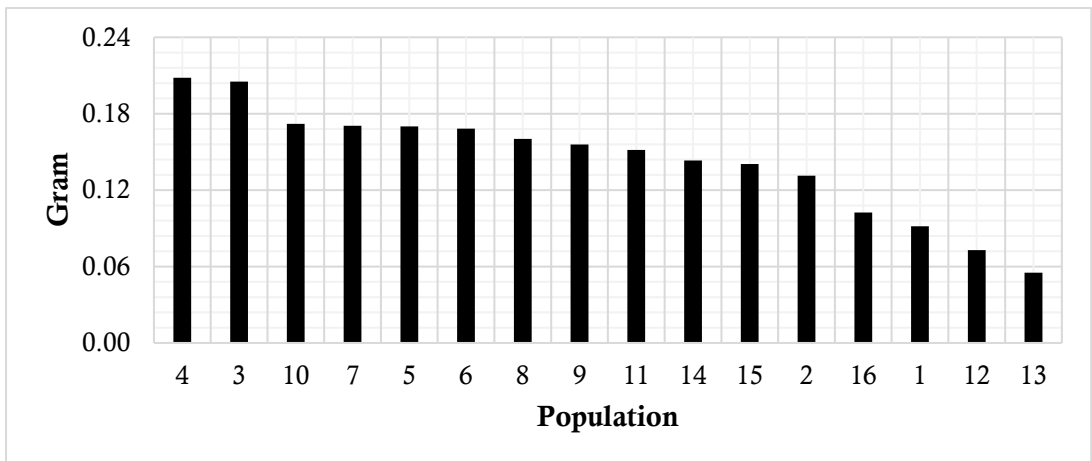


Figure 28. Fruit fresh mass of *T. paniculata* candidate plus trees in Kerala

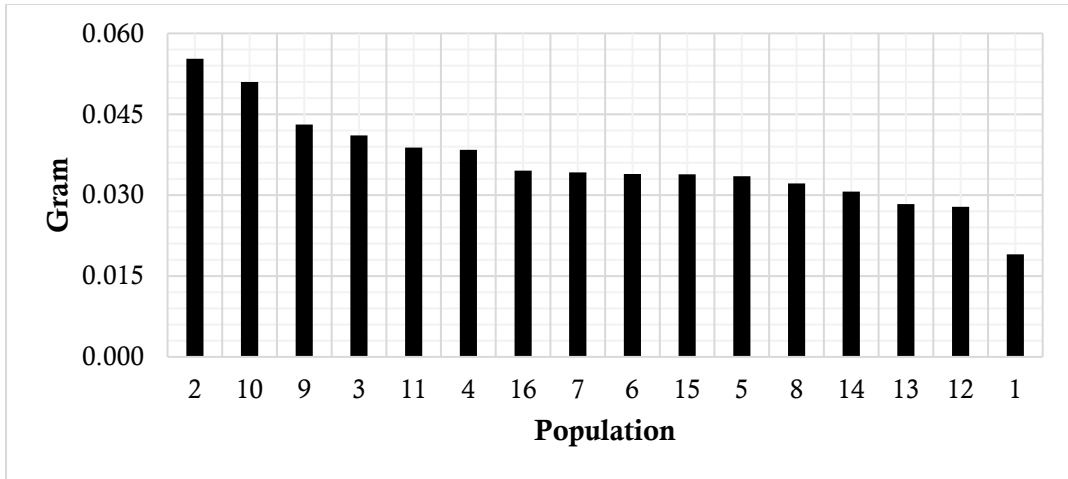


Figure 29. Fruit dry mass of *T. paniculata* candidate plus trees in Kerala

Phenotypic variation studies carried out in the genus *Terminalia* (*T. cuneata*, *T. bellerica*, *T. catappa*, *T. chebula* and *T. myriocarpa*) based on a large number of morphological characters such as tree form, leaf, fruits, wood, biochemical compounds etc. (Oboh et al. 2008; Navhale et al. 2011; Sanjeeva et al. 2013; Singh et al. 2013; Sharma et al. 2015; Wani and Singh 2016; Marjenah and Putri 2017; Das et al. 2020). In genera like *Terminalia*, preference is given to the characters directly connected with timber quality like total tree height, girth/ diameter at breast height, clear bole height/ length etc. In the case of winged fruit-yield *Terminalia*, preferences are given to characters related to fruit and seeds, such as fresh mass, dry mass, morphometry of wings, etc.

Tree characters such as circumference, total tree height, crown diameter, crown length, and a number of primary and secondary branches are directly connected with the commercial feature of a timer tree species and wood, leaf and fruit characters are connected indirectly. Tree girth and total tree height ranged between 2.32 m and 3.24 m (\bar{x} = 2.86 m), 23.80 m to 32.60 m (\bar{x} = 29.14 m), respectively (**Figure 11-12**). The crown diameter and crown length of individuals ranged between 12.40 m and 23.20 m (\bar{x} = 17.73 m), between 11.20 m and 17.20 m (\bar{x} = 13.51 m), respectively (**Figure 13-14**). The number of primary and secondary branches ranged between 4.20 and 7.80 (\bar{x} = 5.45) and between 9.40 and 16.20 (\bar{x} = 11.63), respectively (**Figure 15-16**).

Wood characteristics such as stress wave time, bark thickness, sapwood moisture content and sapwood density ranged between 423.33 μ s and 669 μ s (543.23 μ s) between 16 mm and 32.2 mm (\bar{x} =21.56 mm), between 33.06% and 44.78% (\bar{x} =40.23%), between 585.04 kg m⁻³ and 671.91 kg m⁻³ (\bar{x} = 630.43 kg m⁻³) respectively (**Figure 17-20**). Leaf characteristics such as chlorophyll content, area, fresh mass, dry mass ranged between 43.74 and 53.67 (\bar{x} = 48.29), between 76.50 cm² and 134.69 cm² (\bar{x} = 107.29 cm²), between 1.94 g and 3.34 g (\bar{x} = 2.64 g), between 0.55 g and 1.18 g (\bar{x} = 0.85 g) respectively (**Figure 21-24**). Fruit characteristics such as fresh mass, dry mass, large wing length, width, and small wings length ranged between 0.55 g and 0.21 g (\bar{x} = 0.14 g), between 0.02 g and 0.06 g (\bar{x} = 0.04 g), between 15.19 mm and 21.83 mm (\bar{x} = 18.49 mm), between 9.24 mm and 13.83 mm (\bar{x} = 12.07 mm), between 8.89 mm and 15.73 mm (\bar{x} = 13.22 mm) respectively (**Figure 25-29**). Moisture content in leaf ranged between 61.09% and 81.80% (\bar{x} = 68.26%) while in fruit, moisture content ranged between 43.51% and 87.13% (\bar{x} =72.82%).

From all the tree candidates, 19 tree characters were measured using standard methods and techniques and data were tabled in Microsoft Excel. After the tabulation, minimum and maximum value, mean, standard error, variance, standard deviation, median and coefficient of variation of the entire tree characters were calculated (**Table.13**). Present study helped the documentation of mean value of characters of *T. paniculata* located in its native range. Standard error of all the characters are >1 except ST (7.61), SD (2.78) and LA (2.06). Meantime standard error of fruit dry mass and fresh mass was zero. Similarly variance, standard variance and median differed from one character to another. Coefficient of variation confirmed that tree characters showed significant variation between the populations and distributed in a range between 4.33 (=SD) and 32.59 (=LD).

Sl. No.	Character (Unit)	Min	Max	Mean	SE	Variance	SV	CV
1.	GH (m)	1.72	3.68	2.86	0.05	0.20	0.45	15.84
2.	TH (m)	22.00	36.00	29.14	0.40	12.90	3.59	12.33
3.	CD (m)	10.00	26.00	17.73	0.45	16.38	4.05	22.83
4.	CL (m)	8.00	20.00	13.51	0.29	7.19	2.68	19.84
5.	PB (no. s)	2.00	15.00	5.45	0.19	3.09	1.76	32.23
6.	SB (no. s)	6.00	19.00	11.63	0.32	8.19	2.86	24.61
7.	ST (μ s)	423.33	669.00	543.23	7.61	4629.68	68.04	12.52
8.	BT (mm)	12.17	38.90	21.56	0.58	32.46	5.69	26.43
9.	SM (%)	30.64	48.70	40.22	0.36	12.16	3.49	8.67
10.	SD (g/m^3)	581.12	675.79	630.43	2.78	743.63	27.27	4.33
11.	LC	39.95	61.12	48.29	0.48	22.35	4.73	9.79
12.	LA (cm^2)	74.50	137.17	107.29	2.06	408.42	20.21	18.84
13.	LF (g)	1.29	4.83	2.64	0.07	0.51	0.71	27.04
14.	LD (g)	0.40	1.74	0.84	0.03	0.08	0.28	32.59
15.	LL (mm)	13.02	22.92	18.49	0.22	4.80	2.19	11.85
16.	LW (mm)	8.60	14.90	12.07	0.17	2.68	1.64	13.57
17.	SL (mm)	7.20	20.79	13.22	0.25	6.04	2.46	18.59
18.	FF (g)	0.05	0.29	0.14	0.00	0.00	0.05	31.80
19.	FD (g)	0.01	0.06	0.04	0.00	9.17E-5	0.00	26.57

Table.13. Univariate statistics of tree characters of *T. paniculata* located in Kerala (GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass; Max: Maximum; SE: Standard error; SV: Standard variance; CV: Coefficient of variation).

Popul ation	GH (m)	HT (m)	CD (m)	CL (m)	PB (no. s)	SB (no. s)	ST (μ s)	BT (mm)	SM (%)	SD (kg/ m ³)
TPP01	2.54± 0.37	24.8 ±4.2	12.4 ±2.2	17.2 ±2.2	4.8± 1.1	09.4 ±1.3	647.73± 17.12	19.4 ±4.2	42.0 ±1.4	628.4 ±4.3
TPP02	2.76± 0.44	30.6 ±3.8	16.8 ±5.2	15.2 ±2.3	4.4± 0.9	09.6 ±1.7	619.46± 05.33	16.0 ±2.7	39.9 ±1.9	616.6 ±3.2
TPP03	3.22± 0.29	26.0 ±4.0	20.8 ±2.3	12.6 ±1.1	7.8± 4.1	11.6 ±2.6	610.07± 06.93	20.5 ±3.7	44.8 ±2.5	667.1 ±4.3
TPP04	3.24± 0.25	31.0 ±1.0	18.6 ±2.4	11.8 ±0.5	5.8± 1.3	16.2 ±3.1	471.20± 19.47	18.9 ±4.0	39.7 ±1.3	615.9 ±4.4
TPP05	3.01± 0.49	32.6 ±0.9	16.4 ±2.9	12.6 ±0.9	5.0± 1.2	11.8 ±1.9	466.40± 09.90	18.9 ±2.0	41.1 ±1.5	617.3 ±3.6
TPP06	3.21± 0.56	28.8 ±4.3	21.4 ±2.9	11.2 ±1.8	4.6± 1.3	10.8 ±5.0	444.53± 12.93	18.7 ±3.7	37.6 ±2.5	662.3 ±3.4
TPP07	3.10± 0.41	29.0 ±3.1	21.6 ±3.9	11.8 ±2.7	4.2± 1.8	11.2 ±2.7	458.76± 15.89	17.9 ±1.8	36.5 ±1.7	665.5 ±3.5
TPP08	2.69± 0.32	31.8 ±1.1	17.6 ±3.9	16.2 ±2.9	6.6± 0.9	13.0 ±2.0	461.60± 18.14	19.7 ±4.1	36.8 ±1.8	671.9 ±4.0
TPP09	2.80± 0.35	28.6 ±1.3	14.6 ±2.2	15.8 ±3.3	5.4± 0.6	10.6 ±0.9	573.13± 15.32	27.1 ±3.9	39.2 ±1.7	634.7 ±2.1
TPP10	2.72± 0.41	27.6 ±3.7	16.4 ±2.2	13.8 ±3.6	5.6± 2.5	11.0 ±2.6	592.13± 04.36	29.2 ±3.6	41.8 ±2.0	603.3 ±2.7
TPP11	2.80± 0.35	28.6 ±1.3	14.8 ±1.9	15.8 ±3.4	5.4± 0.6	10.6 ±0.9	573.00± 06.41	32.2 ±4.3	42.2 ±0.8	585.0 ±3.2
TPP12	2.72± 0.41	27.6 ±3.7	16.4 ±2.2	13.8 ±3.6	5.6± 2.5	11.0 ±2.6	608.07± 17.79	18.6 ±3.7	41.2 ±1.6	596.0 ±2.8
TPP13	2.98± 0.33	30.4 ±2.6	23.2 ±1.1	11.6 ±2.6	6.0± 1.9	12.4 ±2.6	594.13± 14.04	26.8 ±2.3	33.1 ±2.4	663.7 ±4.3
TPP14	2.32± 0.46	28.2 ±3.3	13.4 ±1.5	13.4 ±2.1	5.0± 0.9	09.8 ±2.0	539.07± 09.29	19.9 ±4.6	41.9 ±3.2	624.2 ±2.9
TPP15	2.94± 0.24	30.2 ±1.7	20.2 ±3.7	12.8 ±2.2	6.0± 0.6	13.8 ±1.6	552.67± 15.60	19.5 ±4.9	41.6 ±2.5	628.4 ±3.6
TPP16	2.45± 0.28	23.8 ±1.5	14.6 ±1.3	12.4 ±1.7	5.2± 1.3	10.8 ±3.0	479.80± 06.16	21.6 ±4.9	44.1 ±2.4	606.6 ±2.1

Table.14. Population wise tree characters of *T. paniculata* in Kerala

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content & SD: sapwood density).

Popul ation	LC	LA (cm ²)	LF (g)	LD (g)	LL (mm)	LW (mm)	SL (mm)	FF (g)	FD (g)
TPP01	52.4 ±2.4	119.8 ±3.3	2.74± 0.39	0.85± 0.09	15.7 ±1.4	09.2 ±0.3	08.9 ±0.9	0.09± 0.02	0.02± 0.01
TPP02	51.4 ±1.9	115.8 ±2.5	2.11± 0.37	0.68± 0.14	18.5 ±0.9	13.5 ±0.9	14.5 ±1.5	0.13± 0.02	0.06± 0.01
TPP03	51.9 ±3.8	134.7 ±3.2	3.31± 0.97	1.18± 0.36	20.6 ±1.2	12.4 ±0.9	15.7 ±3.1	0.21± 0.02	0.04± 0.01
TPP04	44.9 ±3.1	125.1 ±3.2	3.34± 0.62	1.07± 0.23	21.8 ±1.1	13.4 ±0.6	14.9 ±2.8	0.21± 0.04	0.04± 0.00
TPP05	47.9 ±3.8	129.0 ±3.9	3.33± 0.94	1.16± 0.38	20.0 ±0.6	12.5 ±0.7	15.2 ±1.7	0.17± 0.02	0.03± 0.01
TPP06	46.4 ±3.5	127.1 ±3.6	2.43± 1.13	0.84± 0.42	20.3 ±0.9	12.2 ±1.3	14.2 ±2.1	0.17± 0.02	0.03± 0.00
TPP07	48.5 ±2.9	128.7 ±3.0	2.71± 0.29	0.93± 0.11	19.8 ±1.5	12.3 ±0.9	14.5 ±2.7	0.17± 0.01	0.03± 0.01
TPP08	48.6 ±2.2	128.6 ±2.2	3.08± 0.63	1.03± 0.21	20.3 ±0.9	11.8 ±1.2	14.5 ±1.8	0.16± 0.03	0.03± 0.01
TPP09	49.5 ±3.8	081.3 ±2.6	2.14± 0.44	0.63± 0.21	18.1 ±0.4	13.5 ±1.1	14.2 ±1.2	0.16± 0.01	0.04± 0.00
TPP10	45.8 ±2.4	076.5 ±1.9	1.94± 0.27	0.55± 0.09	20.2 ±0.9	13.2 ±1.2	12.4 ±1.2	0.17± 0.00	0.05± 0.00
TPP11	46.0 ±3.2	080.9 ±3.8	2.52± 0.38	0.78± 0.13	18.9 ±0.7	13.8 ±0.3	14.3 ±0.7	0.15± 0.01	0.04± 0.01
TPP12	53.7 ±1.5	094.6 ±1.5	2.33± 0.61	0.82± 0.22	16.6 ±1.0	09.4 ±0.5	10.7 ±0.9	0.07± 0.01	0.03± 0.01
TPP13	45.0 ±3.6	099.4 ±2.9	2.19± 0.57	0.66± 0.19	15.7 ±0.9	12.1 ±0.9	11.1 ±0.9	0.06± 0.05	0.03± 0.00
TPP14	43.7 ±1.9	091.5 ±2.5	2.59± 0.68	0.75± 0.20	16.2 ±0.4	10.1 ±0.7	11.9 ±1.6	0.14± 0.01	0.03± 0.00
TPP15	44.3 ±3.8	092.7 ±2.9	2.83± 0.27	0.76± 0.11	15.2 ±0.3	10.4 ±0.5	11.6 ±1.0	0.14± 0.01	0.03± 0.01
TPP16	51.9 ±1.2	090.8 ±2.5	2.63± 0.42	0.83± 0.12	17.8 ±2.2	13.3 ±1.6	12.9 ±2.5	0.10± 0.05	0.04± 0.01

Table.14. Population wise tree characters of *T. paniculata* in Kerala

(LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass).

Aryankavu Forest Range (TPP01) population is characterized by minimum crown diameter, several secondary branches, large fruit wings, small wings length, fresh fruit mass and maximum crown length (**Table.14**). Erumely Forest Range (TPP02) is characterized by minimum bark thickness and maximum fruit dry mass. Karulai Forest Range (TPP03) population is characterized by the maximum number of primary branches, sapwood moisture content, leaf area, leaf dry mass and fruit small wings length. Other populations in Karulai Forest Range (TPP04) showed height and girth at breast height, several secondary branches, fresh leaf mass, fruit large wing length and fresh fruit mass.

Meantime TPP05 population located in Karulai showed maximum values for total tree height. Sister populations (TPP06 and TPP07) belonging to Karulai Forest Range showed minimum measure in crown length and number of primary branches, respectively. Another population in Karulai Forest Range (TPP08) had a high value for sapwood density. Parambikulam Forest Range (TPP10) population had a minimum measure in leaf area, fresh leaf mass and dry leaf mass.

At the same time, the sister population (TPP11) had a minimum value for sapwood density and a maximum value for bark thickness and fruit large wing width. The evergreen population belonging to Peechi Forest Range (TPP13) had minimum value for sapwood moisture content, fresh fruit mass and maximum crown diameter.

Sl. No.	Tree characters (Mean value)	Minimum	Maximum
1.	GH (2.86 m)	TPP14 (2.32 m)	TPP04 (3.24 m)
2.	HT (29.14 m)	TPP16 (23.80 m)	TPP05 (32.60 m)
3.	CD (17.73 m)	TPP01 (12.40 m)	TPP13 (23.20 m)
4.	CL (13.51 m)	TPP06 (11.20 m)	TPP01 (17.20 m)
5.	PB (5.45)	TPP07 (4.20)	TPP03 (7.80)
6.	SB (11.63)	TPP01 (9.40)	TPP04 (16.20)
7.	ST (543.23 kg/m ³)	TPP06 (444.53 kg/m ³)	TPP01 (647.73 kg/m ³)
8.	BT (21.56 mm)	TPP02 (16 mm)	TPP11 (32.20 mm)
9.	SM (40.23%)	TPP13 (33.16%)	TPP03 (44.78%)
10.	SD (630.43 kg/m ³)	TPP11 (585.04 kg/m ³)	TPP08 (671.90 kg/m ³)
11.	LC (48.29)	TPP14 (43.74)	TPP12 (53.67)
12.	LA (107.29 cm ²)	TPP10 (76.50 cm ²)	TPP03 (134.69 cm ²)
13.	LF (2.64 g)	TPP10 (1.943 g)	TPP04 (3.343 g)
14.	LD (0.845 g)	TPP10 (0.549 g)	TPP03 (1.18 g)
15.	FL (18.49 mm)	TPP15 (15.19 mm)	TPP04 (21.83 mm)
16.	FW (12.07 mm)	TPP01 (9.24 mm)	TPP11 (13.83 mm)
17.	SL (13.22 mm)	TPP01 (8.89 mm)	TPP03 (15.73 mm)
18.	FF (0.144 g)	TPP13 (0.055 g)	TPP04 (0.208 g)
19.	FD (0.036 g)	TPP01 (0.019 g)	TPP02 (0.055 g)

Table.15. Mean value of plus tree characters

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass).

At the same time, the sister (TPP12) population exhibited top leaf chlorophyll content. People belonging to Sungam Forest Range (TPP14 and TPP15) showed minimum girth at breast height, leaf chlorophyll content and fruit large wing length. The short tree characterizes the population located in Walayar Forest Range (**Table.15**).

4.2.2. Analysis of Variance

Analysis of the variance of the nineteen characters showed significant variation ($p < 0.05$) between the populations (Table.16-17). ANOVA results showed a range of p-value of all the characters between 0.001 and 0.046. Except for the number of primary branches ($p = 0.046$), the p-value of characters ranges between 0.001 and 0.004 (no. of secondary branches). ANOVA confirms that nineteen characters subjected to variation analysis showed significant variation between the populations.

Character	GH	HT	CD	CL	PB	SB	ST	BT	SM
F stat	2.696	4.086	5.613	3.131	1.852	2.598	136.494	4.162	13.229
P value	0.003	0.001	0.001	0.001	0.046	0.004	0.001	0.001	0.001

Table.15. F stat and P value of tree characters between populations

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content).

Character	SD	LC	LA	LF	LD	LL	LW	SL	FF	FD
F stat	386.715	4.969	3.178	4.105	284.94	31.358	21.679	7.45	33.971	20.176
P value	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001

Table.16. F stat and P value of tree characters between populations

(SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass).

4.2.3. Correlation between Tree Characters

Girth at breast height correlated with seven tree characters (Table.18-19). The number of primary branches showed no significant bond with tree characters. Circumference/ girth at breast height of trees showed maximum significant relations with tree characters such as crown diameter ($r = 0.783$), crown length ($r = -0.546$), number of secondary branches ($r = 0.546$), leaf area ($r = 0.539$), fruit large wing length ($r = 0.549$),

fruit small wings length ($r=0.543$) and fruit fresh mass ($r=0.540$). Total tree height showed a significant bond with the number of secondary branches ($r=0.500$) and sapwood moisture content ($r=-0.569$). Crown diameter showed significant relationship with girth at breast height ($r=0.783$), crown length ($r=-0.690$), sapwood moisture content ($r=-0.559$) and sapwood density ($r=0.648$). Crown length showed a significant bond with the crown length ($r=-0.546$) and crown diameter ($r=-0.690$). The number of secondary branches showed a significant bond with girth at breast height ($r=0.546$), total tree height ($r=0.500$) and fresh leaf mass ($r=0.544$). Stress wave time showed a strong correlation only with bark thickness ($r=-0.519$)

Wood characteristics such as bark thickness, sapwood moisture and density showed significant relationships with other tree characters. Bark thickness significantly correlates with leaf area ($r=-0.694$) and stress wave time ($r=-0.519$). Sapwood moisture content had a significant relationship with total tree height ($r=-0.569$), crown diameter ($r=-0.559$) and sapwood density ($r=-0.559$). Sapwood density showed significant relationship with leaf area ($r=0.606$), crown diameter ($r=0.648$) and sapwood moisture content ($r=-0.559$).

Leaf characteristics such as leaf area, fresh leaf mass and leaf dry mass showed a significant relationship with other tree characters; meantime, leaf chlorophyll content had no relationship with other characters. Leaf area showed significant relationship over girth at breast height ($r= 0.539$), bark thickness ($r=-0.694$), sapwood density ($r=0.606$), leaf fresh mass ($r= 0.667$) and leaf dry mass ($r=0.792$). Leaf fresh mass showed significant relation with several secondary branches ($r=0.544$), leaf area ($r=0.667$) and dry leaf mass ($r=0.937$). Leaf dry mass showed a significant relationship with leaf area ($r=0.792$), fresh leaf mass ($r=0.937$) and fruit large wing length ($r=0.512$).

Fruit characteristics such as fruit large wing length, fruit large wing width, fruit small wings length, fruit fresh mass and fruit dry mass showed a significant relationship

with other characteristics. The data shows that fruit's large wing length is significant over fresh fruit mass and fruit's large wing width is significant over fruit dry mass. Fruit large wing length showed a significant relationship with girth at breast height ($r=0.549$), dry leaf mass ($r=0.512$), fruit large wing width ($r=0.638$), and fruit's small wings length ($r=0.825$) and fresh fruit mass ($r=0.812$).

By comparing all the correlation relationships between the nineteen tree characters, it's evident that the number of primary branches and leaf chlorophyll content doesn't show any significant bond with other characters. Maximum ($r=0.937$) strong significant positive correlation was seen between fresh leaf mass and dry leaf mass, followed by fruit large wing length and fruit small wings length ($r=0.825$) and between fresh fruit mass and fresh leaf mass ($r=0.812$). In the meantime, a strong negative significant correlation was seen between bark thickness and leaf area ($r=-0.694$) and crown diameter and length ($r=-0.690$). Other strong negative correlations were seen between girth at breast height/ circumference with crown length ($r=-0.548$), between crown diameter and sapwood moisture content ($r=-0.559$) and between sapwood moisture content and sapwood density ($r=-0.559$). Among the correlation relationships, most of the relations were significant at a 99% confidence level.

Stress wave time is significantly correlated with bark thickness only, and dry fruit mass is associated considerably with fruit large wing width. Bark thickness is critical over stress wave time and leaf area only. Total tree height is significant over several secondary branches and sapwood moisture content. Similarly, crown length is significant over girth at breast height and crown diameter. Among the five fruit characters, fruit large wing length is the only character that significantly correlates with all other characters (large wing width, small wings length, fresh mass and dry mass). Among the nineteen characters, girth at breast height is the character that showed a significant correlation with the maximum ($n=7$) number of characters such as crown diameter,

crown length, leaf area, fruit large wing length, fruit small wings length, leaf area and fruit fresh mass.

	HT	CD	CL	PB	SB	ST	BT	SM	SD	LC	LA	LF	LD	LL	LW	SL	FF	FD
GH	0.40	0.78**	-0.55*	0.23	0.55*	-0.16	-0.13	-0.31	0.42	-0.18	0.54*	0.36	0.45	0.55*	0.33	0.54*	0.54*	0.02
HT		0.37	-0.12	-0.02	0.50*	-0.01	-0.14	-0.57*	0.18	-0.49	0.27	0.19	0.17	0.26	0.19	0.40	0.27	0.05
CD			-0.69**	0.27	0.48	-0.08	-0.15	-0.56*	0.65**	-0.27	0.41	0.12	0.19	0.23	0.14	0.28	0.19	-0.06
CL				-0.02	-0.41	0.02	0.22	0.19	-0.25	0.35	-0.22	-0.19	-0.22	-0.21	-0.14	-0.23	-0.19	0.07
PB					0.45	0.18	0.19	0.19	0.27	0.02	0.07	0.39	0.35	0.14	0.00	0.16	0.23	0.00
SB						0.17	-0.10	-0.23	0.16	-0.43	0.24	0.54*	0.41	0.33	0.15	0.26	0.37	-0.08
ST							-0.52*	0.25	-0.06	0.14	0.28	0.35	0.23	-0.33	-0.33	-0.25	-0.22	0.02
BT								0.01	-0.29	-0.31	-0.69**	-0.43	-0.48	-0.04	0.40	-0.06	-0.06	0.19
SM									-0.56*	0.32	-0.21	0.23	0.17	-0.01	-0.07	-0.01	0.18	0.21
SD										-0.05	0.61*	0.24	0.30	0.16	-0.08	0.21	0.20	-0.34
LC											0.22	-0.01	0.19	-0.04	-0.14	-0.08	-0.28	0.07
LA												0.67**	0.79**	0.48	-0.06	0.41	0.38	-0.24
LF													0.94**	0.38	-0.08	0.38	0.49	-0.36
LD														0.51*	-0.03	0.49	0.48	-0.31
LL															0.64**	0.83**	0.81**	0.37
LW																0.74**	0.48	0.71**
SL																	0.79**	0.45
FF																		0.28

Table.18. Pearson (linear r) correlation between the tree characters of *T. paniculata* in Kerala

GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass)

The correlation analysis using Palaeontological Statistics (PAST) software graphically represents the relationship between the tree characters (**Figure 30**). A positive correlation is displayed in blue colour dots, while a negative correlation is shown in red dots, and the dots' denseness indicates the strong relationship between the characters. Strangeness between the tree characters represents in the form of the denseness of the beads. More dense bubbles show a strong bond, and low, dense dots show a weak relationship between the characters. The second graphical figure is the sorted graph of the former. The significant ($p < 0.05$) bond between the tree characters is only displayed in the blue and red graph, excluding the lower trainable. From the graph, it's evident that the number of primary branches and leaf chlorophyll content did not display any dots in the

graph, indicating that there was no bond between these traits and the tree characters. Compared to a tabular representation of the data, a graphical model is louder to understanding the characters' bond. The coloured data reveals a positive correlation between the characters than a negative correlation between the characters.

Sl. No.	Tree characters	Correlation with
1.	GH	SB, CD, CL, LA, LL, SL, FF
2.	HT	SB, SM
3.	CD	GH, CL, SM, SD
4.	CL	GH, CD
5.	SB	LF, GH, HT
6.	ST	BT
7.	BT	LA, ST
8.	SM	SD, HT, CD
9.	SD	LA, CD, SM
10.	LA	GH, BT, SD, LF, LD
11.	LF	LD, SB, LA
12.	LD	LA, LF, LL,
13.	LL	GH, LW, SL, LD, FF
14.	LW	LL, SL, FD
15.	SL	GH, FF, LL, LW
16.	FF	GH, LL, SL
17.	FD	LW

Table.19. Tree characters showing significant correlation

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass).

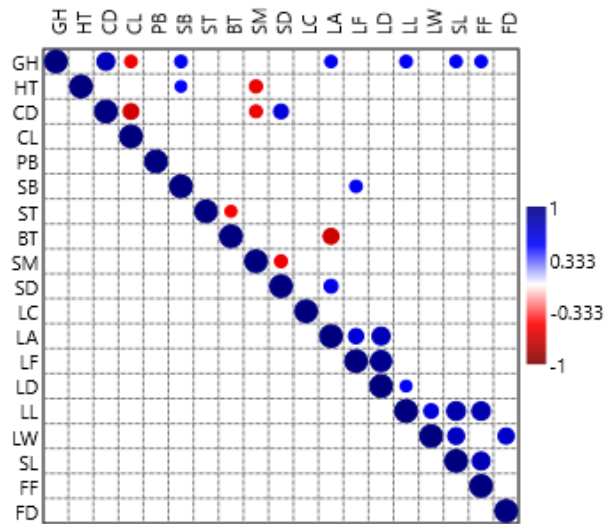


Figure.30. Linear r correlation statistics of tree characters of *T. paniculata* populations in Kerala (exclude lower triangle; significance $p > 0.05$ blank)

(*GH*: girth at breast height, *HT*: total height, *CD*: crown diameter, *CL*: crown length, *PB*: no. of primary branches, *SB*: number of secondary branches, *ST*: stress wave time, *BT*: bark thickness, *SM*: sapwood moisture content and *SD*: sapwood density; *LC*: leaf chlorophyll content, *LA*: leaf area, *LF*: leaf fresh mass, *LD*: leaf dry mass, *LL*: fruit large wing length, *LW*: fruit large wing width, *SL*: fruit small wings length, *FF*: fruit fresh mass and *FD*: fruit dry mass).

4.2.4. Principal component analysis (PCA)

Present PCA of the data resulted with Eigen analysis of the correlation matrix (eigenvalue, proportion and cumulative of principal components), Eigenvectors and graphs such as scree plots, score plot and loading plot of tree characters (**Table.20-21**). Eigen values of principal components from PC01 to PC18 revealed that the first 5 principal components have eigenvalue more than one (>1). The eigenvalue of PC01, PC02, PC03, PC04, PC05 and PC06 is 6.339, 3.132, 2.706, 1.638, 1.465 and 1.075 respectively. All other principal components from PC07 to PC19 have eigenvalue less than one (<1) ranging between 0.839 and 0.

Principle component	Eigenvalue	Proportion	Cumulative
PC01	6.339	0.334	0.334
PC02	3.132	0.165	0.499
PC03	2.706	0.142	0.641
PC04	1.638	0.086	0.727
PC05	1.465	0.077	0.804
PC06	1.075	0.057	0.861
PC07	0.839	0.044	0.905
PC08	0.553	0.029	0.934
PC09	0.467	0.025	0.959
PC10	0.307	0.016	0.975
PC11	0.199	0.010	0.985
PC12	0.168	0.009	0.994
PC13	0.062	0.003	0.997
PC14	0.028	0.001	0.999
PC15	0.021	0.001	1.000
PC16	0.000	0.000	1.000
PC17	0.000	0.000	1.000
PC18	0.000	0.000	1.000
PC19	-0.000	0.000	1.000

Table.20. Eigen analysis of the correlation matrix.

A component with an eigenvalue >1 means that the data shows variability. Here PC1 to PC6 have eigenvalue >1 are the real asset of the variability among the PCs. So PC1 to PC6 were selected for further analysis. Proportion indicates that PC1 is the principal component with accounted maximum to the variability. The first principal component (PC1) accounts for 33.40% of the total variance. Followed by PC2 (16.50%), PC3 (14.20%), PC4 (8.60%), PC5 (7.70%) and PC6 (5.70%). Cumulative value of Eigen analysis of the correlation matrix reveals that PC01 to PC06 explains 86.10% of the variability.

Variable	PC01	PC02	PC03	PC04	PC05	PC06
GH	0.319	-0.036	0.116	0.052	-0.246	0.127
HT	0.210	-0.057	0.256	-0.062	0.246	-0.459
CD	0.257	0.071	0.333	0.053	-0.351	0.202
CL	-0.212	-0.037	-0.221	-0.037	-0.010	-0.701
PB	0.112	0.016	-0.114	0.607	-0.337	-0.165
SB	0.261	0.014	0.124	0.393	0.173	-0.017
ST	-0.261	-0.010	-0.059	0.200	-0.427	-0.154
BT	-0.132	-0.341	0.148	0.344	-0.053	-0.135
SM	-0.108	-0.079	-0.485	0.228	0.080	0.322
SD	0.210	0.229	0.172	-0.099	-0.359	-0.194
LC	-0.099	0.133	-0.368	-0.249	-0.409	0.006
LA	0.288	0.257	-0.154	-0.252	-0.137	-0.116
LF	0.267	0.208	-0.289	0.218	0.208	-0.056
LD	0.288	0.203	-0.319	0.067	0.060	-0.032
LL	0.292	-0.256	-0.181	-0.127	-0.017	-0.039
LW	0.142	-0.480	-0.002	-0.113	-0.082	0.012
SL	0.297	-0.286	-0.148	-0.148	-0.034	-0.080
FF	0.285	-0.225	-0.188	0.049	0.082	-0.042
FD	-0.010	-0.465	-0.087	-0.126	-0.211	0.056

Table.21. Eigen vectors

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass)

The first two principal components such as PC1 and PC2, accounted for 49.90% of the variability. Thirteen PCs, such as PC7 to PC19, explain only 13.90% of the variability. The PCs from PC07 to PC19 resulted in 4.40% to 0% of the variability. Among the PCs, PC16, PC17, PC18 and PC19 did not have any contribution to variability. Cumulative values of PC1 and PC2 as the result of Eigen analysis of the correlation matrix indicated that among the 19 principal components, these two principal

components are enough for further research, such as eigenvectors and graph plots. Eigenvectors of principal components with nineteen tree characters resulted from a correlation between principal components with tree characters.

PC01 was positively correlated with girth at breast height followed by leaf area, dry leaf mass, fruit large wing length, fruit small wings length, and fruit fresh mass. PC02 was negatively correlated with fruit's large wing width, dry fruit mass, bark thickness, and fruit's small wings length. PC03 was positively correlated with the crown diameter and negatively correlated with sapwood moisture content, leaf chlorophyll content, dry leaf mass, and leaf fresh mass. PC04 was positively correlated with the number of primary branches and secondary branches and bark thickness. PC05 was negatively related to the stress wave time, leaf chlorophyll content, sapwood density, number of primary branches and crown diameter. PC06 was positive with sapwood moisture content and negatively related to total tree height and crown length. PC01 had positive coefficients with six characters, followed by PC03 and PC05 with five characters, PC02 with four characters, and PC04 and PC06 with three characters.

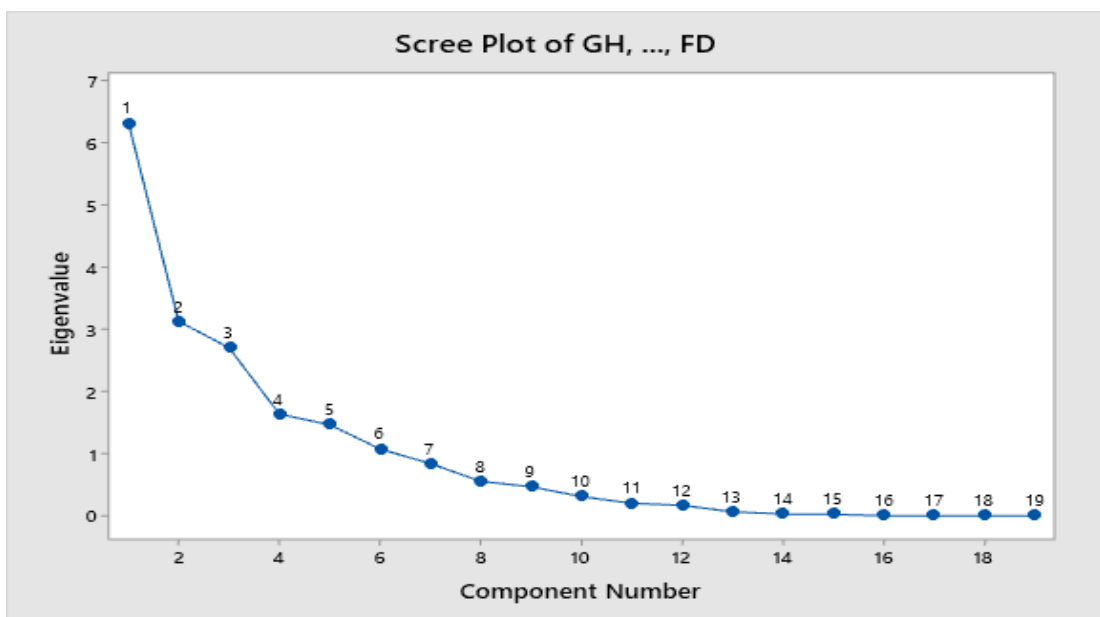


Figure.31. Scree plot of GH, ..., FD.

Eigenvalue plotted against the component number in scree plot revealed the eigenvalue of PC01 to PC19. Scree plot is the graphical representation of eigenvalue as the result of Eigen analysis of the correlation matrix (**Figure.31**). The graph shows that the first six PCs (PC01 to PC06) are placed above the eigenvalue above one. All other PCs (PC07 to PC19) are placed below eigenvalue one. Following the Eigen rule, the first six PCs are an asset of the variability. All other characters are not the real cause of variability.



Figure.32. Score plot of GH,..., FD.

As per cumulative value as the result of Eigen analysis of the correlation matrix, the first two principal components such as PC01 and PC02, accounted for 49.90% of the variability. The first principal component was plotted against the second principal component in the score plot (**Figure.32**). The score plot is similar to the dendrogram, which is divided into four quadrants, and it's evident that plus trees such as 16 (TPP16), 9 (TPP09), 2 (TPP02), 10 (TPP10) and 11 (TPP11) are placed in the first quadrant (-x, -y). The second quadrant (-x, y) included plus trees such as 1 (TPP01), 12 (TPP12), 14 (TPP14), 13 (TPP13) and 15 (TPP15) and the fourth quadrant (x, -y) included plus trees

such as 4 (TPP04). The third quadrant (x, y) included 8 (TPP08), 6 (TPP06), 7 (TPP07), 5 (TPP05) and 3 (TPP03). The score plot reveals that populations in Karulai Forest Range (TPP03, TPP04, TPP05, TPP06, TPP07 and TPP08) showed more similarity than any other population.

Populations located in Parambikulam Forest Range (TPP09, TPP10 and TPP11) showed similarity with the population located in Erumely Forest Range (TPP02), followed by Walayar Forest Range (TPP16). The population in Aryankavu Forest Range (TPP01) showed maximum similarity with that in Peechi Forest Range (TPP12).

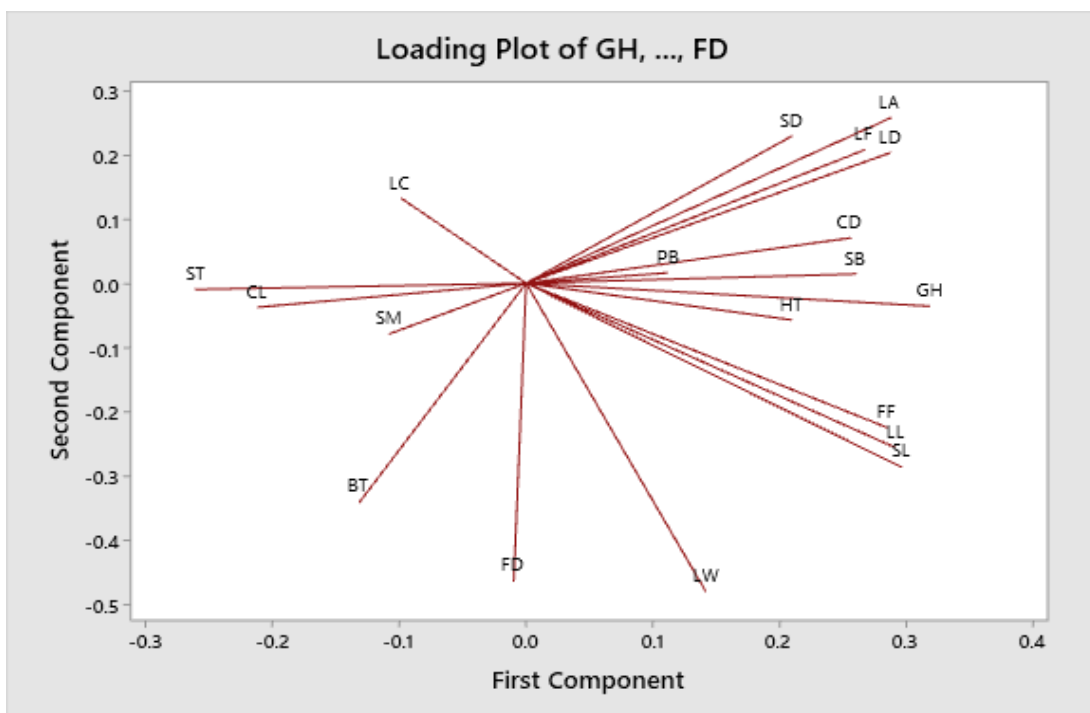


Figure.33. Loading plot of GH, ..., FD

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass).

The loading plot is a graphical representation of the first component against the second component that indicates the relativeness between the tree (**Figure.33**) characters. According to the principle component analysis rule, if the angle between two tree characters is below 90° they are strongly correlated and the angle between the trees characters is above 90° means they are weakly correlated. Present graphical representation in the form of a loading plot indicated the relationship between the tree characters. From the graph it's very clear that components such as LC, ST, CL and SM showed strong correlation with minimum number of components followed by BT. Fruit components LL, LW, SL and LW had strong correlation with maximum number of components (**Table.22**).

Correlation pattern confirmed that the components mentioned above such as LC, ST, CL, SM. and BT are relatively different from other components. As per loading plot, the components/characters placed above the plane (+x axis) are lead characters. From the graph it's clear that the characters such as ST, LC, SD, LA, LF, LD, PB, SB and CD are lead characters, which contributed more to the variation between the populations.

Sl.No.	Character	Strong correlation with
1.	LC	ST, CL, SM, BT, SD
2.	ST	CL, SM, BT, FD, LC
3.	CL	ST, LC, SM, BT, FD
4.	SM	CL, ST, LC, BT, FD
5.	BT	SM, CL, ST, LC, FD, LW
6.	FD	BT, SM, CL, ST, LW, SL, LL, FF, HT, GH
7.	LW	FD, BT, SL, LL, FF, HT, GH, SB, PB, CD, LD, LF, LA, SD
8.	SL	LW, FD, BT, LL, FF, HT, GH, SB, PB, CD, LD, LF, LA, SD
9.	LL	LW, FD, BT, SL, FF, HT, GH, SB, PB, CD, LD, LF, LA, SD
10.	FF	LW, FD, BT, SL, LL, HT, GH, SB, PB, CD, LD, LF, LA, SD
11.	HT	FF, LL, SL, LW, FD, GH, SB, PB, CD, LD, LF, LA, SD
12.	GH	FF, LL, SL, LW, FD, HT, SB, PB, CD, LD, LF, LA, SD
13.	SB	SD, LA, LF, LD, PB, CD, GH, HT, FF, LL, SL, LW
14.	PB	SD, LA, LF, LD, CD, SB, GH, HT, FF, LL, SL, LW
15.	CD	SD, LA, LF, LD, PB, CD, SB, GH, HT, FF, LL, SL, LW
16.	LD	SD, LA, SD, PB, SB, CD, GH, HT, FF, LL, SL, LW
17.	LF	SD, LA, LD, PB, SB, CD, HT, HT, FF, LL, SL, LW
18.	LA	SD, LF, LD, PB, SB, CD, HT, GH, FF, LL, SL, LW
19.	SD	LA, LF, LD, CD, PB, SB, GH, HT, FF, LL, SL, LC

Table.22. Correlation between characters as per loading plot.

(GH: girth at breast height, HT: total height, CD: crown diameter, CL: crown length, PB: no. of primary branches, SB: number of secondary branches, ST: stress wave time, BT: bark thickness, SM: sapwood moisture content and SD: sapwood density; LC: leaf chlorophyll content, LA: leaf area, LF: leaf fresh mass, LD: leaf dry mass, LL: fruit large wing length, LW: fruit large wing width, SL: fruit small wings length, FF: fruit fresh mass and FD: fruit dry mass).

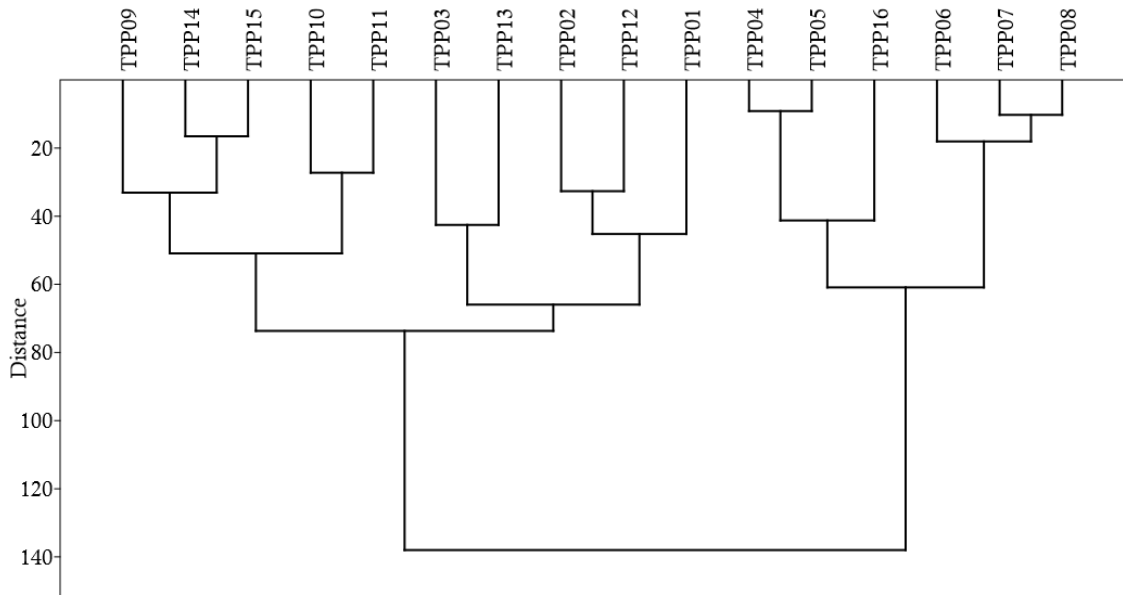


Figure.34. Multivariate classical clustering of tree characters of *T. paniculata* in Kerala (Y-axis: Euclidean distance; X-axis: Populations).

4.2.4. Cluster Analysis

Multivariate classical clustering of tree functional characteristics resulted in prominent hierarchical clustering of 16 populations of *T. paniculata* in Kerala (**Figure.34**). Sixteen selected populations of *T. paniculata* were firstly assembled into an alpha cluster and beta cluster. Alpha cluster includes populations such as TPP01, TPP02, TPP03, TPP09, TPP10, TPP11, TPP12, TPP13, TPP14 and TPP15. The Alpha cluster is again divided into two sub-clusters within 80 Euclidean distances (Ed), and the Beta cluster (TPP04, TPP05, TPP06, TPP07, TPP08 and TPP16) is divided into two sub-clusters within 80 Ed.

It's also clear that *T. paniculata* populations were placed within 140 Ed, and very close populations in the study area were seated within 20 Ed. The Alpha cluster includes ten populations, and the beta cluster contains 06 populations within 80 Ed. Among the six populations in the Karulai Forest Range of Nilambur South Forest Division, 05 (TPP04, TPP05, TPP06, TPP07 and TPP08) were grouped under the beta cluster.

Walayar Forest Range (TPP16) population showed similarity with populations located in Karulai Forest Range. Among the 16 populations, only six in Karulai and one in Walayar Forest Range placed in the alpha cluster. TPP13 (Peechi Forest Range) showed similarities with TPP03 (Karulai Forest Range). Southern populations such as TPP01 (Aryankavu Forest Range) and TPP02 (Erumely Forest Range) showed similarities with TPP12 (Peechi Forest Range). Populations in Parambikulam Tiger Reserve (Parambikulam and Sungam Forest Ranges) were placed in a single sub-cluster.

High similarity (<20 Ed) was shown by TPP04, and TPP05 (Karulai Forest Range), followed by TPP07 and TPP08 (Karulai Forest Range) Alpha cluster was divided into two sub-clusters below 80 Ed. Each sub-cluster included five populations, Two populations (TPP12 and TPP13) are located in Peechi Wildlife Division, but TPP12 showed maximum similarity TPP02 (Erumely Forest Range). TPP13 showed maximum similarity with TPP03 (Karulai Forest Range). Similarly, TPP09 (Parambikulam Forest Range) showed maximum similarity with populations located in Sungam Forest Range (TPP14 and TPP15) than populations in the same Forest Ranges (TPP10 and TPP11). Among the 16 populations of *T. paniculata* located in Kerala, TPP1 (Aryankavu Forest Range) showed maximum dissimilarity with any other character in other populations, followed by TPP16 (Walayar Forest Range). Compared to all, cluster formed by TPP03 and TPP13 placed above 40 Ed.

Among the nineteen characters selected in the study, only five are the tangible assets to cause variation between the populations. Among the characters mentioned above, characters like total tree height, girth at breast height, crown diameter, crown length and several primary branches were responsible for the variation between the populations. Hence present study confirmed that stand-up tree characters need more attention in evaluating tree population variation. The literature studies show that among the *Terminalia*, scholars select various characteristics, including stand-up, wood, leaf,

fruit, seed and biochemical markers. The present research on population variation aspects of *T. paniculata* suggests removing non-significant characters to study variation.

As per PCA, 87.20% of the variation was contributed by six characters: tree height, girth at breast height, crown length and diameter, and several primary and secondary branches. All other characters contributed only 12.80% of the total variation. In this study, three different statistical software's for data analysis included SPSS, PAST and Minitab. From experience, it's evident that compared to other software, most advanced software like Minitab can help present the data more attractively and provide easy understanding. SPSS and Minitab gave different types of output as part of correlation analysis; hence, it's better to use specific software for data analysis. Based on the result, Minitab software was used for further data analysis, including PCA.

4.3. To study the seed characteristics and seed handling techniques.

4.3.1. Fruit Production and Maturity Index

The fruit production pattern of *T. paniculata* individuals located in five individuals in both Peechi Forest Range (three) and Kerala Forest Research Institute campus (two) was estimated by analyzing sixty inflorescences (**Figure.35**). An average of 30.20 flower buds initiated at the first stage of flower phenophases ends with an average of seventeen opened flowers per inflorescence. It seems that 56.30% of flower buds developed into mature flowers, showing that 43.71% of flower buds get futile. Fruiting phenophases initiated with thirteen fruit buds per inflorescence, which is 43% of the total buds developed per inflorescence (**Table.23**).



Figure.35. Flowering from newly originated twigs at a height of 3-metre

Out of the total number of flower buds per inflorescence developed, only 27.80% developed into ripened fruits. It confirms that 72.20% of flower buds get futile before the termination of the reproductive phenophases of *T. paniculata*. Like other *Terminalia*, *T. paniculata* produces many flowers and fruits. A reproductive phenological pattern shows that approximately 25% of the flower buds attain the ripened fruit stage.

Fruit maturity was assessed by analyzing the fruits/seeds of *T. paniculata* at a regular interval of once a week. For analysis, the fruit and seed colour were recorded during the fruit development from the 11th to 25th week of fruit development. Seed is viable during the 11th to 25th week of fruit development selected for assessing fruit maturity index. Even though the Munsell colour chart invented by famous American painter Albert Henry Munsell is an effective colour chart for soil but it's highly valid and reliable to estimate the colour of other categories, including human skin. Along with fruit and seed colour, seed germinability and moisture content were recorded during this period.

Tree No.	Per Inflorescence					
	Flower Bud	Young flower	Opened flower	Fruit Bud	Young fruit	Ripened fruit
1	34	24	20	16	14	11
2	30	27	21	18	16	12
3	28	17	14	09	07	05
4	31	18	16	12	10	08
5	28	18	14	10	08	06
Mean	30.20	20.80	17	13	11	8.40
Futile Percentage		31.12	43.71	56.95	63.58	72.19

Table.23. Flower and fruit set per inflorescence in *T. paniculata*.

Fruit colour during the fruit development period from the 11th week up to natural fruit fall (25th week) begins with greenish-red coloured matured fruits and ends with deep red-coloured fruits. In between these two colour, fruits changes from greenish-red (11th and 12th weeks) to light red (13th and 14th weeks) to red (15th to 20th weeks) to deep red (21st to 25th weeks). Before the 11th week of fruit development, the colour of fruits is green to yellowish-green in colour. Seed colour during the development period begins with light brown coloured ends with deep brown. Before the period (11 to 25th week), the colour of seed looks dull white. Light brown coloured seeds occurred during the 11th to 16th week, and deep brown coloured seeds arose during the 17th to 25th week of fruit development. Germination experiments show that seed germinability ranged between 0.60% (11th week) and 0% (25th week). In between the 11th to 25th weeks, maximum germinability was recorded during the 16th week of fruit development (Table.24).

Sl. No.	Development of fruits (Weeks)	Colour of Fruit	Colour of Seed	Percentage	
				MC	GP
1.	11	Greenish Red	Light Brown	44	0.6±0.1
2.	12	Greenish Red	Light Brown	47	1.0±0.1
3.	13	Light Red	Light Brown	50	1.6±0.1
4.	14	Light Red	Light Brown	53	2.0±0.1
5.	15	Red	Light Brown	56	2.3±0.2
6.	16	Red	Light Brown	59	2.6±0.1
7.	17	Red	Deep Brown	56	2.4±0.1
8.	18	Red	Deep Brown	53	2.2±0.2
9.	19	Red	Deep Brown	50	1.9±0.1
10.	20	Red	Deep Brown	47	1.8±0.1
11.	21	Deep Red	Deep Brown	44	1.8±0.1
12.	22	Deep Red	Deep Brown	41	1.4±0.1
13.	23	Deep Red	Deep Brown	38	1.2±0.1
14.	24	Deep Red	Deep Brown	35	0.2±0.1
15.	25	Deep Red	Deep Brown	30	0.0±0.1

Table.24. Fruit maturity indices of *T. paniculata*
(MC: moisture content; GP: germination percentage)

Germinability during this period shows a pyramidal pattern including log or exponential growth phase and death or exponential decline phase. The growth phase happens from the 11th to 16th week, and the decline phase occurs from the 16th to 25th week. Compare the fruit and seed colour; the fruit and seed with maximum germinability identified appear in red and light brown, respectively, specified with Munsell Soil Colour Charts. As per Munsell's colour chart, the fruit's red colour is recognized by HUE 10 R red 4/6 (**Figure.36**). As you know, forest seed collectors are typically local villagers or tribal people who collect seeds for the forest department's nursery practices and research.



A

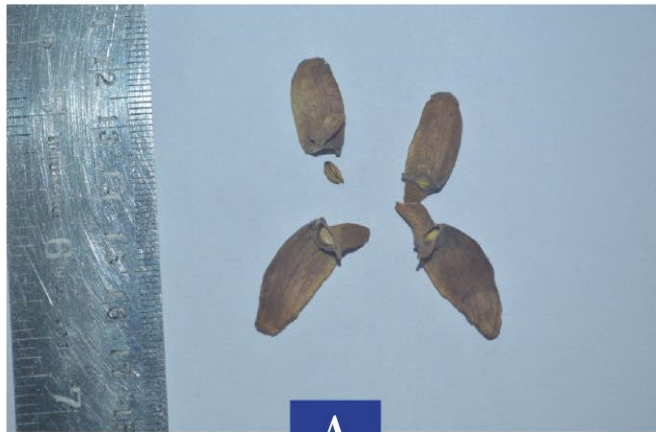


B



C

Figure.36. Fruits of *T. paniculata* collected from Karulai (A: Fruiting branch with ripened fruits; B: Single fruiting unit; C: Munsell colour chart with large fruit wing)



A



B



C

Figure.37. Seed of *T. paniculata*
(A: Seed filled fruits; B: Single seed; C: Seeds with scale)

I think identifying the best seeds through seed viability tests is difficult for local people and tribal people, especially for seeds with high seed emptiness. Simple identification methods like the Munsell colour chart are beneficial for all people. This information is helpful for further research and nursery practices to collect fruit directly from trees with maximum germinability easily for any people with the help of this simple colour chart.

Compared to any other *Terminalia* or tree genus in the study area, the colour of the fruit is beautiful with red colour, which is glossy from November to February. *T. paniculata* is commonly known as Kindal in the timber industry and is also known as Flowering Murdah. Murdah is the anglicized term of Maruva/ Maruth/ Maddi, which is the local name (Hindi, Kannada, Malayalam, Marathi, Tamil and Telugu) for *Terminalia* in Southern India. Massive, gigantic glossy red-coloured fruits during November and February give the name Flowering Murdah because people think that it's the flowering of the species. Also, because of the dominance of the species in the natural forests in Kerala, the fruiting season of *T. paniculata* is known as "when the woods bloom."

Seed Viability

Viability was calculated by conducting rapid cutting tests with the help of seed cutters manually using the seed collected from the Peechi Forest Range of Central Kerala. Eight seed lots were directed to cuttings tests, and seed viability ranged between 2% and 4%, and an average of 2.90% viable seeds were recorded. As Kerala Forest Research Institute recorded earlier, the seed emptiness is very high and ranges between 98% and 96% ($\bar{x} = 97.10\%$). Compared to other *Terminalia* in India, seed emptiness (98% to 96%) is exceptionally high in *T. paniculata* and produces many fruits (**Table.25**). High seed emptiness is a strategy to reduce seed predation by animals (Fuentus and Schupp 1998; Perera et al. 2013; Henn et al. 2014).

Sample No.	No. of Seeds	No. of viable seeds	Emptyseeds (%)	Viableseeds (%)
1.	1000	23	97.70	2.30
2.	1000	34	96.60	3.40
3.	1000	36	96.40	3.60
4.	1000	27	97.30	2.70
5.	1000	40	96.00	4.00
6.	1000	24	97.60	2.40
7.	1000	20	98.00	2.00
8.	1000	26	97.40	2.60
Mean	1000	28.80	97.10	2.90

Table.25. Viability of *T. paniculata* seeds from Peechi Forest Range.

1. Name of collector	Sanal C Viswanath
2. Type of source	Natural forest
3. Place of collection	Karulai
4. Forest Range and Division	Karulai and South Nilambur
5. Collection method	Pruning off seed-bearing plants
6. Altitude	58 m
7. Collection bag	Cotton sack
8. Period of collection	January 2017 (2ndweek)
9. Type of seed	Drupe (or Pseudo-samara)
10. Number of bags	06
11. Distance from the collection site to the laboratory	135 km

Table.26. Seed collection data from Karulai

Seed Morphometry and Moisture Content

Among the Indian winged fruit *Terminalia*, *T. paniculata* and *T. myriocarpa* are characterized by three-unequal wings. Both species are different in fruit size and size of the middle wing. Among three wings, the middle wing is large in *T. paniculata* and vice versa in *T. myriocarpa*. Seed dispersal occurs through hydrochory and anemochory; the dispersal is supported by the tiny size of the fruits and winged fruit character. *T.*

paniculata is the only three-winged *Terminalia* with a sizeable middle wing that supports the anemochorous seed dispersal mode. *T. paniculata* fruits or seeds measured using the seeds collected from six different populations in the Karulai Forest Range were recorded (Table 26-27).

Population	Large wing length (mm)	Large wing width (mm)	Small wings length (mm)	Fruit fresh mass (g)	Fruit dry mass (g)
TPP03	20.61 ± 1.18	12.44 ± 0.93	15.73 ± 3.06	0.21 ± 0.02	0.04 ± 0.01
TPP04	21.83 ± 1.09	13.35 ± 0.57	14.93 ± 2.78	0.21 ± 0.05	0.04 ± 0.00
TPP05	20.02 ± 0.63	12.50 ± 0.73	15.17 ± 1.69	0.17 ± 0.03	0.03 ± 0.01
TPP06	20.30 ± 0.89	12.18 ± 1.34	14.19 ± 2.05	0.17 ± 0.02	0.03 ± 0.00
TPP07	19.76 ± 1.48	12.32 ± 0.90	14.53 ± 2.71	0.17 ± 0.01	0.03 ± 0.01
TPP08	20.33 ± 0.90	11.80 ± 1.16	14.48 ± 1.81	0.16 ± 0.03	0.03 ± 0.01
Mean	20.47 ± 1.19	12.43 ± 1.02	14.84 ± 2.29	0.18 ± 0.03	0.04 ± 0.01

Table.27. Fruit characteristics of *T. paniculata* populations in Karulai.

The length of the large middle wing ranged between 19.76 mm and 21.83 mm, and the width ranged between 11.8 mm and 13.35 mm. Also, the length of the two small wings ranged between 14.19 mm and 15.73 mm. The fresh fruit mass (0.16 mg to 0.21 mg) and dry mass (0.03 mg to 0.04 mg) of fruits were recorded, showing that 81.25% to 80.95% of the fresh mass includes moisture content. Karulai forest range is part of the evergreen rainforests of the Nilgiris section of Western Ghats, India, and the high moisture content of seeds is may because of the location. The tiny size of the fruit supports Anemochorous and hydrochorous mode of seed dispersal. The mass of fruits/ seeds collected from the Karulai Forest Range indicates that one kilogram includes approximately 5555 fresh and 33333 dry seeds. The number may vary from location to location.



A



B



C

Figure.38. *T. paniculata* seed germination trials
(A: ripened fruits; B: seed sown in small trays, 100 seeds/ tray;
C: seed sown in large trays, 1000 seeds/ tray)

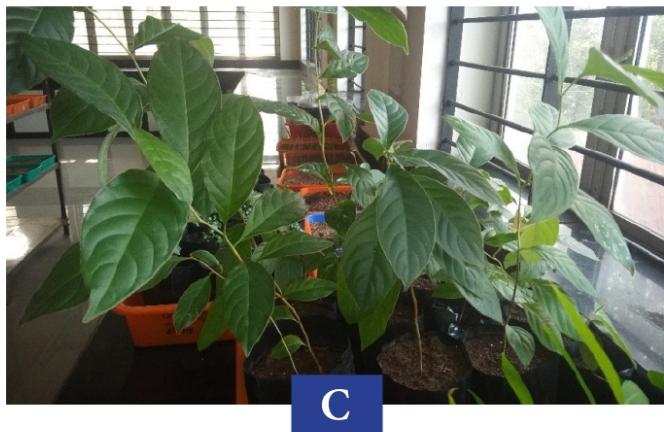
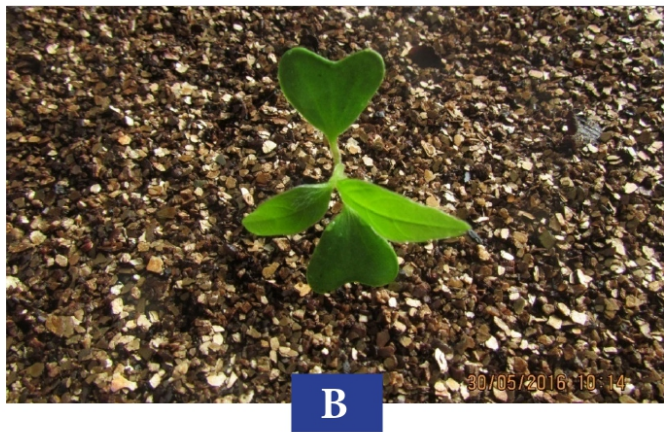


Figure.39. Seed germination stages in *T. paniculata* (A: first day of germination; B: four leaf stage; seedlings in polythene bags).

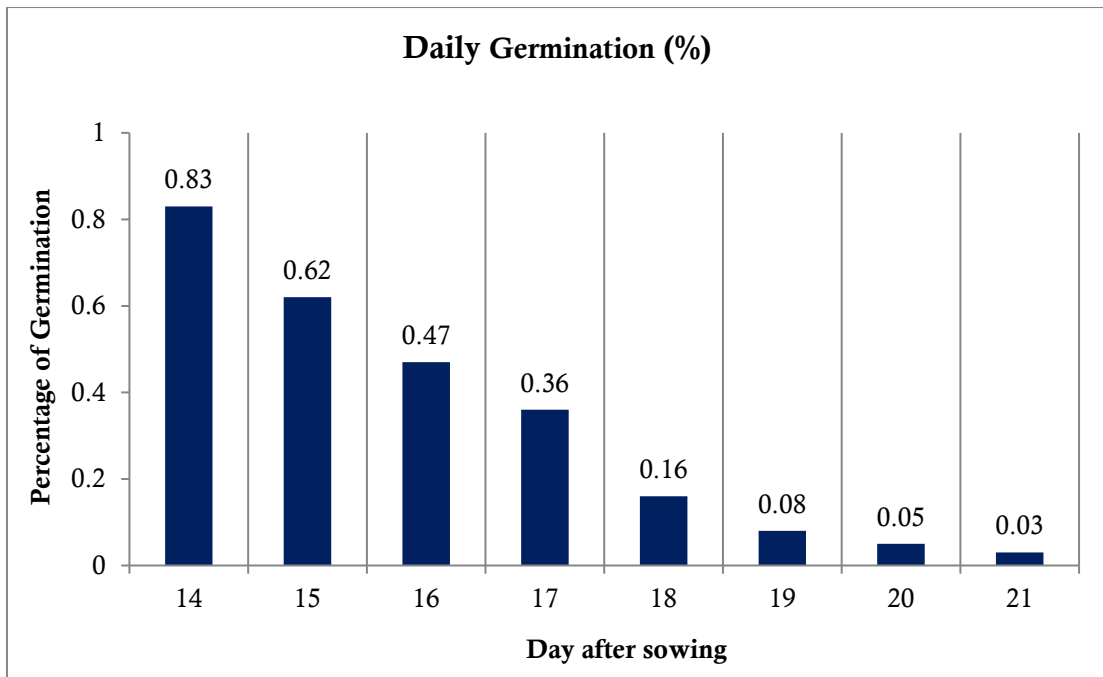


Figure.40. Daily germinability of *T. paniculata* seeds from Karulai

4.3.2. Germination Experiments

Germinability was calculated by conducting germination trials, and the results of the germination experiments show that initial germination occurs 14-days after sowing and germination end 21-days after sowing (**Figure.37-40**). Germination occur 14-days to 21-days after sowing, so one week is required to complete the germination. Maximum germination occurs on the first day of germination (14th day) is 0.83% and ends with minimum germination on the 21st day at 0.03%. Germination experiment reveals that the total germinability is 2.60% of seeds collected from Karulai Forest Range of South Nilambur forest division of Kerala. It's confirmed that maximum germination occurred during the 16th week of fruit development, and maximum germination is 2.60%. During the germination period, it's evident that a gradual decrease occurs in daily germination from the first day to the final day of germination. For this experiment, seeds of the 16th week of fruit development for germination trials and an average of 26 seedlings were germinated.

Sl. No.	Treatment	Seed type	No. of germinants out of 1000 seeds	Germination Percentage
1.	Fresh seeds (Control)	Winged	24	2.40
2.	Water soaking (12 hr)	Winged	24	2.40
3.	Water soaking (24 hr)	Winged	24	2.40
4.	Water soaking (48 hr)	Winged	25	2.50
5.	Water soaking (12 hr)	De-winged	26	2.60
6.	Water soaking (24 hr)	De-winged	25	2.50
7.	Water soaking (48 hr)	De-winged	25	2.50

Table.28. Pre-sowing results of *T. paniculata* seeds from Karulai

ANOVA Summary					
Source	Degrees of Freedom	Sum of Squares	Mean square	F stat	P value
Between groups	6	9.1429	1.5238	10.6649	0.002
Within groups	14	2.0003	0.1429		
Total	20	11.1433			

Table.29. ANOVA summary of germination percentage between treatments

4.3.3. Pre-Sowing Treatments

Pre-sowing treatments resulted in a maximum of 2.60% germination of *T. paniculata* seeds collected from the Karulai Forest Range of the South Nilambur Forest Division of Kerala (Table.28-29). Seven pre-sowing treatments were tried, including control. The study confirmed that a range between 24 and 26 germinants was recorded after sowing. Among the different pre-sowing treatments, seeds without any treatment (control) and water-soaked seeds for 12 hrs show 2.40% germinability, followed by water-soaked seeds for 48 hrs, water-soaked de-winged seeds for 24 hrs, and water-soaked de-winged seeds for 48 hrs with 2.50%. Maximum germinability of 2.60% occurred with water-soaked seeds for 24 hrs and water-soaked de-winged seeds for 12 hrs. Statistical

analysis of the germination rate results in no significant difference ($p > 0.005$) between the treatments.

Germination Percentage	Type of container					Mean
	Jute sack	Plastic sack	Cotton sack	Aluminium container	Plastic container	
04	2.20	2.30	2.20	2.40	2.40	2.30
16	2.10	2.20	2.10	2.30	2.30	2.20
25	1.80	2.10	1.80	2.20	2.30	2.00
Mean	2.00	2.20	2.00	2.30	2.30	

Table.30. Germinability of one-month stored *T. paniculata* seeds from Karulai.

ANOVA Summary					
Source	Degrees of freedom	Sum of squares	Mean square	F stat	P value
Between groups	04	0.244	0.061	2.7724	0.087
Within groups	10	0.220	0.022		
Total	14	0.4641			

Table.31. ANOVA summary of type of containers between treatments

4.3.4. Seed Storage and Longevity

Seed stored in different cold storage conditions in Kerala Forest Seed Centre. Seeds (16th week of fruit development) are stored in containers or sacks. Germination trials were conducted 1-month after storage, and results show that the germinability ranged between 1.80% and 2.40% (Table.30-32). Top seeds germinated after one month in an aluminium container and thick plastic container at 4°C and minimum germination occurred in seeds stored in jute and cotton sack at 25°C. The plastic container is the best

storage container compared to the jute sack, cotton sack, aluminium container, and plastic sack cover with 2.30%. Similarly, after one month of storage, germinability is high at 4°C at 2.30%. Comparing the temperature conditions and storage mediums, jute and cotton sack containers and room temperature are inadvisable.

Source	Degrees of freedom	Sum of squares	Mean square	F stat	P value
Between groups	02	0.172	0.086	3.5346	0.0621
Within groups	12	0.292	0.0243		
Total	14	0.464			

Table.32. ANOVA summary of different temperatures between treatments

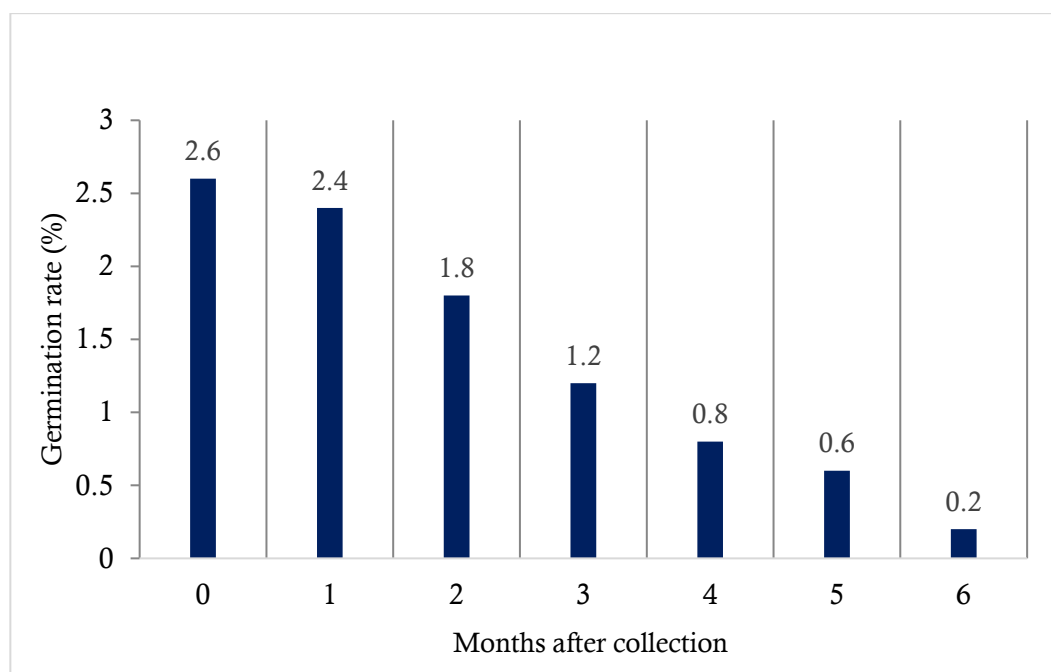


Figure.41. Germinability of stored seeds of *T. paniculata* from Karulai in different periods

Seed longevity experiments resulted in a seed that shows viability up to 180-days (6-months) using the seeds stored at 4°C temperature in a plastic container. The seed germinability pattern shows a gradual decline from 1st month after collection to the 6th

month and ends with 0.20% germinability (**Figure.41**). Germination starts with 2.60% germinability, followed by 2.40% germinability in the next month (**Table.33**). Even though the species shows a slight (0.20%) viability loss in 30-days, which is 7.60% of the maximum germinability. The sixth month ends with a viability loss of 2.40% of the maximum germinability, which is a 92.30% loss of the total. Hence, a slight decline in the viability of a high seed emptiness tree can significantly impact seed viability.

Months after seed collection	0	1	2	3	4	5	6
Germination (%)	2.60	2.40	1.80	1.20	0.80	0.60	0.20
Futile rate (%)	-	7.70	30.80	53.90	69.20	76.90	92.30
Difference (%)	-	0.20	0.60	0.60	0.40	0.20	0.40

Table.33. Germinability of stored seeds of *T. paniculata* from Karulai
(Stored temperature: 4°C; stored container: Plastic).

Carpology is a branch of biology that deals with the seeds and fruits with particular reference to the structure and morphology of seeds and fruits. But the study regarding seeds is generally referred to as seed biology, so carpology is a rare term. The present study covers the seed characteristics and handling techniques of *T. paniculata*, which can collectively be named as seed biology of *T. paniculata*. The significance of the investigation regarding seed biology of *T. paniculata* is that winged-fruit tropical tree species with three-unequal wings and trivial size and mass, high seed emptiness (between 96% and 98%) and low natural regeneration status (>2%), colossal flower and fruit production end with deep blood-red ripened fruits gives the name “Flowering Murdah” for the species and terms “When The Wood Blooms” is because of the massive fruiting of a large number of individuals. So the study regarding seed biology of *T. paniculata* is also a seed biological study of a species with winged fruits, high seed emptiness, low natural regeneration, and a tree named because of its fruiting character.

Scholars generally describe the fruit type of *T. paniculata* as drupe (Sasidharan, 2011). Scholars from the Hainan University of China recorded the diversity of Samara fruit type in Angiosperms, especially in trees. They classified samara fruit type into six categories: Single-winged Samara, Perigynous (Butterfly, Round Winged) Samara, Sepal winged Samara, Lanceolate winged Samara, Rib winged Samara and Bract winged Samara (Tan et al. 2018). Genus *Terminalia* include order Myrtales known for winged rib samara, Perigynous Samara and Lanceolate winged Samara fruit types. Even though the fruit of *T. paniculata* looks like a Samara, but can't mark the fruit under any category of Samara. So the study suggests the term Pseudo-samara as the fruit of *T. paniculata*.

Studies regarding reproductive phenology, seed maturation, and colour-based fruit maturation index are the basic study supporting the estimation of fruit production and fruit maturity index (Schimdt 2007). Fruit color-based maturity studies on Indian Beach (*Pongamia pinnata*), Divi-divi (*Caesalpinia coriaria*), Himalayan Firethorn (*Pyracantha crenulata*) and Sandal (*Santalum album*), etc. confirmed that fruit colour is virtuously related to fruit maturity. Light brown pod and brown seeds give maximum germinability (84%) in Indian Beach (Srimathi et al. 2013). At the same time, dark brown pods provide maximum germinability (44%) in Divi-divi (Deepakkumar and Ramanan 2016). A similar Himalayan Firethorn experiment resulted in maximum germinability (84%) detected with light orange fruits (Shah et al. 2006). Fruit maturity studies in the Sandal resulted in maximum germinability observed with black (59%) and reddish-brown seeds (56%) (Manonmani and Vanangamudi 2001).

The present study also reveals the colour-based fruit maturity index of *T. paniculata*. Maximum germinability (2.60%) was ensured during the 16th week of fruit development, characterized by red fruits (HUE 10R red 4/6) and light brown seeds. Only 28% of flower buds attained the ripened fruit stage. Many methods are available to identify the colour in connection with the estimation of the fruit maturity index. The fruit

colour of *T. paniculata* was determined using the Munsell colour chart (Reeder et al. 2014).

After estimating the fruit maturity index and fruit production pattern, seed viability and seed emptiness of *T. paniculata* were estimated. Former studies regarding germinability recorded high seed dormancy and low germinability of *T. paniculata* (Murali 1997; TNFD 2005; Pillai and Chandrasekhara 2011; Pillai 2017). According to Perea et al. (2013), seed emptiness is a mechanism of plants, especially trees, to prevent seed predation by arboreal animals and birds. Seed fertility studies in Axlewood (*Anogeissus latifolia*) reported that the species showed about 95% seed emptiness (Singh et al. 2015). *Anogeissus* is considered the synonym of the genus *Terminalia* by the royal botanical garden Kew.

Hence Axle wood is a sister species of *T. paniculata*. Viability analysis of *T. paniculata* resulted in seed emptiness ranging between 98% and 96% ($\bar{x} = 97\%$) and viability ranging between 2% and 4% ($\bar{x} = 2.90\%$). The present study regarding seed germinability of *T. paniculata* confirmed the former studies (**Table.34**). Seed emptiness of *Terminalia* (*T. cuneata*, *T. bellerica*, *T. catappa*, *T. chebula*, *T. elliptica* and *T. travancorensis*) ranged between 80% and 40% (Chacko et al. 2002). As per the literature reviews and results of the present study, *T. paniculata* is the species that belongs to the particular genus with high seed emptiness and is also one among tropical angiosperm trees (Emanikor et al. 2020). Teak (*Tectona grandis*), one of the associate species of *T. paniculata* shows high seed emptiness ranges between 80% and 60% (Chacko et al. 2002).

Sl. No.	Germination percentage	Germination duration	References
1.	00.75	-	Pillai and Chandrasekhara 2011
2.	02.00	-	Chacko et al. 2002
3.	02.60	21 days	Present study
4.	15.00	-	Murali 1997
5.	20.00	25 days	TNFD 2005

Table.34. Germination percentage of seeds of *T. paniculata*

Anything regarding seed biological study doesn't complete without explaining the seed sample size. According to the forest seed testing rule seed germinability studies need between 40 and 100 seeds per sample (Mulawarman et al. 2003; Chacko 2009; Ribeiro-Oliveira et al. 2016). But in the case of seeds with high seed emptiness, a sample with 40 to 100 seeds is not enough for seed biological studies; hence, 1000 seeds are selected per sample and eight samples per ISTA rule (ISTA 2018). As per the ISTA rule, the number of seeds per sample is purely based on seed emptiness, seed mass and size and germinability. Hence it's projected that, except for all other *Terminalia*, *T. paniculata* required 1000 seeds per sample for conducting seed experiments.

Sl. No.	Species	Dimension	Weight (fruits/kg)	Emptiness
1.	<i>T. arjuna</i>	4-5 cm x 2.50- 3 cm	176- 375	Low
2.	<i>T. bellerica</i>	1.50 cm x 2.70 cm	97- 176	Low
3.	<i>T. catappa</i>	5- 6 cm x 3- 4 cm	150- 850	Low
4.	<i>T. chebula</i>	2.50- 4 cm x 1.50- 2 cm	141- 220	Low
5.	<i>T. crenulata</i>	3- 4 cm x 3.50 cm	441- 551	Low
6.	<i>T. paniculata</i>	15 x 8 mm (large wing)	26103- 59966	97%
7.	<i>T. paniculata</i> (Present study)	19-21 mm x 11-12 mm (large wing)	25000- 33333	96-98%

Table.35. Seed characteristics of selected *Terminalia* in India (Chacko et al. 2002)

Fruit morphometry of *T. paniculata* documented the dimension of large fruit wing length and width, fruit small wings length, and fruit fresh and dry mass. The research entitled “Patterns of seed size, germination and seed viability of tropical tree species in Southern India” is one the original research regarding seed biology. Murali (1997) recorded the seed weight (0.20 g), viability (180-days), and germination days (15-days). The present study recorded seed fresh (0.18 ± 0.03 g) and dry mass (0.04 ± 0.01 g) of *T. paniculata* collected from the Karulai Forest Range of Nilambur South Forest Division. Dimensions of fruit wings, such as the length of the large wing (20.50 ± 1.20 mm), the width of the large wing (12.40 ± 1 mm), and the length of small wings (14.80 ± 2.30 mm) recorded in the first time. Seed dimension and fruit weight of selected Indian *Terminalia* confirmed that *T. paniculata* is small in dimension and seed weight with high seed emptiness (Table.35).

Sl. No.	Species	Pre-sowing Treatments	Ger. %	References
1.	<i>T. bellerica</i>	Cold-WS for 48 hrs.	93	Hossain et al. 2014
2.	<i>T. catappa</i>	WS and drying of de-pulped seeds (6 days; 12 hrs interval) and soaking in 2% CaOCl ₂ (12 hrs).	98	Masilamani et al. 2013
3.	<i>T. chebula</i>	Nicking the broad end and WS (36 hrs).	76	Benjamin et al. 2019
4.	<i>T. cuneata</i>	10KRF+ Pre-soaking.	85	Wani & Singh 2018
5.	<i>T. elliptica</i>	Complete seed scarification.	97	Negi & Todaria 1995
6.	<i>T. ivorensis</i>	Soaking in 2M H ₂ SO ₄ .	37	Amponsah et al. 2018
7.	<i>T. laxiflora</i>	High temperature (78 ^o C) treatment for 10 min.	80	Mewded et al. 2018
8.	<i>T. myriocarpa</i>	No need.	37	Bahuguna et al. 1987
9.	<i>T. paniculata</i>	No need	2.60	Present study
10.	<i>T. sericea</i>	Nicking and WS for 12 hrs.	13	Likoswe et al. 2008
11.	<i>T. superba</i>	Soaking in 2M H ₂ SO ₄ .	84	Amponsah et al. 2018

Table.36. Pre-sowing treatment of selected *Terminalia* (WS: water soaking; Ger.: Germination).

Seed dispersal of *T. paniculata* was recorded as an Anemochous mode of distribution (Tadwalkar et al. 2012). Anemochorous mode of seed dispersal occurs supported by the three wings mentioned above. Not only a dispersal supporting unit, but the wings of *T. paniculata* are also a critical taxonomic character to identify the species using seeds or fruits (3-unequal winged) among the *Terminalia* and winged fruit trees (Deshpande and Yadav 2017).

Genus *Terminalia* produces two types of fruit types such as winged and non-winged fruits. Within the winged fruit category, species are diverse in terms of the number and size of wings. De-pulping followed by water-soaking is the superlative pre-sowing treatment for wingless fruits, and de-winging followed by water-soaking is the excellent pre-sowing treatment for winged fruits (Chacko et al. 2002; Pillai and Chandrasekhara 2011) (**Table.36**). Germinability of both winged and wingless fruits ranges between 35% and 85% with pre-sowing treatments, too (Chacko et al. 2002). Based on wing character, *T. paniculata* is closely associated with *T. myriocarpa* because it also has three-winged fruit. A germination study on *T. myriocarpa* concluded that no pre-sowing treatment is needed for a maximum germinability of 38% (Bahuguna et al. 1987). Meantime, *T. paniculata* resulted in 2.60% germination only and required 21-days to complete germination after sowing. Seed germination experiments on *T.paniculata* recorded values ranging between 0.75% and 20% and duration of 25-days.

Pre-sowing treatments always aim to enhance seed germinability and reduce germination duration, which is very effective in the genus *Terminalia*. Seed germinability of winged fruit *Terminalia* like *T. cuneata*, *T. elliptica* confirmed that pre-sowing treatments have a high impact on germinability (Chacko et al. 2002). *T. laxiflora* is also a winged-fruit tree species native to the African subcontinent and shows enhanced seed germinability (80%) by the impact of pre-sowing treatment, seeds in 78°C for 10 min (Mewded et al. 2018). Pre-sowing treatment (nicking and water soaking for 12 hrs) on *T.*

sericea resulted in 13% seed germinability (Likoswe et al. 2008). But a present study on *T. paniculata* confirmed that pre-sowing treatments don't impact seed germination. Germination studies on *T. myriocarpa* show that there is no need for pre-sowing treatments, which supports the present study (Bahuguna et al. 1987; Bisht et al. 2001).

Cold storage is the best seed storage condition and is generally adopted throughout the tropics by keeping the seed without any change in moisture content. Seed storage studies on genus *Terminalia* resulted that seeds can be viable for a period, which varies from one species to another, and cold storage is the best storage method (Chacko et al. 2002) (Table.37). Seed storage studies on *T. superba* resulted in hermetic glass bottles being better than plastic bottles, and freezer and refrigerator temperatures are ideal for storage (Asomaning 2020). The present study concluded that cold (4°C) storage is the best method, and an aluminium or plastic container is the best storage medium for storing the seeds of *T. paniculata*. Seed storage studies of *T. paniculata* also revealed that viability lasts for 6-months and ends with 0.20% seed germinability, which supports the study by Murali (1997).

Sl. No.	Species	Seed Longevity	Reference
1.	<i>T. bellerica</i>	360-days	Murali, 1997
2.	<i>T. cuneata</i>	360-days	Murali, 1997
3.	<i>T. elliptica</i>	360-days	Murali, 1997
4.	<i>T. myriocarpa</i>	180-days	Bahuguna et al. 1987
5.	<i>T. paniculata</i>	180-days	Murali, 1997
6.	<i>T. paniculata</i>	180-days	Present study

Table.37. Seed longevity of genus *Terminalia* distributed in India

Seed longevity experiments resulted in seeds showing viability for up to 180-days using the seeds stored at 4°C temperature in a plastic container. Seed germinability

pattern shows a gradual decline from 1st month after collection up to 6th month and ends with 0.20% germinability. Germination starts with 2.60% germinability, followed by 2.40% germinability in the next month. Even though the species shows a slight (0.20%) viability loss in 30-days, which is 7.60% of the maximum germinability. The sixth month ends with a viability loss of 2.40% of the maximum germinability, which is a 92.30% loss of the total. Hence, a slight decline in the viability of a high seed emptiness tree can significantly impact seed viability.

4.4. Development of a protocol for propagation in *T. paniculata*.

4.4.1. Propagation through seeds

For the experiment, the fruits/seeds of 16 weeks of fruit development were collected and sown in different mediums. Initial germination starts 13- 15 days after seed sowing and ends in 21-26 days after seed sowing (**Table.38**). Seed germination rate ranged between $2.40\pm 0.20\%$ and $2.60\pm 0.10\%$, and germination duration ranged between 21 ± 1 -days and 26 ± 2 -days. Seeds in soil medium show the longest germination duration (11 days) followed by sand, soil-sand and sand vermiculite mediums (10 days). Seeds in soil medium show the low germination (2.40%) followed by soil-sand and sand-vermiculite (2.50%). ANOVA results that both germination duration and germination percentage shows significant (p -value < 0.05) variation between the treatments (**Table. 39-40**). The observations indicated that vermiculite shows least germination duration and best for early germination and germination rate.

Sl. No.	Sowing medium	Germination in days			Germination percentage	Mean daily germination
		Initial	Final	Duration		
1.	Soil	15	26	11	2.4	0.09
2.	Sand	14	24	10	2.6	0.11
3.	Vermiculite	13	21	08	2.6	0.12
4.	Soil- Sand	15	25	10	2.5	0.10
5.	Soil- Vermiculite	14	23	09	2.6	0.11
6.	Sand- Vermiculite	14	24	10	2.5	0.10

Table.38. Germinability of *T. paniculata* seeds from Karulai

The development of propagation protocols is an advanced phase of a tree breeding program to produce many individuals within a limited period using a minimum number of resources and expenses. Usually, angiosperm trees are propagated through seeds. Sometimes propagation through seeds will not happen due to different types of reproductive constraints. *Terminalia* is a genus that belongs to the order Myrtales, propagated commonly through seeds. However, because of the multipurpose uses of the trees, researchers have attempted different types of propagation methods for the production of a large number of individuals. Propagation protocols include seed and vegetative propagation, such as natural and artificial means. Vegetative propagation through natural means is not recorded in the genus *Terminalia*. The present study evaluates the scope of vegetative propagation through artificial standards such as cutting, layering, etc.

ANOVA Summary

Source	Degrees of Freedom	Sum of Squares	Mean Square	F-stat	P-value
Between groups	05	19.1662	3.8332	13.7973	0.0001
Within groups	12	03.3339	0.2778		
Total	17	22.5001			

Table.39. ANOVA summary of germination duration between the treatments

ANOVA Summary

Source	Degrees of Freedom	Sum of Squares	Mean Square	F-stat	P-value
Between groups	05	0.105	0.0210	12.6141	0.0002
Within groups	12	0.020	0.0017		
Total	17	0.125			

Table.40. ANOVA summary of germinability between the treatments

Seed propagation is the standard method of plant propagation than any other method like vegetative propagation. Seed characteristics such as reproductive phenology, production pattern, dispersal and viability, germination, and regeneration rate will vary from one species to another and within the species due to biotic and abiotic factors. The reproductive phenophases of *T. paniculata* located within the campus of Kerala Forest Research Institute show a wide range of variance in terms of the age and size of the trees. The reproductive phenophases of the individuals on campus start during the first week of September up to the 2nd week of April (7.5-months). But a single individual on the campus starts flowering during September (4th week) and ends during March (4th week); hence 6-months are required to complete the reproductive phenophases.

Seed germination of genus *Terminalia* ranges between 37% and 98% with or without pre-sowing treatments. Seed germination of wingless *Terminalia* (*T. bellerica*, *T. catappa*, *T. chebula*) ranged between 76% and 98%, and winged *Terminalia* (*T. cuneata*, *T. elliptica*, *T. myriocarpa*) ranged between 37% and 85%. Compared to genus *Terminalia*, seed germination of *T. paniculata* is very low. Sister species, *T. myriocarpa* showed 37% seed germinability without any pre-treatments. The present study concluded that seeds during the 16th week of fruit development are the best seed collection period, and there is no need for pre-treatment. Germination experiments also concluded that germination is independent of the nature of sowing mediums. High germination rate ($2.60 \pm 0.10\%$) and

short duration (21 ± 1 -days) were recorded in a vermiculite medium. Low germination rate ($2.40\pm 0.20\%$) and long time (26 ± 2 -days) were observed with pure soil medium.

➤ **Propagation using stem cuttings and epicormic shoots**

There was no rooting in young stem cuttings (**Table.41**). All treatments except epicormic shoots treated with IBA 500 ppm failed to root (**Table.42**). Hence, stem cuttings and epicormic shoots treated with plant rooting hormones in different concentrations is not advisable for *T. paniculata* vegetative propagation. 26.66% of epicormics shoots treated with 500 ppm IBA had rooting and so this treatment is success (**Figure.42**). Present study also supported that vegetative propagation practises are futile in *T. paniculata* except epicormics treated with 500 ppm IBA. A study by the IISc Bangalore, already listed *T. paniculata* and *Lagerstroemia microcarpa* as one of the most problematic tree species in the Western Ghats regarding propagation through vegetative cuttings and seeds and present research supporting the experiment by IISc.



Figure.42. Stages of vegetative propagation using epicormic shoots in *T. paniculata* (A and B: Development of epicormic shoots from stem cuttings; C and D: Hormone treated epicormic shoots in root trainers; E: rooted epicormics; F: 1-year old plants).

Treatment	T01	T02	T03	T04	T05	T06	T07	T08	T09	T10	T11	T12	T13	T14	T15	T16
Rooted cuttings	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Percentage of rooted cuttings	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Table.41. Rooting percentage of hormone treated stem cuttings.

Treatment	T01	T02	T03	T04	T05	T06	T07	T08	T09	T10	T11	T12
Treated epicormics (in no.s)	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3	10 x 3
Rooted epicormics (in no.s)	8	0	0	0	0	0	0	0	0	0	0	0
Rooting (in %)	26.66	0	0	0	0	0	0	0	0	0	0	0

Table.42. Rooting percentage of IBA treated epicormics.

A high amount of phenolic compounds is recorded in *T. paniculata* especially the deep blood red coloured fruits, because of the presence of tannin. Probably the presence of tannin in the cut end of the plant, rooting hormones can't have an impact on rooting. Hence the application of plant hormone on vegetative cuttings is futile even planted in wood charcoal mixed vermiculite.

Propagation experiments on Indian *Terminalia* confirmed that vegetative propagation is successful in *T. chebula*, *T. cuneata*, *T. myriocarpa*, etc, with the support of rooting hormones in different concentrations. Propagation experiments on *T. chebula* concluded that apical stem cuttings treated with rooting hormone could be helpful for the mass production of seedlings (Jose and Thomas 1993). A similar experiment in *T. cuneata* concluded that shoot cuttings treated with rooting hormone could be used for mass production (Kusum et al. 2017). An investigation on *T. myriocarpa* concluded that

woody branch cuttings without rooting hormone resulted in low rooting (Kumar et al. 2011). Generally, the genus *Terminalia* has both successful and futile vegetative propagation attempts (Bhardwaj et al. 1993). The present study on *T. paniculata* also concluded that vegetative propagation using woody branch cuttings and epicormics results in 26.66% rooting of epicormics. The study suggests that epicormics treated with rooting hormone can be used for mass production for planting activities.

Propagation through air layering

Compared to Indian *Terminalia*, air-layering is successful in *T. cuneata* only, and the present experiment confirmed the earlier report (Singh and Jolly 1969). In *T. cuneata*, layering resulted in a 92% survival rate after transplanting the rooted layers after 30-days. No rooting was observed as the result of air layering in *T. paniculata* located on the Kerala Forest Research Institute campus. The lack of root formation could be due to the high amount of phenolic compounds.

Experiments on *T. paniculata* using seeds, vegetative cuttings and air-layering resulted in propagation through seeds, and vegetative propagation using epicormic shoots treated with 500 ppm IBA is successful. *T. paniculata* produce a large number of seeds to overcome the reproductive constraints due to high seed emptiness. Because of the availability of many seeds, propagation through seeds is advisable. Seeds of *T. paniculata* are also readily available due to the dominant nature of the tree species in Kerala. Epicormic shoots treated with low concentrated IBA are also effective for the large-scale production of *T. paniculata* seedlings. Scholars developed propagation protocols for large-scale production of forest trees, especially in the case of forest trees, to overcome reproductive constraints and effectively present a large number of seedlings. The present study suggests propagation of *T. paniculata* through seeds and epicormic shoots through the protocol mentioned above for many seedling productions.

CHAPTER 5
SUMMARY AND CONCLUSION

Chapter 5

Summary and Conclusion

The present study on plus tree selection, variation, seed characteristics and development of propagation protocol of *Terminalia paniculata* has been carried out to select the plus trees of *T. paniculata* from the different populations in Kerala, to assess the variation between diverse populations, to analyze the seed characteristics of the target species and to develop propagation protocol for the large-scale production of the species. The study was conducted in the Kerala part of Peninsular India. The experiments were carried out in the laboratory of the Forest Genetics and Tree Breeding department and the laboratory of Kerala Forest Seed Centre at Kerala Forest Research Institute, Thrissur, Kerala, India, from 2016 to 2021.

T. paniculata is endemic to peninsular India, is the dominant species throughout Malabar Coast, and is one of the globally important commercial timber species recognized by botanic garden Conservation International, Food and Agricultural Organization, etc. The species shows high seed emptiness. Like other Terminalias, *T. paniculata* is a timber species (household, industrial, and tertiary sector) well known for its multipurpose usage as economically significant green manure, ethno medicine, tannin, and dye production raw material and ecologically important host tree for many faunal species. Even though the species is dominant and widely distributed throughout Kerala, natural populations outside protected areas have been declining due to many anthropogenic activities. *T. paniculata* shows high seed emptiness and low germination. Thus, studies on seed characteristics and development propagation protocols are essential for a successful tree improvement program.

Plus trees are the phenotypically superior trees in terms of growth rate, high wood quality, and resistance to diseases and pest attacks. Plus trees were selected from candidate plus trees, which were chosen from the base population. A detailed population

survey was conducted throughout the study area. Even though *T. paniculata* is a dominant commercial timber tree species, no plantations of this species have been developed so far. From the selected base populations, candidate plus trees were selected using stem straightness, clear bole height, diameter, forking, branch angle and thickness, self-pruning, fluting, straight grain, disease and Epicormic shoots. The regression selection method was used to select plus trees from the identified candidate plus trees, and characters like circumference at breast height, total height, and crown diameter and crown length were used.

Studies were conducted using different characters to identify the variation between diverse populations. It included characteristics that can be easily observed and measured at any season of the year independent of different phenophases (circumference at breast height, total tree height, crown length, crown diameter, primary branches, and secondary branches), characters that can be understood and measured only by tests (bark thickness, sapwood moisture content, sapwood density, leaf chlorophyll content, leaf area, leaf fresh mass, and leaf dry mass) and Characters that are not directly connected with timber production (fruit large wing length, fruit large wing width, fruit small wings width, fresh fruit mass, and fruit dry mass). Statistical analyses like ANOVA, correlation, principal component, and cluster analysis were performed using different statistical software.

Study on seed characters and seed handling techniques, including production pattern of flowers and fruits, maturity index, collection and morphometry, moisture content, viability, seed germinability and pre-sowing treatments, longevity, and storage of seeds were studied. All the experiments were conducted at the laboratory of Kerala Forest Seed Centre, Peechi. Trees located in Peechi Forest Range, Kerala Forest Research Institute Campus and Karulai Forest Range was used in the study.

Different propagation techniques were used to develop a propagation protocol for *T. paniculata*, including propagation using seeds, vegetative cuttings, epicormic shoots, and air-layering. Seeds collected from six populations of Karulai Forest Range were used for propagation studies using sources. Cuttings taken from matured trees located at Kerala Forest Research Institute were used for the propagation experiments using vegetative cuttings and epicormic shoots. Phytohormones like NAA and IBA were used for these experiments. Air-layering was performed on six mature trees located in Kerala Forest Research Institute.

The tree improvement program involves selecting species, assessing variation, improving desired characters, and large-scale production of the improved individual. For the success of every tree breeding program, knowledge regarding the selected species is obligatory. *T. paniculata* is endemic to peninsular India, recorded from Karnataka, Maharashtra, Tamil Nadu and Kerala states. The species is commonly distributed in Kerala and Karnataka parts of Peninsular India. Due to high anthropogenic pressure, natural populations of *T. paniculata* outside the protected area, including home gardens and agricultural lands, drastically declined. Plus tree selection is the beginning of tree breeding, and different methods scholars adopt for selecting plus trees. Based on the literature survey, a detailed population survey was conducted throughout the study area from May 2016 to December 2017. Sixteen populations were identified through the extensive field survey, and candidate trees were identified from these populations using selection criteria. Sixteen plus trees were selected using the regression selection method.

Wild tree species have a large amount of genetic variation, which is very useful in tree breeding programs. Like all other tree breeding program, the purpose of the program is to meet the demand for timber of *T. paniculata* through tree improvement approaches. Plus tree selection is the breeding program, and different methods scholars adopt to select

plus trees. The primary tree breeding program of forestry trees, including Terminalias conducted in India, is done through the comparison tree method and is based on many variables. Rudolf proposed a plus tree selection through regression selection method, which is efficient for selecting plus trees from uneven-aged populations. The regression selection method was chosen because plantations are not yet developed, and the individuals' ages are unknown. This method is independent of the behaviour of tree breeder or geneticist and based on statistical backgrounds, and only four variables are required for the selected program.

Even though the species is dominant throughout Kerala, only 16 plus tree populations have been recorded from the state; among the 16 plus tree populations, six belong to Karulai Forest Range, and five belong to Parambikulam Tiger Reserve. These two regions are the significant pockets of *T. paniculata* and are located in two agro-ecological zones such as Foothills (Walayar and Erumeli) and High Hills (Parambikulam, Sungam, Karulai, Peechi, and Aryankavu). A minimum of five candidates plus trees were selected from each population for a further plus tree selection program. The growth characteristics of each tree were recorded. Tree selection through regression method designated for selecting plus trees from candidates used for this study. Compared to the comparison tree method, the regression method of plus tree selection is purely determined by four variables: tree height, girth, crown diameter, and length. Also, regression analysis is suitable for both aged-known and aged-unknown tree individuals and is independent of human behaviour. While comparing the growth characteristics of plus trees and candidate plus trees of *T. paniculata* in Kerala, candidate plus trees show supremacy over circumference at breast height and total tree height.

Genetic variation is the naturally occurring genetic differences among individuals of the same population. These variations permit a population's flexibility and survival in the face of changing environmental circumstances. Most tree characters were connected with their heredity to a certain degree, while ecological factors highly modify some. This

study estimated variation between the selected populations by analyzing the variation in the phenotypic characters. Eighteen phenotypic characters were used for calculating the variation between the chosen people. These characters include the characters that can be observed and measured throughout the year and those that can be understood and measured by experiments and tests. Statistical analyses like correlation analysis, analysis of variance, and principal components were performed to analyze the variation.

The correlation analysis showed that circumference at breast height shows the maximum number of meaningful bonds with seven tree characters, and the number of primary branches shows no significant bond with any tree characters. The wood characteristics such as bark thickness, sapwood moisture, and sapwood density showed a meaningful relationship with other tree characters under study. The fruit characteristics such as large wing length, large wing width, tiny wings length, fruit fresh mass, and fruit dry mass also significantly correlated with other factors. By comparing all the correlation relationships between the 18 tree characters, it's evident that the number of primary branches and leaf chlorophyll content doesn't show any significant bond with the other characters.

Principal component analysis was performed to determine the variation in data by defining the minimum number of principal components. Eigenvalue, proportion, and scree plots were used to assess variation. Eigenvalue of principal components from PC1 to PC18 reveal that the first five principal components have eigenvalue of more than one. They are the real asset of the variability in the data and were selected for further analysis. Eigenvectors of principal components with 18 tree characters resulted from a correlation between principal components with tree characters. Eigenvalue plotted against the component number in scree plot reveals the eigenvalue of principal components PC1 to PC18. The score plot reveals that populations located in Peechi (TPP12 and TPP13) show similarities with Sungam (TPP14 and TPP15) and Aryankavu (TPP1). Also, populations located in Parambikulam (TPP9, TPP11 and TPP12) show similarities with

populations located in Walayar (TPP16) and Erumely (TPP2). Populations in the Karulai Forest Range (TPP3, TPP4, TPP5, TPP6, TPP7 and TPP8) placed together in the score plot also show the similarity between populations.

Multivariate classical cluster analysis of tree functional characteristics resulted in prominent hierarchical clustering of 16 populations of *T. paniculata* in the Kerala part of Peninsular India. Among 16 selected populations of *T. paniculata* were assembled into an alpha cluster and beta cluster. The alpha cluster contains eleven populations, and it is subdivided into sub-clusters. The beta cluster contains five populations and is subdivided into two sub-clusters within 37.50 Ed. Among the selected populations, TPP13 is the single population that left the cluster within 37.50 Ed. Even though the different populations fall under other agro-ecological units, cluster analysis was contradictory.

Study on seed characters and handling techniques, including production pattern of flowers and fruits, maturity index, collection and morphometry, moisture content, viability, seed germinability and pre-sowing treatments, longevity, and storage of seeds were studied. An average of 30.2 flower buds initiated at the first stage of flower phenophase ends with an average of 17 opened flowers per inflorescence. It seems that 56.30% of flower buds developed into mature flowers. Out of the total number of flower buds per inflorescence generated, only 27.80% developed into ripened fruits. The reproductive phenological pattern shows that approximately 25% of the flower buds attain the matured fruit stage. Fruit maturity was assessed by analyzing the fruits of *T. paniculata* at a regular interval of once a week. Seed is viable during the 11th to 25 weeks of fruit development selected for assessing fruit maturity index. The maximum germinability was recorded during the 16th week of fruit development. As per Munsell's colour chart, the fruit's red colour is recognized by HUE 10 R red 4/6.

Seed viability was calculated by conducting rapid cutting tests and recording an average of 2.90% viable seeds. Seed morphometry of *T. paniculata* reveals that the fruit

has three-unequal wings. Among the three wings, the moderate wing is large, and the lateral wings are small. One kilogram includes approximately 5555 fresh seeds and 33333 dry seeds. The number may vary from one location to another. Germinability was calculated by conducting germination trials, and the results of the germination experiments show that initial germination occurring 14-days to 21-days after sowing. Maximum germination occurs during the 16th week of fruit development, and maximum germination is 2.60%.

Among the different pre-sowing treatments, seeds without any therapy and water-soaked seeds for 12 hrs show 2.40% germinability, followed by water-soaked seeds for 48 hrs, water-soaked Dewinged seeds for 24 hrs, and water-soaked de-winged seeds for 48 hrs with 2.50%. Maximum germinability of 2.60% occurred with water-soaked seeds for 24 hrs and water-soaked de-winged seeds for 12 hrs. Germination trials were conducted 1-month after storage, and results show that the germinability ranged between 1.80% and 2.40%. Top seeds germination was observed in aluminium containers and plastic containers at 40C and minimum germination occurred in seeds stored in jute and cotton sacks at 25^oC.

Every tree breeding program aims to mass-produce quality seedlings using vegetative propagation protocol at a low cost. Seed propagation is the standard method of plant propagation than any other method like vegetative propagation. Fruit maturity index calculated as part of seed characteristics study confirmed that during the 16th week of fruit development shows comparatively very high germination value (2.60%) in vermiculite medium at room temperature. High germination rate and short duration were recorded in vermiculite medium meantime low germination rate and long term was observed with pure soil medium.

Vegetative propagation techniques like propagation using young stem cutting, stem cutting, epicormic shoots and air layering. Propagation using young stem cuttings

treated with artificial plants rooting hormones such as IBA and NAA in two different methods: the dipping method and the soaking method, was futile by lacking root formation. Like rooting hormone treated with stem cuttings, epicormic shoots treated with plant rooting hormone IBA were tried. All other treatments in the dipping method get futile by lacking root formation. Both vegetative propagation experiments by treating the stem cuttings and epicormic with artificial plant rooting hormones like IBA and NAA were ineffective and didn't result in rooting or root formation. Only epicormics treated with IBA 500 ppm resulted in a minor success rate (26.66%). The air layering method was also tired to develop the less expensive and time-consuming propagation practices for mass production.

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