PHENOLOGY AND GERMPLASM VARIATION OF *Tetrapleura tetraptera* (SCHUM. AND THONN.) TAUB.

BY

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DEDICATION

This thesis is dedicated to my late mum of blessed memory, Alhaja Alirat Iyabo Adeyanju, who by the grace of Almighty Allah made this work a reality. May Allah grant her soul eternal rest.

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ABSTRACT

Tetrapleura tetraptera is a tree species of great ethnobotanical importance that is highly utilised and as such its natural population is threatened. For sustainable utilisation of the species, there is need for development of protocols for its domestication. However, critical information on species phenology, germplasm characteristics and macro-propagation, which are required for the domestication process are limited. Therefore, the flowering and fruiting patterns; germplasm variation and macro-propagation techniques for *Tetrapleura tetraptera* were investigated.

Tetrapleura tetraptera trees were purposively selected from Forestry Research Institute of Nigeria (FRIN) and National Horticultural Research Institute (NIHORT) in Ibadan for phenological studies, based on availability. Onset and duration (days) of flowering and fruiting, number of flowers/inflorescence and synchrony (*Z*), were monitored on the trees for 20 months. Visiting insects, frequency of visits, pollen load and fruiting efficiency were assessed. For germplasm variation, 150 fruits each were purposively collected from Ibadan, Mamu, Iwo and Aponmu within the rainforest zone, based on access. Variation in fruit- length (cm), width (mm); number of seeds (NS) and seed weight (g) were determined for each location. In a completely randomised design experiment, height (cm), diameter (mm), and number of leaves of 50 uniformly growing seedlings were measured for six months. Stem cuttings (top, middle and base) obtained from seedlings from all locations were macro-propagated. Cuttings survival, number of roots/cutting (NR) and root length (RL, cm) were assessed using standard procedures. Flowering frequency (FF) was correlated with temperature, relative humidity and rainfall. Data were analysed using descriptive statistics, correlation analysis and ANOVA at $\alpha_{0.05}$.

Flowering occurred twice a year (January-July; October-December), while fruiting occurred between February-August and November-December in both sites. Flowering duration was 104.3 ± 7.6 , 48.7 ± 10.6 and 93.8 ± 12.3 , 62.3 ± 4.4 at FRIN and NIHORT, respectively. Fruiting duration was similar in both sites (70-131 and 18-38). Number of flowers/inflorescence varied from 294.2 ± 40.3 to 296.6 ± 37.3 . The highest Z (FRIN: 0.67 ± 0.1 , NIHORT: 0.88 ± 0.1) occurred in the first flowering cycle and least in second (FRIN: 0.34 ± 0.27 , NIHORT: 0.43 ± 0.29). Eight and 13 insect species were encountered at FRIN and NIHORT, respectively. *Monomorium minimum* was most frequent (FRIN: 27.9%; NIHORT: 22.9%). *Bombus* sp. had highest pollen load (25%),

while fruiting efficiency ranged from 0.25-0.99% in both sites. Fruit- length ($16.7\pm1.48-20.9\pm2.5$), width ($37.2\pm5.1-52.4\pm5.9$) and NS ($10.8\pm3.9-15.6\pm2.4$) were significantly different. Aponmu had highest seed weight (16.2 ± 0.1), while Mamu had least (10.9 ± 0.3). Seedling height ($53\pm17.8-61.8\pm12.6$) and number of leaves ($14.1\pm2.9-16.3\pm2.9$) were significantly different. Cuttings survival (56.3%), NR (2.19 ± 2.3) and RL (2.8 ± 2.6) were highest at base and least at top (1.8%, 0.07 ± 0.9 and 0.04 ± 0.3). Mamu had highest survival (38.2%), NR (1.5 ± 2.3), RL (1.56 ± 2.2) while Ibadan had least (12.4%, 0.4 ± 1.2 , 0.6 ± 1.9 , respectively). Flowering frequency was positively correlated with temperature (r=0.53 (FRIN); 0.52 (NIHORT), but negatively correlated with relative humidity [r=-0.60 (FRIN); -0.22 (NIHORT)].

Tetrapleura tetraptera exhibited extended and synchronised phenological patterns. Fruit and seed source affected early growth and development. Stem cuttings from the base had better rooting ability and are most suitable for macro-propagation.

Keywords: Flowering pattern, Floral synchrony, Tree domestication, *Tetrapleura tetraptera* **Word count**: 485

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CHAPTER ONE

INTRODUCTION

1.1 Background of the study

The tropical forest is a veritable renewable resource that has a dominant role in the economic well-being of most rural dwellers as well as many urban people (Olajide and Udofia, 2008; Awodoyin et al., 2015). It contains high biodiversity particularly of trees which provide timber and non-wood forest products (NWFPs) such as fruits, nuts and vegetable, oils, gums, fibres, resins, dyes and herbs (Aigbe and Oluku, 2012). Apart from the economic benefits of timber products which are highly valued at national and international levels, non-timber forest products from indigenous tree species like Tetrapleura tetraptera (Schum. and Thonn.) Taub. plays significant role in contributing to rural income, food security and health care (Endamana, 2016). Hence, indigenous forest resources are vital for the social well-being, economic, and healthcare of low income people (Jimoh et al., 2013). Unfortunately, due to over exploitation, deforestation, urbanisation and seasonal bush fires, the biological diversity of the Nigeria's tropical forest where most of these resources are domiciled is constantly being diminished (Ladipo et al., 1997; Ouinsavi and Sokpon, 2010; Suleiman et al., 2017). This situation is affecting the structure and function of forests causing substantial loss of plants germplasm and posing threats to conservation, decreasing survival rates, growth and reproduction of individual trees as well as causing the degradation of major gene pools (Oumorou et al., 2010).

Tetrapleura tetraptera is an indigenous tropical tree species whose leaves, bark, roots and fruits are utilized for food, medicine and aromatic purposes in Africa (Adesina *et al.*, 2016). The species contains potent constituents that are useful in biological or pharmacological processes. The plant components have been proposed as active ingredients in drug formulation (Adesina *et al.*, 2016). These components have been

found to be molluscidal, anti-ulcerative, anti-inflammatory and anti-microbial (Abii and Elegalam, 2007). For instance, it is locally utilized for control and management of *Diabetes mellitus* (type 2).

In Eastern Nigeria, the pods have been shown to have positive lactation properties and have been used to prepare meals for breastfeeding mothers on the first day after birth to avoid postpartum contraction (Enwere, 1998). Studies conducted by many researchers have established the efficacy and potential of *T. tetraptera* plants in the control of other ailments like kidney failure and cardio-vascular illnesses (Ajayi *et al.*, 2011), schistosomiasis (Aladesanmi, 2007), bacterial (Ekwenye and Okorie, 2010) and microbial (Okoronkwo and Echeme, 2012) infections. Due to the threats imposed on this economically and medicinally important tree species by unsustainable anthropogenic activities, deliberate research work on the species phenology, germplasm variation and development of appropriate domestication protocols became necessary with a view to establishing a sustainable management action plan for the conservation of remaining germplasm.

1.2 Statement of Problem

The dearth of information on the phenology and maturation of tropical fruits and seeds is a major challenge facing efforts aimed at domestication and conservation of the species (Ladipo *et al.*, 1992). Tree species differ in their phenology with respect to year and season; little is known about the pattern of biological events in *T. tetraptera*. Infact, majority of researches on reproductive phenology of tropical trees reported initiation and duration of blooming at the population level (Engel and Martins, 2005; Ettinger *et al.*, 2018) or community flowering patterns (Medeiros *et al.*, 2007). Environmental and genetic effects of individual trees on flowering phenology depend on the behaviour of the individual species on a spatio-temporal basis (Grodgan and Loveless, 2013). However, the numerous medicinal and nutritional benefits of *T. tetraptera* have encouraged research mostly on silvicultural requirements suitable for its successful establishment while aspects of reproductive phenology necessary for continued existence have not been fully explored. Hence, there is a knowledge gap in this aspect of the species. Climatic variables such as moisture, temperature, irradiance, relative humidity, day length and photoperiod may be associated with flowering and fruiting trends of many tropical plants (Mohandass *et al.*, 2018). There is inadequate information on the climatic factors that initiate flowering and fruiting phenology in *T. tetraptera*.

Information on floral traits and ratio of pollen grains to ovules determines the breeding system of plant species (Cruden, 2000). Floral morphology and arrangements largely determine the floral visitors (Faegri and Van Der Pijl, 1979). Unfortunately, information on floral morphology and the insect visitors to *T. tetraptera* is scanty. Nevertheless, information on this aspect of reproductive biology would aid the understanding of the evolution of pollination ecology of the species (Etcheverry *et al.*, 2008). The Low fruiting efficiency reported for this species in the south-south geographical zone of Nigeria (Omokhua and Ukoimah, 2008) necessitates the need for similar studies in southwestern locations to cover the endemic range of the species in Nigeria. This would help corroborate or debunck general assumptions about the species reproductive output.

The morphological variation suspected to exist within the geographical distribution of *T. tetraptera* is not well established. Propagules of tropical tree species like *Juniperus procera* are known for their genetic variability due to their outcrossing nature (Mamo *et al.*, 2006). Thus, genetic differentiation results from extensive variation in atmospheric conditions within the geographical range of the species. However, limited scientific efforts have been channelled towards understanding these variations in *T. tetraptera* and how they influence the tree species reproductive cycle.

The scarcity of information on sources of superior planting stocks has been recognized as a major constraint of agroforestry establishment in tropical areas (Simons, 1996). *Tetrapleura tetraptera* is a leguminous species that can be planted with arable crops in case of alley cropping and/or farming. However, the potential for its inclusion in agroforestry system will further be determined by the productivity of green biomass on a regular basis through seedling growth evaluation. Surprisingly, little is known about its early growth characteristics particularly for different seed sources across its geographical range. Most *T. tetraptera* stands are found in the natural forest as a component of agroforestry farms (Omokhua and Ukoimah, 2008), and are becoming rare (Olajide and Udofia, 2008; Omokhua and Aigbe, 2014; Okereke *et al.*, 2014).

Information on protocols for macropropagation of *T. tetraptera* through stem cuttings is scanty in literature unlike *Nauclea diderrichii, Pentaclethra macrophylla* and *Triplochiton scleroxylon*. The uncontrolled exploitation of fruits and/or seeds of *T. tetraptera* for spice and herbal condiments limit the amount of germplasm for regeneration. Also, the seeds are dormant (Opabode *et al.*, 2011), with low germination percentage under natural conditions (Jimoh, 2005), this requiring pre-sowing treatments in order to break their dormancy before sowing. In addition, seeds of the species are prone to pre-germination mortality as a result of herbivory, diseases and unfavourable environmental conditions, thus, limiting regeneration of the species in the wild. Hence, there is need to develop alternative methods of mass propagating the plant for sustainable utilization of the species.

The use of auxin has been largely reported to enhance rooting ability in some tropical trees (Kebede *et al.*, 2013). However, information is lacking regarding auxin roles on rootability of *T. tetraptera* stem cuttings. High cost of growth regulators makes the procurement sometimes difficult for farmers, thus natural growth regulators have been suggested as alternatives. They have been shown to also promote rooting of plant cuttings (Usman and Akinyele, 2015). Nevertheless, information on the potentials of natural growth regulators in increasing rooting of *T. tetraptera* stem cuttings is poorly understood.

The influence of stockplant source as a factor in achieving rooting success of *T. tetraptera* is also scarce in literature. The inability of stem cuttings to root uniformly and high mortality rate of planting stock of this species suggest that some interacting factors may be inhibiting rooting success. However, nodal/cutting positions on the stem and/or shoot have been noted to influence rooting ability in tropical trees (Adeyanju *et al.*, 2013; Saifuddin *et al.*, 2013). It is pertinent to provide empirical evidence in identifying the limiting factors to its successful asexual propagation.

1.3 Objectives of the study

1.3.1 Main objective

This study assessed the flowering and fruiting phenology, germplasm/early growth variation and macro-propagation of *T. tetraptera* with a view to providing relevant data for successful conservation and improvement of the species.

1.3.2 Specific objectives

- **i.** To assess the flowering, anthesis and fruiting phenology of *T. tetraptera* and determine the influence of climatic variables (rainfall, temperature and relative humidity) on them,
- **ii.** To evaluate the floral morphology, insect visitors dynamics and fruiting efficiency of *T. tetraptera*,
- iii. To determine the variation in morphological traits of fruits of *T. tetraptera* from four different sources in south-west Nigeria,
- **iv.** To assess the effect of seed sources on seedling variations, early seedling growth and biomass accumulation of *T. tetraptera* seedlings,
- v. To develop protocols for macro propagation of *T. tetraptera* using stem cuttings.

1.4 Justification

Onset and length of flowering and fruiting events is a key trait of reproductive output and inherent variation in progenies of plant species (Elzinga *et al.*, 2007; Wei, 2016). It shapes mating possibilities between synchronous plants, thereby influencing the transmission of genetic diversity from generation to generation and affecting its degree and structure in populations (Ison *et al.*, 2014). The schedule of phenological events of *T. tetraptera* may indicate the influence of biotic and/or abiotic environmental conditions (Mohandass *et al.*, 2018). For instance, temperature and rainfall have been shown to influence pollinator visit, pollen transfer and invariably fruit production in most trees (Bustamante and Búrquez, 2008). However, flowering plants are found to respond differently to seasonal variation in rainfall and temperature (Singh and Kushwaha, 2005). Researches into phenological events of some tropical tree species have provided an understanding on patterns of plant physiology including the significance of abiotic conditions and biotic factors as selective cues for phenological events (Khanduri, 2014). Therefore, the

determination of the flowering and fruiting patterns of *T. tetraptera* are essential components in understanding its reproductive ecology. Such knowledge is also vital for its successful domestication, improvement and conservation of indigenous plant genetic resources (Omondi *et al.*, 2016). They help in the understanding of the forest ecosystem functioning and dynamics as well as the regeneration success of plant species (McLaren and McDonald, 2005). Information on schedule of anthesis and fruiting events of plant species is one of the indicators that have been used and accepted by many Researchers to monitor climate change (Rötzer and Chmielewski, 2001). This is because change in patterns of phenological event may indicate changing climatic variability. Hence, understanding phenological sequence of tropical trees and their interacting environmental factors is crucial to assessing the health of a forest ecosystem.

An understanding of floral trait of Tetrapleura tetraptera is necessary to determine the possible pollinators and study the species behaviour (Barret and Harder, 1996), and to clarify evolutionary relationships among angiosperms (Oyelana and Ogunwenmo, 2012). The characteristics of a flower are crucial to enhancing the pollen transfer from anthers to the stigma because of their role in pollinator attraction (Ohara and Higashi, 1994). The inability of floral organ to attract efficient pollinators may result in pollen restriction thus, limiting seed formation (Ashman et al., 2004). This can also limit reproductive output in certain cases (Trueman, 2013). Pollination success of most tropical angiosperms is controlled by the behaviour of insect visitors and floral characteristics (Sornsathapornkul and Owens, 1998). The sexual systems (monoecious and dioecious), floral pollination syndromes and characteristics can influence the ecosystem (forest) changes and regeneration processes (Machado et al., 2006). Derived knowledge from resource allocation between the pollen and ovules of a flower is a critical trait to comprehending the breeding sytem of T. tetraptera (Cruden, 2000). Pollen-ovule ratio also reflects the efficiency of pollen transfer in a flowering plant. This determines the chances of pollen that will fertilize every ovules in an ovary to ensure maximum seed set. Therefore, transfer efficiency of pollen grains is increased in species with clumped pollen grains because of their lower polyad-ovule ratio (Cruden, 1997). Hence, analysis of pollen-ovule ratio of *T. tetraptera* will provide a better understanding of its reproductive system.

Pollinating insects have a pertinent role in transmission of genetic material within and between populations of rainforest trees due to their ability in promoting species diversity and evolutionary dynamics over time (Oyelana and Ogunwemo, 2012). Lack of plant-pollinator association often causes species extinction and population dispersal or fragmentization. Knowledge of insect visitors and their interaction with *T. tetraptera* trees will aid identification of potential pollinators and understanding of reproductive success. Adequate information on the foraging pattern and behaviour of insect visitors of *T. tetraptera* trees will enhance better understanding of its pollinating syndrome, breeding methods and population structure of insect species (Mitchell *et al.*, 2004).

Knowledge on fruiting efficiency of *T. tetraptera* trees (which include fruit set and gynoecium drop) would provide vital information on species improvement, domestication and conservation. Synchronized flowering, the availability of compatible pollen and its efficient transfer and deposition on stigma affect fruit and seed set in plants (Baranelli *et al.*, 1995).

Information on morphological variation of *T. tetraptera* fruits from different sources across its geographic range would be beneficial to its genetic improvement programmes (Singh *et al.*, 2010). Infact, experiments on provenance evaluations are the preliminary steps for any tree species improvement (Mamo *et al.*, 2006). Such studies are very important in identifying the outstanding seed source that can also withstand adverse environmental conditions. This would enhance the choice of *T. tetraptera* seed sources and standardize the procedures for germplasm collection for plantation establishment. It would also improve productivity if such species is introduced into agroforestry systems (Takuathung *et al.*, 2012). Evaluation of morphological traits of fruits and seeds of *T. tetraptera* from different sources are critical steps to enhance tree species productivity because improvement in a morphological trait would promote improvement in the corresponding character (Abraham *et al.*, 2006; Sudrajat, 2016). Consequently, this would enhance the development of superior individuals and reduce exploitation pressure on wild populations with the establishment of plantations of superior genotypes in a fruit orchard.

The physiological characteristics of plant species with wide geographic range are usually affected by differences within and between climatic zones (Assobadjo et al., 2010). Evaluation of the pattern of genetic variation within the natural range of a T. tetraptera would also be imperative to guide selection of outstanding phenotypes (Unival et al., 2002). Growth traits variations ensure biological stability and enable species to withstand climatic conditions. Infact, successful tree breeding programme start with screening of seed sources for provision of outstanding (superior) trees (Franzel et al., 2008). Knowledge of early growth rates of T. tetraptera seedlings are equally important information to be considered in seedling establishment for inclusion in agroforestry systems such as alley or hedge row cropping. However, considering the long gestation periods of forest tree species, field evaluation of different seedlots is capital and labour intensive and the merits may not justify the investment. Hence, Ladipo et al. (1992) pointed out that screening at nursery stage offers an excellent opportunity for the assessment of genetic differences. Therefore, knowledge on genetic variability between and within populations of T. tetraptera trees will provide useful guidelines for seed sourcing and genetic improvement.

The amenability of stem cuttings to rooting in some tropical trees species has been reported (Atangana *et al.*, 2006; Akinyele, 2010). Asexual propagation of *T. tetraptera* using stem cuttings would ensure the transfer of desirable characters from ortet to progenies (Hartmann *et al.*, 2002) and also facilitates the mass production of quality plant materials (Leakey and Simons, 2000). However, the uses of growth regulators also affect the rate of propagation success in leafy stem cuttings (Leakey, 2004). Accessibility and cost of procuring growth regulators is therefore a major barrier for low income growers. Recent research efforts have shown that alternate growth regulators like coconut water tend to eliminate this constraint (Usman and Akinyele, 2015; Dunsin *et al.*, 2016). Hence, an assessment of the potentials of auxins and alternate growth regulators for propagation of *T. tetraptera* using stem cuttings was timely.

Provenance variations in stockplants and nodal positions of cuttings have been suggested to influence rooting success of some tropical trees (Tchoundjeu and Leakey, 2001). Stem cuttings obtained from various positions on the seedling shoots vary in their rooting ability. This difference in rooting response of cuttings has been attributed to within and between shoot factors which affect physiological processes (Leakey, 2014). Therefore, evaluation of influence of stockplant's source and cutting positions on the shoot of *T. tetraptera* seedlings would be essential for mass propagation of cuttings, maximising propapagation cost and profitability.

1.5 Scope of the study

The entire research study was undergone in the south-western part of Nigeria. The phenological assessment of T. tetraptera trees was carried out at Forestry Research Institute of Nigeria (FRIN) and National Horticultural Research Institute (NIHORT), Ibadan, Oyo State, Nigeria. Similarly, timing of flowering and fruiting, duration, anthesis and intensity and fruit growth were monitored in the study areas. The floral morphology was analysed at the Plant Anatomy Laboratory of Department of Botany, University of Ibadan, Nigeria. Pollen morphology and quantification were carried out at the Palynology Laboratory, Department of Archaeology, University of Ibadan, Nigeria. Morphological traits of fruits and seedlings obtained from the four seed sources in South-West Nigeria (Ibadan (Oyo State), Iwo (Osun State), Mamu (Oyo state) and Aponmu (Ondo State) were examined at the Department of Forest Production and Products, University of Ibadan. The early growth and biomass accumulation in seedlings from the different sources were evaluated. The multiplication of T. tetraptera propagules through vegetative propagation was done using leafy stem cuttings with exogenous application of growth regulators at varying concentrations to facilitate rooting. The comparative effect of alternate growth regulator (coconut water) and auxins (IBA and NAA) on the rooting of stem cuttings were assessed. Finally, the effect of stockplant source and nodal position of stem cuttings on the shoot were also investigated.

CHAPTER TWO

LITERATURE REVIEW

2.1 Description and botanical features of *Tetrapleura tetraptera*

Tetrapleura tetraptera (Schum. and Thonn.) Taubert belongs to Fabaceae plant family and the sub-family is Mimosoideae (Keay, 1989). The species has different local names in Nigeria: aridan (Yoruba), oshosho (Igbo) and edeminyang (Efik) (Keay, 1989). *Tetrapleura tetraptera* trees are distributed in the tropical forest of Africa especially in the rainforest. It is deciduous and the height of a matured tree ranged from 20 to 25m while the girth varies from 1 to 3 m (Orwa *et al.*, 2009). Adult trees have slender bole with small and sharp buttresses (Plate 2.1). The tree has glabrous compound leaves that are arranged on the stalk. The length of the pinnae varies from 5 to 10 cm and they are arranged in pairs (5 to 9), usually opposite or alternate. Leaflets are sessile and alternately arranged on each side of the pinna stalk and their number ranged between 6 and 12. The length of the leaflet varies from 12 to 25 mm while the breadth is between 6 to 12 mm. The leaflets are rounded at the two ends, elongated and elliptic (Orwa *et al.*, 2009).

Tetrapleura teraptera floral colours are pinkish-cream turning to orange during anthesis or sometimes creamy yellow. The flowers are clustered in spikelike inflorescences (length; 5-20 cm), arranged in pairs on the twigs. Each flower has a thin stalk of 10 stamens with the anthers bearing secretions at the pinnacle (Orwa *et al.*, 2009). *Tetrapleura tetraptera* fruit hang at the end of the branch on stout stalks of 25 cm long and is persistent. The fruit (length; 15 - 25 cm, width; about 5 cm) is shiny, glabrous, dark brown and slightly bent with four vertical-winglike ridges about 3 cm broad. The genus name (*Tetrapleura*) originates from a Greek word implying 4 ribs fruit while specific epithet (*tetraptera*) refers to 4 wings. Two of the fruit wings had soft, sugary pulp that is oily and aromatic, while the other two are woody. The seeds are orthodox, small, black, hard, and flat. The length of seed is about 8 mm and it is embedded in the mesocarp.



Plate 2.1. A matured *Tetrapleura tetraptera* **tree** Source. Field survey (2017)

There are about 6 290 seeds per kilogram. The timber is Reddish to brown, fairly hard heartwood and white sapwood. *Tetrapleura tetraptera* shed leaves in December while flowering is initiated at late February and lasted till early April in Ghana. Fruit fall occurs from September and is over in December (Orwa *et al.*, 2009).

The unique pleasant smell of the fruits attracts seed dispersals in the wild. The fruits are also edible and have been indicated to contribute to nutritional needs and well-being of indigenous people as they contain certain compounds like crude protein (7.4 - 17.5 percent), lipid (5 - 20 percent), fiber (17 - 20 percent), carbohydrates (43 - 49 percent) and 234 to 380 g/calory of food energy (Okwu, 2003). In Southern Nigeria, the unique aroma of the dry fruit explains its utilization as a spice condiment (Essien *et al.*, 2009). The presence of major minerals such as Calcium (Ca), phosphorus (P), potassium (K), zinc (Zn) and iron (Fe) was also demonstrated by the utilization of the fruits as a condiment and aromatic agent was the Crude lipid fraction in the fruits is higher than what is normally found in common spice like bonnet pepper, onion, curry and ginger, therefore enhancing its processing and improving its use for commercial purposes (Essien *et al.*, 1994).

The leaves, barks, roots as well as kernels of *T. tetraptera* fruits are used for medicinal purposes. The fruit extracts are used as recipes of various medicinal treatments for inflammations, abdominal aches, convulsions, flatulence and indigestion in Western Africa (Adesina *et al.*, 2016). Infusion of whole fruit can be used as an enema, emetic or to treat for feverish conditions and constipation (Adesina *et al.*, 1980). It is also an active ingredient in drug preparations to manage pains, arthritis, high blood pressure, type 2 diabetes, epileptic conditions, high bilirubin level, schistosomiasis, asthma, feverish conditions and microbial infections (Burkill, 1985). In Nigeria, it is used locally to make body cosmetics to treat fever, ulcers and skin rashes (Adelaja and Fasidi, 2012).

The species domestication has also been reported, seedlings can be raised successfully with organic and inorganic fertilizers (Offiong *et al.*, 2010). According to these Authors, application rates and fertilizer type influenced early growth and development of the

species. The seeds of T. tetraptera are dormant and therefore require pre-germination treatment to break the dormancy in order to achieve optimum germination (Jimoh et al., 2005). The most efficient means of breaking T. tetraptera seed dormancy was to soak seeds in concentrated H_2SO_4 acid (Ibiang *et al.*, 2012). This was also consistent with Alaba et al. (2006) and Wakawa and Akinyele (2016) that soaking T. tetraptera seeds in concentrated H₂SO₄ acid recorded the highest percentage germination compared to other pre germination treatment such as mechanical scarification and heat treatment. Germination experiments examining exogenous application of auxins on T. tetraptera seeds revealed that Napthalene Acetic Acid (NAA), Indole-Butyric Acid (IBA) and Indole Acetic Acid (IAA) promoted seedling sprout and regeneration at low concentration of 0.005 g/ml (Maku et al., 2014). Loamy-sand had been shown to be the most appropriate soil textural class for *T. tetraptera* establishment (Akpan and William, 2016). However, the species has the potentials to be established in different ecological zones in Nigeria (Wakawa and Akinyele, 2016). Studies on the fruiting pattern of T. tetraptera in the Ekpoma as well as Benin region of Edo State in Nigeria have shown low fruiting efficiency in the species (Omokhua and Okoimah, 2008).

2.2 Flowering and fruiting phenology of tropical trees

2.2.1 Timing of flowering and fruiting

Reproductive phenological schedule in indigenous tropical trees is a very important character of plants because it determines the number of mating opportunities available to a gynoecium and consequently, effective reproduction of cross-pollinated species (Dominguez and Dirzo, 1995). Timing of blooming has been studied at the community level (Engel and Martins, 2005; Ettinger *et al.*, 2018) and at the individual level (Grodgan and Loveless, 2013; Sinebou *et al.*, 2016). Flowering and fruiting pattern of plants in a population are described by periodicity, length, frequency or intensity of flowering (Bawa *et al.*, 2003). Flowering phenology may result from physiological response to a proximate environmental cue such as photoperiods (Glover, 2007), temperature, irradiance (van Schaik *et al.*, 1993), rainfall (Lobo *et al.*, 2003) as well as ultimate factors (availability of pollinators and seed dispersals) that select for a particular reproductive phenology. The timing of biological events (flowering/fruiting) in plants has significant effect on survival

or reproduction success (Elzinga et al., 2007). This indicates the period when floral organs associate with plant resources for fruit and seed development (Rodriguez-Perez and Traveset, 2016). For instance, flowering events in tropical trees have been associated to period when pollinators are abundant while fruiting events is associated to period of maximum water availability that may favour seed germination and seedling establishment or the time when seed dispersals are most abundant (Funch et al., 2002). Reproductive phenological events of tropical trees vary within and among populations in addition to seasonal variation (Williams-Linera, 2003; Wei, 2016). For instance, flowering events in the seasonally tropical forests occurs during the change from late dry to early rains (Murali and Sukumar, 1994). However, flowering activities in tropical trees such as Tamarindus indica was initiated at the start and end of the dry period whereas fruit production was a wet season event (Fandohan et al., 2015). Hence, the schedule of these phenological events was synchronized with relative humidity and rainfall (Okullo et al., 2004). It is expected that tropical trees whose flowering occured in the dry season often develop fruit in the rainy period (Singh and Kushwaha, 2006). Temporal change in water status of soil for some plant species has been identified as the proximate and ultimate index triggering phenological events in seasonal forest (Borchert, 1994). This was consistent with De Bie et al. (1998) who indicated that seasonal variation in flowering and fruiting patterns of tropical trees was associated with water availability in the soil and temperature change. Peak flowering events of most trees in the late dry season also confirmed that temperature and rainfall were important climatic factors controlling flowering in the tropical rainforest of Southwestern China (Mohandass et al., 2018). Seasonal variations in irradiance have also been noted as proximate environmental cue that signal flowering near the peak of dry season in some tropical trees (Adler and Kielpinski, 2000) apart from temperature (Nadarajan and Pujari, 2018). Seasonal changes in resource availability or peaks of irradiance greatly determined timing of flowering in wet tropical forest (Wright and Van Schaik, 1994). Variations in day-length is another essential factor that is associated with reproductive phenology in both seasonal and aseasonal tropical climates (Morellato et al., 2000). Exposure of some tropical tree species to either short or long photoperiod has been correlated with floral buds initiation (Glover, 2007). On the other hand, flowering phenology may also be indirectly influenced by leaf phenology because both leaf growth and reproductive events involve nutrient and energy utilization (Rivera et al., 2002). Therefore, resource availability and partitioning among vegetative and reproductive organs may affect staggering flowering period. Both Flowering time and leaf shedding period determine the duration of fruiting activity in tropical deciduous trees during the annual cycle. The ability of some tropical trees to initiate floral buds during leaf fall in the dry season is a unique adaptation to withstand seasonal climate (Bochert et al., 2004). According to these Authors, leaf shedding phenomenon enables tropical trees to minimize water stress during the dry season flowering therefore, resulting in temporal partitioning of resources for vegetative growth and flowering events. Flowering and fruiting processes of plants are affected not only by biotic and abiotic variables, but also the genetic factors inherent in plants (Sundarapandian et al., 2005). Flowering activities in many angiosperms was under strong genetic influence (Weis and Kohler, 2004). Flowering in some tropical tree species can occur at any period during the year although coexisting trees differ in timing (Sakai, 2001). Variations in time of flowering in some plant species have also evolved to prevent plantpollinator competition and inter-specific pollen transfer with other genetically associated species (Anderson and Hill, 2002).

2.2.2 Duration and frequency of flowering in tropical trees

The duration or length of blooming period is an important parameter in assessing flowering pattern of a species. It can be described for individual flower, flower head or cluster, tree, community and species (Primack, 1985). Tropical trees within a population vary widely in their flowering length ranging from a few days to several months (Opler *et al.*, 1980). Flowering duration may be long in some tree species, whereas in others, it may be brief. Flowering duration can be described in terms of seasonal and extended flowering pattern. For instance, seasonal flowering pattern connotes brief flowering that is associated with a particular season (dry or wet) while extended pattern ranged from two weeks to five months or more (Frankie *et al.*, 1974). Plant reproduction may be guaranteed in extended blooming by increasing pollination opportunities, manage plant investments in reproductive organs, and/or avoid competition for seed dispersals. The

length of flowering in an angiosperm regulates pollen flow or feeding behaviour of a pollinator, thus affecting the pollination success of such plant (Bawa *et al.*, 2003).

For instance, pollinator movement between plants and invariably pollen transfer is controlled by the rate of daily production of floral resources and this is a means to avoid selfing in flowering plants (Bawa, 1983). Besides genetic control, duration of flowering is also influenced by variation in environmental conditions of plant species. For example, plant species could bloom for extended periods in temperate areas because of similar climatic conditions therefore reflecting no distinct selection for flowering phenology (Marquis, 1988). The flowering duration of *Vitellaria paradoxa* in the Sudanian zone of Cameroon, was five months (February to June) although flowering peak occur in March, whereas in the Guinean zone, it was four months (November to December) with peak flowering in December (Nguemo *et al.*, 2014). Flowering duration of *Senegalia senegal* was also extended during the long rainy events than the brief rainy periods in Kenya (Omondi *et al.*, 2016).

Flowering frequency varies with different species and it ranges from trees that flower once a year (annual), to bi-annual (sub-annual), some are less frequently than once a year (supra-annual) and others are more or less continually in 12 months (continual). The distribution of tree species exhibiting each of these patterns also varies with forest types. Annual flowering tree species are more frequent than sub-annual and continual (Sundarapandian *et al.*, 2005), while supra-annual species are the least frequent in the tropical forest (Bawa *et al.*, 2003). Annual flowering species are highly distributed in forest with stronger seasonality (Sakai, 2001; Mohandass *et al.*, 2018). However, sub-annual species are the most frequent than annual flowering species in the rainforest of Costa-Rica (Newstrom *et al.*, 1994). Sub-annual tree species may be influenced by time of fruit maturity and maximum time for seed dispersal. For instance, repeated flowering events in a tree species may not be possible if maturity of seeds and fruits of such trees span over a long time. Selection for annual species (such as lianas, herbs and shrubs) appeared to have evolved because ovules are dispersed by wind in the summer (Newstrom *et al.*, 1994). Studies have also shown that phenological patterns in species such as *Guiera*

senegalensis are not influenced by changes in abiotic factors whereas in other species like *Combretum aculeatum* and *Acacia adansonii* fluctuate with years (Grouzis, 1991).

2.2.3 Flowering intensity of tropical trees

Flowering peak in tropical trees are distinguished and ranges from major to minor in seasonal environments. This variation in flowering intensity is often related to climatic variation (Bawa *et al.*, 2003). For instance, flowering peak of most trees in tropical environments occurred in the transition period from late dry to early rainy periods (Mohandass *et al.*, 2018). Such flowering activity may be an indication that flowering patterns is associated with variations in climatic conditions in the tropics or seasonal environments. For example, peak of flowering in *Acacia mangium* occurred from September to December while in *Acacia auriculiformis*, November to February in (Ngiem *et al.*, 2011). Peak flowering of most tropical trees often coincides with period of optimum resource availability and temporal abundance of pollinators while peak fruiting coincides with period of abundant seed dispersals and soil moisture that may influence seed germination and seedling establishment (Funch *et al.*, 2002).

2.2.4 Floral synchrony and anthesis of flowering plants

Floral synchrony refers to simultaneous opening of flowers and can be described in terms of pattern of flowering in trees with respect to other trees in the population. This is an important factor for plant fitness especially self-incompactible species or out-crossing trees. Anthesis (flower opening) is described as the period when flower parts became active and this coincide with floral bud break. Anthesis may be synchronous or asynchronous within an inflorescence. Synchronized flowering patterns of some tropical trees in a population may indicate a natural selection for cross-pollination (Auspurger, 1980). For instance, high flower synchronization exhibits a large floral display that facilitates increased flower visitation rates by insects, increased rate of pollen transfer, higher chances to find mates and potential for cross pollination thus, leading to increased reproductive success (Kudo and Harder, 2005). However, plant species varies widely with temporal pattern of floral display. Sornsathapornkul and Owens (1998) reported synchronous opening of flower buds in the *Acacia* hybrids inflorescence. Asynchronous

flowering was also exhibited on inflorescence of some tropical plants including *Vitex doniana* (Sinebou *et al.*, 2016) and *Solanum* species (Oyelana and Ogunwenmo, 2012). Similar flowering pattern was reported in *Pentadesma butyracea* with the apical or terminal flowers blooming first while flowering lasted 3 to 5 days on an inflorescence (Ewedje *et al.*, 2015). This flowering phenomenon is beneficial in extended flowering species because unpollinated flowers might have chance of being pollinated after anthesis occurred the following day (Sinebou *et al.*, 2016).

2.2.5 Flower/inflorescence morphology and insect visitors of tropical trees

Flower morphology is a critical trait for perceiving the reproductive biology of plant species. These are essential characters that determine the movement of pollen grains from the androeciuem to the gynoecium (Ohara and Higashi, 1994). Usually, flowers are classified according to their receptiveness and form of the floral resources, size, symmetry, and colour. Pollinating syndromes can be described with these characteristics which may give hint on possible pollen vectors of plants, although not in all plant species (Ollerton, *et al.*, 2009). These syndromes are also necessary tool for assessment of the ecosystem diversity functions (Machado *et al.*, 2006).

Floral asymmetry determines the types of pollinating agent of a plant. For instance, the measure and shape of sepals and petals of *Vigna caracalla* are proportional to the size of their pollinators (Etcheverry *et al.*, 2008). This also agreed with the asumption that floral traits (pollination syndromes) may restrict assess to less functional groups of pollinating agents (Rosas Guerrero *et al.*, 2014). For example, large and bulbous corolla would permit greater number of pollinators, within-plant interaction and transfer of genes (Mitchell *et al.*, 2004). This floral trait in *Butea monosperma* was also attributed to ornitophilly; the bird pollinating syndrome (Tandon *et al.*, 2003). However, presence of long and slightly curved beak in sunbirds seems to enable them to recruit nectareous glands from the base of flowers in a number of tree species. In *Acacias*, accessibility of nectars is also restricted to insects with long proboscis capable of penetrating the corolla tube (Bernhardt and Walker, 1984). Sweet-strong scent, brightly coloured petals and abundant nectar have been regarded as floral characteristics of bee pollinating syndrome (mellitophilly)

according to Borges *et al.* (2009). Plant species with small-sized corolla and small nectar secretions have also restricted visits to small insect species such as ants because of their low energy requirement and excluded larger pollinators with greater nutritional requirement.

Number of flowers and their arrangement on inflorescence is another variable that can be used to elucidate the plant-pollinator interaction. Inflorescence refers to cluster of flowers on a stem (floral shoot). Floral arrangement on inflorescence can be determinate (cymose) or indeterminate where the youngest buds are located at the apical axis (such as raceme, globose, panicle, catkin, spike or elongate). Different species of *Acacia* has been recognized by the number and arrangement of flowers on their floral heads (Kenrick, 2003). According to this Author, spicate inflorescence was noted to contain over 500 flowers while a globose inflorescence may contain as few as three flowers. For instance, *Acacia caesia* was reported to have a globose inflorescence with an average of 32 flowers (Solomon Raju *et al.*, 2006); *Acacia drepanolobium* has few flowers per inflorescence while *Acacia nilotica* has many flowers per inflorescence (Stone *et al.*, 2003). On the other hand, *Butea monosperma* has a paniculate racemose inflorescence with an average of 167 flowers (Tandon *et al.*, 2003).

Number of stamens and pollen presentation is another floral morphology that is species specific because of the variation in their size and arrangement. Spatial pollen presentation is a floral morphological variable that is imperative for pollinating visitors of plant species especially in *Acacia sp* (Stone *et al.*, 1998). Mature pollen grains may be presented and shed as a unit (monad), two (dyad), three (triad), four (tetrad) and eight or more (polyad). The fusions of pollen grains in polyads are typical of Mimosaceae such as *Acacia sp* (Panicker and Sreedevi, 2004). The size and quantity of pollen grains in each polyad also varies with species and this may range from 4, 8, 16 or 32. Pollinators of these species collect pollens as units and their food requirement may be an index for selection of plants in addition to their size. However, the difference in the presentation of pollen is crucial for flower visitors with high energy and nutritional requirement because pollen import and export often increases when pollinators visit more flowers on massive displays (Kudo and

Harder, 2005). For instance, large insect species restricted visits to those plant species with densely flowered inflorescence (Sornsathpornkul and Owens, 1998). Such behaviour tends to save energy rather than raking pollen from inflorescences with few flowers (spatial arrangement) which appears to involve greater energy. The size or weight of an insect species could also be attributed to pollinator's preference for specific inflorescence because densely flowered inflorescence appears to support the weight of large insect species thus, make foraging easy unlike the spatial inflorescence that may collapse with large-sized species (Stone *et al.*, 2003).

Generally, floral traits with bees and diverse insects pollinating syndromes do not have specialized morphological mechanisms to restrict any flower visitors (Momose *et al.*, 1998). It is worthy to note that the quantity of seeds produced in outcrossing species will depend greatly on the activities of pollinating agent or pollen vectors. The main pollinators of most tropical trees are species from the Hymenoptera, Diptera, Coleoptera and Lepidoptera order (Bawa, 1990; Renner and Feil; 1993; Cruden, 1997). For instance, insect visitors of the Hymenoptera order (such as honey bee and sweat bee) were indicated as the most efficient pollinators of *Acacia hybrid* because of the frequent contact between the anther and stigma (Sornsathapornkul and Owens, 1998). The frequent visits of pollinators to an inflorescence have been established to maximise seed set and genetic differences among propagules (Sahli and Coner, 2007).

2.3 Morphological variation in fruits of tropical trees

Information on variability in fruit and seed characters within a species's natural geographical range is essential for tree improvement and choice of the best seed source. Leakey *et al.* (2000) established that significant variations in the morphological characters of fruit and nut traits of *Irvingia gabonensis* are important indicator for its genetic improvement. This information is critical to produce genetically superior phenotypes of plant species. Significant differences in fruit morphological traits also existed in *Pentaclethra macrophylla* (Tsobeng *et al.*, 2012) and *Adansonia digitata* (Assogbadjo *et al.*, 2010). The observed differences in morphological characters could be explained by variation in the species's genotype or the conditions where the species are grown (Shankar

and Synrem, 2012). However, the most variable morphological character in tropical fruit trees has been reported to be fruit and seed weight. These traits have also been noted as the most variable in Chrysophyllum albidum which implies that they are good indicators for tree selection and improvement (Dadegnon et al., 2015). Similar variations have also been reported among Chrysophyllum albidum provenances in Nigeria (Ladipo et al., 2000), Faidherbia albida in Kenya (Fredrick et al., 2015) and Prunus nepaulensis in India (Shankar and Synrem, 2012). This trait (weight) appeared to be controlled by environmental factors while the dimensions (width and length) are assumed to be genetically influenced (McAllister, 2005). However, variations in seed morphology characters of *Dalbergia sissoo* were attributed to resource availability such as nutrients, light and water (Singh and Bhat, 2008), during seed development which varies over season. Amadi (2014) also attributed genetic and environmental variability to differences in fruits and seeds morphological characters of Plukenetia conophora from different sources in southwest Nigeria. Significant relationship has been established between fruit morphological variables of plant species and climatic variables. Sudrajat (2016) opined that significant variations in the morphophysiological traits of Anthocephalus cadamba seedas could be explained by the genetic variations and natural conditions in their geographic location. These assumptions were also supported by Akinyele (2007) who recorded significant variations in length, diameter, volume and weight of Buchholzia coriacea seeds. Fruit character and seeds character of tropical trees are critical steps to enhance tree species productivity because improvement in a morphological trait would cause simultaneous change in the other character.

2.4 **Provenance variation in tree species**

The role of provenance evaluation in plant species is to elucidate sequence of genetic variation that will guide selection of genetically superior and well adapted phenotypes (Oyebade *et al.*, 2012). This information would enable the best productive seed sources to be identified and selected for effective conservation and safe use of genetic resources from plants (Fredrick *et al.*, 2015). Aside from the floristic richness, most indigenous fruit trees exhibit considerable intraspecific variations in tree growth, phenology of flowering and fruit characters (Dadegnon *et al.*, 2015). The genetic variation among progenies of

tropical trees could be explained by the out crossing nature of trees because progenies segregate with respect to parental traits (Simons, 1996). Significant variations have been recorded in provenance evaluation of both indigenous and exotic trees. Rochon et al. (2007) observed considerable genetic changes in early growth characteristics of *Guazuma* crinite (Mart.) due to provenance influence. Okere (2013) also reported a broad genetic diversity among the sources of Khaya grandifoliola. Similar variations in seedlings growth traits was also reported in Adansonia digitata (Assogbadjo et al., 2010), Jatropha curcas (Ghosh and Singh, 2011), Faidherbia albida (Fredrick et al., 2015) and Cordia aficana (Abraham et al., 2006). Such genetic variation offers opportunity for selection of species with high adaptation to wide range of environmental conditions. Preliminary results among provenances of Ziziphus mauritiana (Kalinganire et al., 2008) and Parkia biglobosa from broad range of West African countries also indicated considerable amount of variation in growth characteristics (Teklehaimnanot, 1997). However, variation has been described as a key to long-term species survival and continued evolution (Guries, 1982). Tree improvement harnesses the differences in the ecological range of tree species and utilizes it in the enhancement of tree performance in a desired trait.

2.5 Vegetative propagation

The production of a whole plant from a part of a mother plant, and simultaneous formation of associated cells, tissues and/or organs asexually is vegetative propagation (Mudge and Brennan, 1999). The development of vegetative propagation protocol is the second step after plus trees selection in the domestication process of plant species for species genetic improvement. The methods of vegative propagation range from macro (such as rooting of stem cuttings, budding and grafting, air layering) to micro or invitro propagation (Leakey, 2004). This technique became necessary when uniformity of genetically superior individuals is required. It is often used when seeds are not readily available or there is difficulty with seed storage, long juvenile period in plants (especially in indigenous trees), when a dioecious species is needed, and when certain individual species is of great importance (Okafor and Lamb, 1994). Vegetative propagation has been successfully carried out in a wide range of tropical trees. These include *Prunus africana*, *Syzygium guineense* (Kebede *et al.*, 2013), *Picralima nitida* (Gbadamosi, 2014) and

Massularia acuminata (Usman and Akinyele, 2015). However, some interacting factors such as stock plant physiology, propagation environment, auxins concentration, within-shoot/nodal position and leaf area affect vegetative propagation success and can only be enhanced when these factors are taken into consideration (Leakey, 2014). Khasa *et al.* (1995) also noted that higher rooting percentages in stem cuttings of *Racosperma auriculiforme* and *Racosperma mangium* were enhanced when important factors were optimised. Studies on vegetative propagation of a number of tropical timber species indicated a direct relation with propagule maturity and the lower the propagule age, the higher the rooting success and vice-versa (Teklehaimanot *et al.*, 1996; Simons and Leakey, 2004).

2.5.1 Effect of growth regulators and concentration on rooting of juvenile stem cuttings

Auxins have been extensively reported to be crucial in enhancing root formation in stem cuttings of tropical trees (Leakey, 2004). Auxins (growth regulators) are natural endogenous substances in plants that regulate growth processes. Depending on endogenous auxin concentration, exogenous application of auxin may promote, ineffective or inhibit root growth in plants. Auxins are usually measured in parts per million and applied in small quantities (Whiting et al., 2014). Rooting percentage of some stem cuttings have been reported to increase with auxin treatment (Leakey, 2004). However, tree species can vary in their response to auxin treatment and concentrations. For instance, vegetative propagation of Pentaclethra macrophylla indicated that Naphthalene Acetic Acid (NAA) enhanced rooting ability of the species compared to either Indole-3-Butyric acid (IBA) or Indole Acetic Acid (IAA) (Tsobeng et al., 2013). Atangana et al. (2006) also made similar assertion that NAA gave the highest rooting performance on stem cuttings of Allanblackia floribunda. Shekhawat and Manokari (2016) also noted that NAA significantly improve rooting in Couroupita guianensis compared to IBA and IAA. On the other hand, IBA significantly stimulated rooting in stem cuttings of Dacryodes edulis (Mialoundama et al., 2002) and Prunus Africana (Tchoundjeu et al., 2002) than NAA and IAA. Auxins concentration is another factor that affects propagation success in stem cuttings and this may either enhance or inhibit root

initiation. Mesen *et al.* (1997) reported significant roles of IBA applications on stem cuttings of *Cordia alliodora* (Ruiz & Pav.) Oken. Bud growth was inhibited in this species with increased concentrations of IBA. Gbadamosi and Oni (2005) also shared similar view that higher concentration of IBA decreased rooting percentage of *Enantia chloranta* stem cuttings. This is comparable with Gbadamosi (2014) that vegetative growth of *Picralima nitida* cuttings is inhibited by auxin concentration higher than 0.1g/L. On the contrary, rooting ability in *Aesculus indica* was significantly influenced at higher concentration (2000ppm) of IBA (Mojeed *et al.*, 2009). Similar trend was also observed in *Switennia macrophylla* where rooting performance was highest in cuttings treated with 0.4% followed by 0.2% while untreated cuttings had the lowest (Hossain *et al.*, 2004). However, juvenile stem cuttings of some tropical plants like *Massularia acuminata* rooted easily with or without growth regulator treatment (Akinyele, 2010; Usman and Akinyele, 2015).

Apart from synthetic hormones or auxins, natural growth regulator has also been reported to enhance rooting behaviour in some tropical plants. For instance, the use of coconut water has been proven to be effective as an alternate growth regulator in rooting of *Massularia acuminata* stem cuttings when compared to auxins (Usman and Akinyele, 2015). Ssimilarly, Kayode *et al.* (2016) recorded a major positive rooting effect of coconut water on *Dioscoreophyllum cumminssii* juvenile stem cuttings. Rooting rates was enhanced significantly with coconut water in Bougainvillea (Okunlola, 2016) and *Parkia biglobosa* (Dunsin *et al.*, 2016) compared to auxins. The stimulatory role of coconut water is explained by its chemical constituents including the phytohormones, antioxidants, sugar, inorganic compounds and minerals (Yong *et al.*, 2013).

2.5.2. Effect of stock plant source and nodal positions on rooting of juvenile stem cuttings

The successes of vegetative propagation in tropical trees by rooting can be enhanced if the factors (pre-severance and post-severance) controlling rooting of stem cuttings are optimized (Leakey, 2014). Adequate knowledge of these factors is essential to promote the rooting success and cost effectiveness of macro-propagation programme. Tchoundjeu and Leakey (2001) attributed variation in rooting response of Lovoa trichilioides cuttings to differences associated with stock-plant source and their physiological characteristics. Rooting success in tropical tree species differs with respect to nodal positions of stem cuttings on the stockplant (Leakey and Mohammed, 1985). For instance, rooting success in Triplochiton scleroxylon was significantly enhanced from apical cuttings than from basal (Leakey and Storeton-west, 1992). Similar observations were also found in Lovoa trichilioides (Tchounjeu and Leakey, 2001) and Azadirachta indica (Palanisamy and Kumar, 1997). These observations can be explained by variation in gradient of endogenous growth substance and cellular activity within-shoot of the stockplant (Hartmann et al., 1997). The high rooting response achieved in apical cuttings was also explained by high level of soluble carbohydrates in the flushing shoots than the basal cuttings. Similar observation was also made in *Stevia sp.* that the apical cuttings contain meristematic cells because of high concentration of auxins and cellular activities (Beemnet and Solomon, 2012). On the other hand, Tchounjeu and Leakey (2001) opined that higher rooting success in the basal nodal (cutting) positions of *Khaya ivorensis* was attributed to the presence of dormant nodes on the shoot which facilitates retention of sugars and consequent root initiation. According to Saifuddin et al. (2013), root development was enhanced in basal cutting positions than the apical positions because of the high sugar gradient and organogenic acitivity of basal nodal position. Dick et al. (1996) also attributed significant rooting variability in *Calliandra calothyrsus* to cutting location on the shoot and origin of the propagule.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Description of study locations

The entire research investigation was located in the lowland rainforest zones of Nigeria cutting across four states namely Oyo, Osun, Ogun and Ondo. The selection of these states was guided by reconnaissance survey across the species natural range. Phenological monitoring of *Tetrapleura tetraptera* was carried out at the Forestry Research Institute of Nigeria (FRIN) and National Horticultural Research Institute (NIHORT) both in Ibadan (Figure 3.1) while fruit collection for morphological variation and growth study were carried out in four locations within lowland rainforest zone of Nigeria (Figure 3.1).

3.1.1 Seedling's growth experiment

The seedling's growth and macropropagation experiments were conducted in the forest nursery of the Department of Forest Production and Products, University of Ibadan, Ibadan, Nigeria. The nursery is located on latitude 7°26'58"N and longitude 3°53'49"E with an altitude of 277m above sea level. The tropical climate has distinct dry and wet seasons. The dry season occurs between November and March while the wet season is usually between April and September. The mean minimum and maximum temperatures are 22°C and 34°C.

3.1.2 Laboratory experiment

Floral morphology was conducted at the Anatomy laboratory, Department of Botany, University of Ibadan, Nigeria while pollen morphology and quantification was executed at Palynology laboratory in the Department of Archaeology, University of Ibadan, Nigeria.

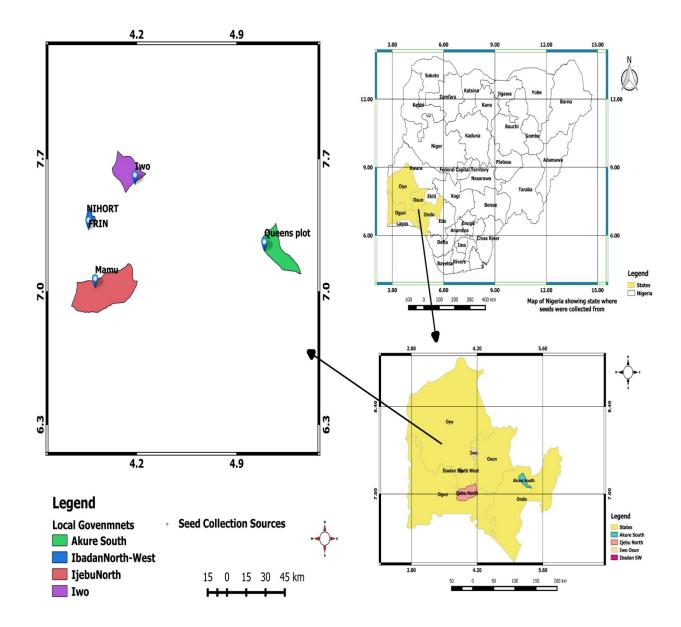


Figure 3.1. Selected locations with stands of *Tetrapleura tetraptera* in the Lowland Rainforest of Nigeria (inset: Map of Nigeria)

Source: Field survey (2017)

3.2 Phenological assessment study of *Tetrapleura tetraptera*

3.2.1 Selection of sample trees

A reconnaissance survey was carried out to ascertain fruiting population of the species in the selected locations within the rainforest region of Nigeria. Trees in the two locations (FRIN and NIHORT) were purposively selected for intensive observation of the phenological cycles based on availability and ease of access. Hence, four trees (>25 years) while six trees (>10 years) were tagged. All sampled trees had diameter greater than 30 cm at breast height (1.3m). The morphometric characters of *T. teraptera* trees were shown (Appendix 1). The tree diameter at breast height and crown diameters were determined using measuring tapes. The crown diameter was estimated as the mean of highest and lowest crown diameter of a tree. The height of the tree was calculated using a measuring tape and altimeter to determine the distance between the observer and the tree by triangulation.

One branch was selected from each of the four cardinal directions of the tree (north, west, south and east) and twenty five twigs were tagged on each branch for phenological monitoring from January, 2017 to July, 2018. Onset of flowering phenophase was closely monitored on the sampled twigs on a weekly basis. The onset of a phenophase was taken as the first day when organs on one or more branches were observed to have entered that phase while the end of a phenophase was taken as the day when no branch was observed to be carrying organs in that phase according to the methodology of Omondi *et al.* (2016). Duration (days) of a phenophase on individual tree was calculated as the difference between the onset and end of that phenophase. The number of tree species in flowering or fruiting phenophase per month was also determined as frequency of flowering or fruiting trees. Climatic data (monthly rainfall, minimum and maximum temperature and relative humidity) during the phenological assessment period were obtained from FRIN meteorological station, Ibadan.

3.2.2 Duration of anthesis per inflorescence of *Tetrapleura tetraptera*

Two trees were selected to assess duration of flower opening per inflorescence; one at FRIN and the other at NIHORT. Four main branches were chosen (one in each cardinal direction) and five inflorescences were tagged on each branch (one per twig). A total of twenty infloresences with flowers at bud stage were randomly selected and tagged. Inflorescences of tall trees were assessed by climbing using iron scaffold. The inflorescences were carefully labelled on each of the flowering trees. Four stages as described for flowering phenophase were monitored (Ewedje et al., 2015). Flowering bud initiation (fl1), well-developed bud (fl2), flower at anthesis (fl3), and flower withering (fl4). Daily floral phenology was followed between stage fl2 (well-developed bud) until the commencement of anthesis on the sampled inflorescences following the method of Sine'bou et al. (2016). Observations were made on the sampled inflorescences from 7.00 am to 1.00 pm each day to record the number of opened flowers. Due to the dense insertion of T. tetraptera flowers on inflorescence, the opened flowers were counted and carefully marked using a permanent marker at the end of each day. The exercise continued until all the flowers had opened. The duration of anthesis per inflorescence was taken as the days required for all flowers to complete anthesis on the inflorescence following the methodology of Kudo and Harder (2005). The peak of flower opening (days) on an inflorescence was taken to be the the day of highest number of opened flowers from the commencement of anthesis on the inflorescence.

3.2.3 Intensity and floral synchrony of *Tetrapleura tetraptera*

The intensity of flowering phenophase in the sampled trees in each month was determined visually with the aid of binoculars following the semi-quantitative method of Fournier (1974). Ordinal classes were used as follows according to Sine'bou *et al.* (2016):

- 0 = no open flowers observed;
- 1 = 1-25% of open flowers observed;
- 2 = 26-50% of open flowers observed;
- 3 = 51-75% of open flowers observed;

4 = 76-100% of open flowers observed;

The relative flowering intensity of *T. tetraptera* on a given monthly basis in each location was estimated by averaging the sum of individual tree's value using the method described by Ngiem *et al.* (2011).

$$F.I = \sum_{i=1}^{N} \frac{Fi}{4N} \dots 3.1$$

F I = Fournier index

 F_i = Phenophase intensity for each tree at a given time

N = Total number of observed trees

Peak flowering and fruiting were defined as the period in which the phenophase intensity of a population attained maximum. The floral synchrony defines the degree to which a plant's flowering duration (days) overlapped with the flowering of other sampled trees in the locality.

The floral synchrony index (X_i) per tree was calculated and per population (Z) following Auspurger (1983):

$$Xi = \left[\frac{1}{n-1}\right] \left[\frac{1}{fi}\right] \sum_{j \neq i}^{n} eij \dots 3.2$$
$$Z = \left[\frac{1}{n}\right] \sum_{i=1}^{n} Xi \dots 3.3$$

Where n = number of trees in the population,

 f_i = number of flowering days of the individual tree *i*,

eij = number of days on which individual tree *i* and *j* flower in a synchronized way ($j \neq i$).

The floral synchrony (X_i) ranges from 0 to 1 where 1 represents total flowering overlap indicating all trees in the population are flowering during the flowering period of the individual I while $X_i = 0$ means there is no flowering overlap among the trees in the population.

Z measures the mean flowering overlap of trees within a population.

3.2.4 Duration of fruiting phenophase per inflorescence

Twenty-five fruiting inflorescences were selected from five trees at NIHORT. This location was selected because *T. tetraptera* canopy were easily accessible for effective monitoring of the fruiting events. Fruiting phenophase was monitored forthnightly and four stages were defined; fruit set (fr1), young green fruits (fr2), fruit emerged turns brown (fr3) fruits maturation (fr4).

3.2.5 Data analysis

Initiation and completion of flowering and fruiting events of each tree were recorded to construct a phenological chart for each location. Duration of a phenophase on individual tree and inflorescence was calculated as the difference between the onset and at the end of that phenophase. Intensity of flowering was determined monthly and descriptive statistics was used to analyse duration of anthesis; fruiting per inflorescence, trees, location, floral synchrony and intensity. The number of trees (frequency) in flowering and fruiting per month was recorded. Spearman's rank correlation coefficient was used to determine relationship between climatic variables and number of trees in a phenological event. Mean values and standard deviation of morphological traits was calculated using Microsoft excel while comparisons between the means of the locations were done using student's t-test in SPSS.

3.3 Study 2: Flower and inflorescence morphology, insect visitors and reproductive efficiency of *Tetrapleura Tetraptera*

3.3.1 Flower morphology

Twenty flowers at anthesis were randomly selected from four inflorescences on two trees from the two locations and stored in 50% ethanol prior to morphological observation at the laboratory. These flowers were observed using a binocular dissecting microscope at X40 magnification to describe the arrangement of floral and sexual organs (androecium and gynoecium), and to document the number of stamens. The dimensions of floral organs (petals, sepals, anther, stigma, style and ovary) were determined using a microcaliper and light microscope at X40. The colour of the flower was noted.

For ovule count, ten matured flowers were selected and their style and ovary was separated. An ovary was placed in a glycerol drop on a slide and divided into two halves following Ngiem *et al.* (2011) under a dissecting microscope at 40X magnification. The number of ovules was counted for each ovary while images were taken using a digital camera and a photomicroscope.

3.3.2 Quantification and morphology of pollen grains

(i) Pollen grains morphology determination

One hundred anthers from ten flower buds (developed) were collected from one tree in each location (NIHORT and FRIN) and preserved in 70% ethanol. The flower buds were transferred from the vials to calibrated plastic centrifuge tubes and anthers were crushed using different glass rod for different samples. The tubes containing crushed anthers were centrifuged at 5000 rpm for ten minutes before the clear supernatant was decanted. The samples were acetolysed following Erdtman (1969). Acetolysis mixture was added to the sample and heated in a water bath, 70°C to boiling point for 10 minutes. After cooling, the sample was centrifuged and distilled water was used twice to wash off the acetolysis mixture completely. The acetolysis mixture is a very strong mixture which dissolves the cytoplasmic content of the pollen such that the exine structure becomes clearly visible. After the final washing, sample was treated with 50% glycerol. The residue was stirred and made up to 1 ml with 100% glycerol and transferred to labeled vials before mounting on pre-cleaned and labeled slides and viewed at 40X magnification under Olympus CH30 binocular microscope with vernier scale. The shapes of pollen grains were obtained by dividing the polar axis (length) with the equatorial diameter. The sizes of the pollen grains were classified according to Erdtman (1969).

(ii) Pollen count

Ten (10) μ l of the pollen was pipetted onto the slide using a micro-pipette after mixing each sample thoroughly. This was replicated on three different slides. Pollen grains were counted with a modified dilution method (Dafni *et al.*, 2005) using an Olympus CH30 binocular microscope (40X) with vernier caliper. To facilitate the counting of the pollen grains, a manual counter was used and the numbers of pollen per slide were counted. The mean quantity of pollen grains in 10 μ l was estimated for each location whereas the quantity of pollen grains in 1ml glycerol mixture was taken as the number of pollen grains in the anthers used in the preparation using the formula,

 $\frac{n x \operatorname{1ml} x \operatorname{10^3}}{10 \,\mu l} \dots 3.4$

Where n = mean number of pollen grains in 10 µl. Number of pollen grains in single anther was obtained by dividing the mean number of pollen grains in 10 µl by the number of anthers used. The number of pollen grains produced per flower was obtained by multiplying the number of anthers per flower and the number of pollen grains in an anther.

The pollen-ovule ratio of *T. tetraptera* flower was determined by the ratio of pollen grains in a polyad to the number of ovules in the ovary of that flower. Pollen grain shape was described based on the ratio between the length of the polar axis (P) and equatorial diameter (E), size and structure according to Erdtman (1969).

3.3.3 Determination of inflorescence morphology

Twenty five inflorescences were sampled from two trees each in the two locations during the first flowering cycle. The number of flowers on sampled inflorescence was counted using a tally counter while the length of each inflorescence was measured on the field using a metre rule graduated in centimeters. The number of inflorescences on a twig (inflorescence clusters) of *T. tetraptera* was determined by sampling fifty twigs from four trees in each location.

The number of matured fruits per inflorescence was determined by sampling fifty fruiting inflorescence from four trees in each location.

3.3.4 Insect visitors and interaction with *Tetrapleura tetraptera*

The insect visitors associated with *T. tetraptera* flowers were observed from the top of the selected trees at NIHORT and FRIN in the second blooming cycle during the peak flowering season. Observations took place from 7.00 am to 5.00 pm for three days. The observations were conducted every two hours for 5 minutes during the sampling period

according to the methodology of Sine'bou *et al.* (2016). Insect species or taxonomic group, time of visitation, frequency (number of visits per minute to an inflorescence) and duration of visit were monitored and documented. The time spent by an insect on an inflorescence was monitored using a stop watch. Insect visitor abundance and behaviour were also assessed. The insect visitors were categorized into pollinators and non-pollinators according to their observed behaviours. Insect visitor is considered a pollinator if it made contact with the sex structures and non-pollinator if it did not contact. Insect visitors were caught using sweep net and preserved in specimen bottles containing formalin soaked cotton wool. Images of the insect visitors were taken and viewed under binocular dissecting microscope at x40 magnification for presence of *T. tetraptera* pollen on them (Gan *et al.*, 2013).

The pollen carrying capacity was evaluated directly by counting the polyads on the various parts of the insect body. Where more than one insect was caught for a particular species, the mean of the polyads count was taken. Field observations in addition to these data were used to determine which insect visitors were likely to be pollinators. *Tetrapleura tetraptera* pollen was easily distinguished on insect bodies due to the understanding and identification of the pollen shapes and sizes during the previous studies on pollen morphology. Specimens of insect visitors were identified and deposited at the Entomology unit of Forestry Research Institute of Nigeria, Ibadan.

3.3.5 Reproductive efficiency of *Tetrapleura tetraptera*

Two trees were selected in each location and twenty inflorescences were randomly sampled and left open. An inflorescence was chosen as the sample unit because the flowers were small and densely compacted (Pires and Freitas, 2007). The number of flowers on each of the sampled inflorescences was counted and recorded. The fruit set rate of *T. tetraptera* of trees were determined in each location at phenological stage Fr1 (persisted expanded ovary) during the first fruiting cycle. On each sampled inflorescence, the total number of fruits set was counted and recorded for each site. Fruit set rate was determined for each site by expressing the number of fruits set per inflorescence as a percentage of the number of flowers per inflorescence.

Where FSR = Fruit set rate per inflorescence,

F = Number of fruits effectively set

H = Total number of flowers on an inflorescence

At phenological stage fr3 (developed fruit with no sign of abortion), the fruit drop rate (FDR) was estimated by counting number of developed fruit on each of the sampled inflorescence of *T. tetraptera*. Percentage fruit drop rate was calculated as;

FDR (%) = $\frac{\text{Number of mature fruits-Number of fruits set}}{\text{Number of gynoecium produced}} \times 100.....3.6$

3.3.6 Data analysis

The mean values of floral variables and standard deviation per each location was calculated and comparisons between the means of the two locations were carried out using student's t-test in Statistical packages for social sciences (SPSS). Abundance and frequency distribution of insect visitors to *T. tetraptera* trees was estimated using Mann-Whitney U-test. Frequency distribution chart was used to estimate the capacity of insect visitors for carrying pollen grain. Data on reproductive efficiency were presented using descriptive statistics.

3.4 Study 3: Morphological variation in fruits/seeds of *Tetrapleura tetraptera* from four different sources in southwest Nigeria

One hundred and fifty matured fruit-pods were selected from each of the four sources in a completely randomized design (Table 3.1). These fruits were harvested from four to six trees depending on availability which were 20 to 50 metres apart to avoid relatedness. The width, length and number of seeds were measured for each fruit-pod. Length of fruit was determined by tracing the edge of the pod from the point of attachment to the twigs up to the tip and transferring the measurement onto a metric ruler for accurate reading, while the width was measured using a digital vernier caliper across the widest portion. Each pod was broken and the total number of seeds counted. A total of 100 seeds were

batched per source in three replicates and their weight determined using an electronic digital weighing balance (Model E300).

3.4.1 Data analysis

The data on fruit morphological traits were analysed using analysis of variance (ANOVA) and Tukey's test was used to separate significantly different means. Coefficient of variation (%) was estimated to determine the amount of variation in fruit traits within and between sources. Pearson correlation coefficient (r) was used to test relationship between fruit traits and geo-climatic variable.

| Sources | Latitude | Longitude | Temperature | Rainfall | Relative | Altitude |
|---------|-------------|------------|-------------|----------|-----------|----------|
| | (N) | (E) | (°C) | (mm) | humidity | (m) |
| | | | | | (%) | |
| Ibadan | 7°23'36′ - | 3°51'44‴ - | 22 - 31 | 9.4 – | 59 - 86.5 | 150 – |
| | 7°24'22" | 3°50' 56" | | 293.1 | | 250 |
| Iwo | 7°30 '36" - | 4°01′12″ - | 24 - 30 | 9 - 237 | 60 - 72 | 200 – |
| | 7°43′12″ | 4°16′12″ | | | | 500 |
| Aponmu | 7°12′ - | 5°3′36″ - | 21 - 29.7 | 9 - 244 | 70 - 78 | 230 – |
| | 7°24′ | 5°10′48″ | | | | 600 |
| Mamu | 6°54′0″ - | 3°38′24″ - | 19 – 28 | 11 - 227 | 70 - 85 | 60 - 145 |
| | 7°02′60″ | 3°47′24″ | | | | |

Table 3.1. Geographical and meteorological description of fruit collection sites

Source: Nigerian Meteorological Agency (NIMET), 2018

3.5 Early growth assessment of *Tetrapleura tetraptera*

3.5.1 Effect of seed sources on germination and early growth of *Tetrapleura tetraptera* seeds

From each seed source, two hundred (200) seeds were soaked in concentrated H_2SO_4 acid (90%) for 5 minutes to break the dormancy as a result of hard seed coat (Alaba *et al.*, 2006). The seeds were later washed in distilled water before sowing in labeled germination plastic trays filled with sterilized-washed river sand (Plate 3.1). These trays were laid out under a high humidity propagator in the nursery while watering was done daily to field capacity. At two leaf stage, fifty uniformly growing seedlings from each source were transplanted into 16 cm x 14 cm x 12 cm black polythene pots filled with topsoil (Plate 3.2). A completely randomized design experiment was used to evaluate early growth and development of seedlings (Table 3.2). The seedlings were watered daily to field capacity. The following variables were measured fortnightly for twenty four weeks: seedling height, collar diameter and number of leaves. The height of seedling was measured from the the tip of the youngest leaf to the root level; stem collar diameter was measured with a digital vernier caliper at 1cm height above the soil surface; while leaf development was assessed by directly counting the number of leaves.

3.5.2 Assessment of biomass accumulation of *Tetrapleura teraptera* seedlings from four different sources

Five harvests were made during the assessment period over twelve month's period. At each harvest, five seedlings were chosen at random from each seed source. The seedlings were then uprooted and put in a water container so that the soil around the roots could be carefully dislodged and washed away. Then, the uprooted seedlings were divided into root and shoot components while stem biomass included the leaf stalks. The root and shoot components of each seedlings were weighed to determine the fresh weight before oven-drying at 80°C to constant weight. The samples were allowed to cool before the dry weights of the samples were measured using an electronic weighing balance (Model E 300).



a. Tetrapleura tetraptera seeds



b. Germinated Tetrapleura tetraptera seedlings

Plate 3.1. Germination of *Tetrapleura tetraptera* seeds in the nursery



Plate 3.2. Tetraplura tetraptera seedlings at two-leaf stage

| AP | IW | IB | MM | AP | IW | IB | MM | IB | IW |
|----|----|----|----|-----|----|----|----|----|----|
| IB | MM | AP | IW | IB | MM | AP | IW | AP | MM |
| MM | IW | IB | AP | MM | IW | IB | AP | IB | IW |
| IW | IB | IW | MM | Iwo | IB | IW | MM | IW | IB |
| IB | MM | AP | IW | IB | MM | AP | IW | AP | MM |
| MM | IW | IW | AP | MM | IW | IB | AP | IB | IW |
| AP | IB | MM | IW | AP | IB | MM | IW | MM | IB |
| IW | AP | IW | MM | IW | AP | IW | MM | IW | AP |
| IB | MM | AP | IW | IB | MM | AP | IW | AP | MM |
| AP | IW | IB | MM | AP | IW | IB | MM | IB | IW |
| MM | AP | IW | IB | MM | AP | IW | IB | IW | AP |
| IW | IB | MM | AP | IW | IB | MM | AP | MM | IB |
| IB | MM | AP | IW | IB | MM | AP | IW | AP | MM |
| AP | IW | IB | MM | AP | IW | IB | MM | IB | IW |
| MM | IB | IW | AP | MM | IB | IW | AP | IW | IB |
| IW | AP | MM | IB | IW | AP | MM | IB | MM | AP |
| IB | MM | AP | IW | IB | MM | AP | IW | AP | MM |
| AP | IW | MM | IB | AP | IW | MM | IB | MM | IW |
| MM | IB | IW | AP | MM | IB | IW | AP | IW | IB |
| IW | AP | IB | MM | IW | AP | IB | MM | IB | AP |
| | | | | | | | | | |

Table 3.2. Experimental layout of *Tetrapleura tetraptera* seedlings in the nursery

* AP – Aponmu, IB – Ibadan, MM – Mamu, IW - Iwo

The mean leaf area of a seedling was estimated by finding the area of a pinnule using the grid method. Since *T. tetraptera* has a bipinnately compound leaves, estimating leaf area of all the pinnules using the grid method is laborious and time consuming, thus the following variables were estimated;

Mean area of fifty pinnules, mean number of pinnules on 50 leaflets (pinnae), mean number of pinnae per leaf on a 50 seedling and the number of leaves on a seedling. Thus, the mean leaf area (cm^2) of a seedling was determined as multiples of mean area of pinnules, mean number of pinnules per pinnae, mean number of pinnae per leaf and number of leaves per seedling.

The dry weights and mean leaf area were then used to calculate the relative growth rate (RGR) and absolute growth rate (AGR), and net assimilation rate (NAR) using equations 3.7, 3.8 and 3.9.

$$W_1 =$$
 Initial Dry Weight

- $W_2 =$ Final Dry Weight
- $A_{I} =$ Initial leaf area
- $A_2 =$ Final leaf area
- $T_1 =$ Initial Harvest Time
- $T_2 =$ Final Harvest Time
- ln = Natural logarithm

3.5.3 Data analysis

The data obtained were analysed at P < 0.05 using generalized linear model (GLM) in Minitab v.17. Coefficient of variation (%) was estimated to calculate the amount of variation in growth characteristics within and between sources using SPSS and descriptive statistics.

3.6 Study 5: Vegetative propagation of *Tetrapleura tetraptera* using leafy stem scuttings

Double node leafy stem cuttings were collected from seventy uniformly growing seedlings aged eight months from each of the four seedling sources (Ibadan, Iwo, Aponmu and Mamu) using a secateur. Stem cuttings (6 cm long) were taken concurrently on the shoot of the stockplant, while the location of each cut was noted on the shoot. From the seedlings raised in study 4, four hundred (400) leafy stem cuttings were derived. The exercise was done early in the morning and the severed cuttings were kept moist after collection by immersing them in water. In all, a total of one thousand six hundred (1600) cuttings were taken from the four sources. All cuttings had their leaves reduced to half, to prevent excessive transpiration but aid photosynthesis. Plant growth regulators; Indole-3-Butyric acid (IBA), Naphthalene Acetic acid (NAA), combination of IBA and NAA (IBA + NAA) were used on the cuttings at different concentrations. The growth regulators (auxins) were prepared following the standard procedure of Leakey et al. (1990). Thus, a micro pipette was used to dissolve 0.1mg, 0.15mg and 0.2mg of each regulator into 10 ml of ethanol. The resultant solution was subsequently mixed with 90 ml of distilled water. From the resulting solution, the needed concentrations (100ppm, 150ppm and 200ppm) were derived. Coconut fruits were harvested from a home garden in Ibadan and the coconut water was extracted into a conical flask within the nursery. The coconut water was extracted using a filter paper and mixed thoroughly with distilled water to achieve 25% dilution following the method of Agele et al. (2013). The cuttings's end was treated with these growth regulators using the quick dip method (Akinyele, 2010) for five seconds. After the treatment, the cuttings were placed in germination trays filled with sterilized river sand to a depth of 1cm. Five cuttings from each source, nodal position and regulator concentration were set in a germination tray and replicated twice. In all, there were twelve treatments in addition to the control (no hormone treatment). Thereafter, the germination trays were placed in a high humidity propagator and the treatments arranged in 3x4x4 factorial using a completely randomized design.

Watering was performed daily with a hand sprayer to field capability and the following morphological variables were assessed after sixty days.

- i. Percentage survival (%)
- ii. Number of roots per cutting
- iii. Total root length
- iv. Length of longest root
- v. Shoot height
- vi. Number of leaves per cutting

Four seedlings were also selected from each of the stockplant source for biomass assessment (shoot dry weight). Relative growth rate (RGR) and average growth rate (AGR) was determined.

3.6.1 Data analysis

The data obtained were analysed statistically using SPSS v.16 (SPSS Chicago, USA). Rooting and survival data were transformed using Arcsin transformation in order to normalize the data. The differences among mean values were separated using Duncan's multiple range tests at p < 0.05. Pearson correlation coefficient (*r*) was used to test relationship between two stockplant variables and rooting variables.

CHAPTER FOUR

RESULTS

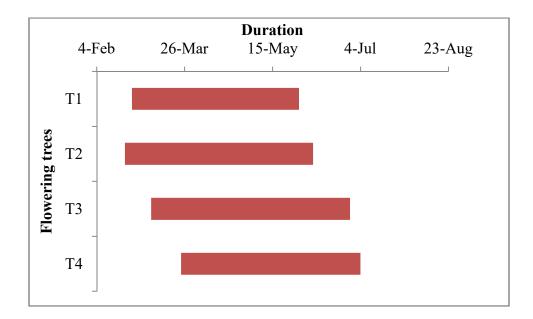
4.1 Flowering phenology of *Tetrapleura tetraptera* at the study sites

4.0

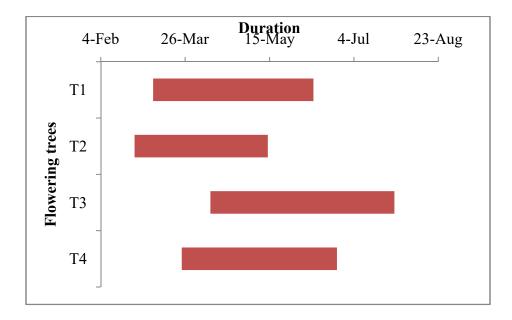
Three flowering cycles were observed during the study period, two in 2017 and one in 2018. Flowering was first observed in February and lasted till July while second flowering onset was recorded in the mid of October to end of December at both locations (Figures 4.1 and 4.2). The third flowering cycle was observed from mid January to August at FRIN while that of NIHORT commenced from mid-February to early July in 2018 (Figure 4.3). The first and third flowering coincided with the peak of dry season and extending to early rainy season immediately after leaf flush while the second flowering occured at the end of rainy season coinciding with the beginning of dry season. Flowering was observed on all tagged trees at FRIN during the first flowering cycle while four out of six trees flowered in NIHORT during the first flowering cycle (Figure 4.1). The difference in the onset of flowering among the first flowering cycle (Figure 4.1). The difference in the onset of flowering among trees in the second flowering cycle ranged from 7 to 14 days within the locations while that of the third flowering cycle ranged between 7 and 14 days in NIHORT and 96 days at FRIN (Figures 4.2 and 4.3).

4.1.1 Duration of flowering of *Tetrapleura tetraptera* at the study sites

Flowering duration of *T. tetraptera* trees was similar in both locations and ranged between 95 and 113 days in FRIN, while in NIHORT, it ranged between 79 and 109 days during the first flowering cycle (Figure 4.1). In second flowering cycle, flowering ranged from 39 to 60 days at FRIN site, while that of NIHORT ranged from 59 to 60 days. However in third flowering cycle, flowering duration ranged from 82 to 108 days in FRIN, while NIHORT ranged from 98 and 133 days respectively (Figures 4.3a and 4.3b). Flowering duration was shorter in the second cycle as compared to the first and third cycle in both locations.

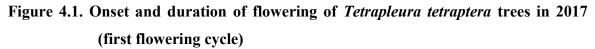


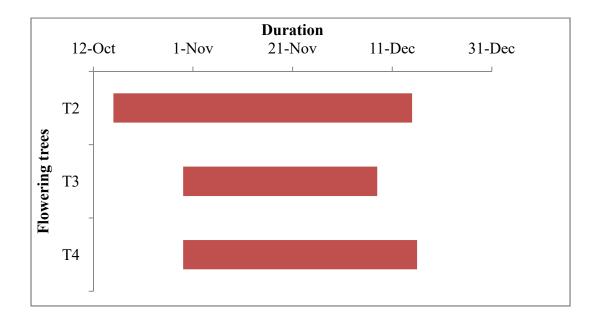
Location i. FRIN



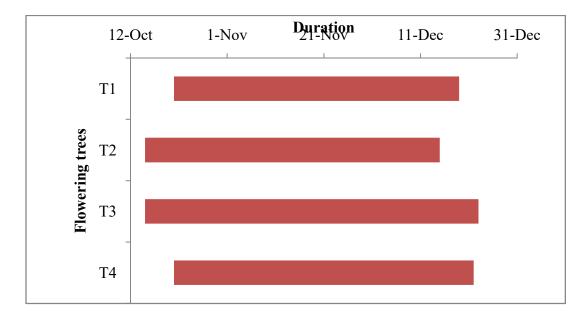
Location ii. NIHORT

*T1, T2, T3 and T4 denote Tetrapleura tetraptera flowering trees





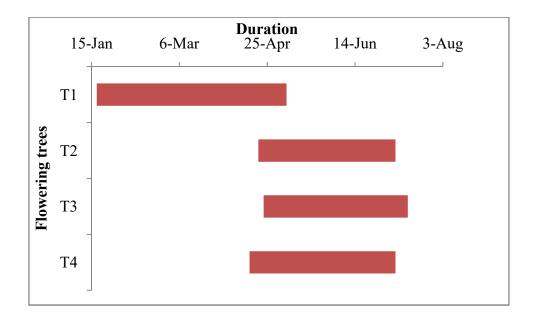
Location i. FRIN



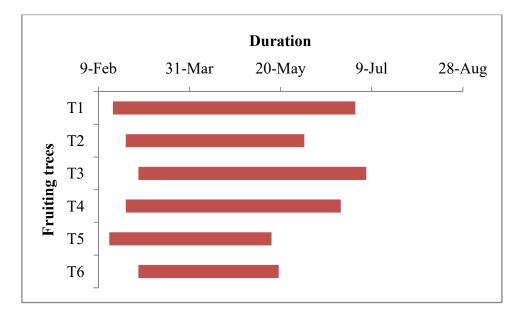
Location ii. NIHORT

*T1, T2, T3 and T4 denote Tetrapleura tetraptera flowering trees

Figure 4.2. Onset and duration of flowering of *Tetrapleura tetraptera* trees in 2017 (second flowering cycle)



a. FRIN



b. NIHORT

*T1, T2, T3, T4, T5 and T6 denote Tetrapleura tetraptera flowering trees

Figure 4.3. Onset and duration of flowering of *Tetrapleura tetraptera* trees in 2018 (third flowering cycle)

The flowering duration of *T. tetraptera* trees at FRIN was highest $(104 \pm 7.6 \text{ days})$ in the first flowering cycle, followed by the third flowering cycle $(88 \pm 13.7 \text{ days})$ while the least $(49 \pm 10.6 \text{ days})$ flowering duration was recorded in the second flowering cycle (Table 4.1). However, the flowering duration at NIHORT site in the second flowering cycle $(62 \pm 4.4 \text{ days})$ was shorter than the first $(94 \pm 12.3 \text{ days})$ and third flowering cycle $(107 \pm 22 \text{ days})$. When compared, the flowering duration of trees at FRIN during the first cycle $(104 \pm 7.6 \text{ days})$ was higher than NIHORT $(94 \pm 12.3 \text{ days})$, whereas the mean flowering duration at NIHORT in the second flowering cycle $(62 \pm 4.4 \text{ days})$ was higher than FRIN ($49 \pm 10.6 \text{ days}$) (Table 4.1).

4.1.2 Floral synchrony of *Tetrapleura tetraptera* trees at FRIN and NIHORT (Ibadan)

The floral synchrony index of *T. tetraptera* trees in FRIN was highest (0.82 ± 0.03) during the first flowering cycle, followed by third flowering cycle (0.67 ± 0.22) , but was least during second flowering cycle (0.43 ± 0.29) (Table 4.1). The individual tree floral synchrony index (X_i) at FRIN ranged from 0.78 to 0.86 in the first flowering cycle (Appendix 3). During the second flowering cycle, the floral synchrony index ranged from 0.47 to 0.66 while that of the third flowering cycle varied between 0.34 and 0.74 (Appendix 3).

The floral synchrony index was highest among *T. tetraptera* trees in the third flowering cycle ($Z=0.88 \pm 0.11$), followed by the first cycle ($Z=0.66 \pm 0.1$) and was least (0.34 ± 0.27) during the second cycle (Table 4.1). The floral synchrony index (X_i) of each *T. tetraptera* tree at NIHORT ranged from 0.51 to 0.73 in the first flowering cycle. The second flowering cycle ranged from 0.47 to 0.54 while the third flowering cycle ranged from 0.81 to 1.0.

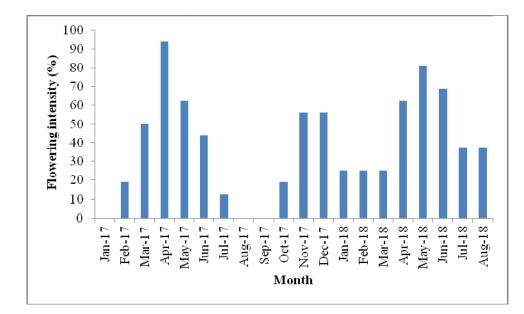
4.1.3 Flowering intensity among *Tetrapleura tetraptera* trees at FRIN and NIHORT study sites in Ibadan

Three flowering peaks were recorded during the observation in respect to the flowering cycles. At FRIN, the first peak (93.8%) in the species flowering was recorded in April (2017), second peak (56.3%) in November and December (2017) and third peak (81.3%)

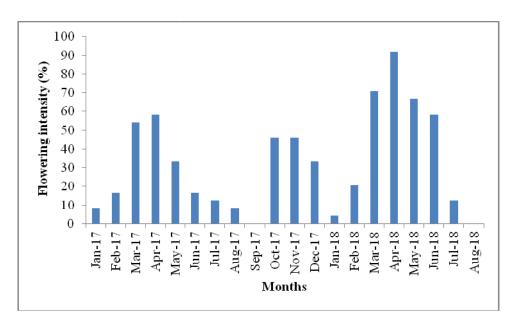
in May (2018) (Figure 4.4a). However, NIHORT trees attained their first peak (58.3 %) in April (2017), second peak (45.8 %) in November (2017) and the third peak (91.6 %) was recorded in April (2018) (Figure 4.4b). Flowering events ceased in August (FRIN) and September (NIHORT) but resumed in October signalling the beginning of another flowering cycle.

Table 4.1. Flowering duration and Floral synchrony (Z) of *Tetrapleura tetraptera* (mean \pm sd) at Forestry Research Institute of Nigeria (FRIN) and National Horticultural Research Institute (NIHORT), Ibadan

| | Flowering duration (days) Floral synchrony | | | | | |
|-----------------|--|----------------|-----------------|----------------|--|--|
| Flowering cycle | FRIN | NIHORT | FRIN | NIHORT | | |
| First | 104.3 ± 7.6 | 93.8 ± 12.3 | 0.82 ± 0.03 | 0.66 ± 0.10 | | |
| Second | 48.7±10.6 | 62.3 ± 4.4 | 0.43 ± 0.29 | 0.34 ± 0.27 | | |
| Third | 87.8 ± 13.7 | 106.7 ± 22.0 | 0.67 ± 0.22 | $0.88\pm~0.11$ | | |







b. NIHORT

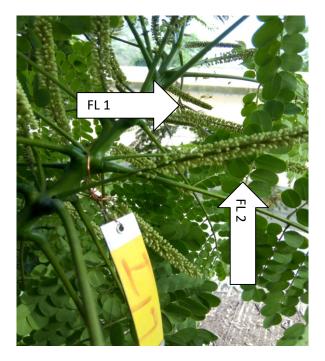
Figure 4.4. Flowering intensity of *Tetrapleura tetraptera* trees at Forestry Research Institute of Nigeria (FRIN) and National Horticultural Research Institute (NIHORT), Ibadan

4.1.4 Floral development and rate of anthesis of *Tetrapleura tetraptera* inflorescence at FRIN and NIHORT, Ibadan

After initiation, flower buds (fl1) attained stage fl2 (developing buds), between 14 to 21 days. Buds became well developed (stage fl3) between 21 to 26 days after bud initiation (Appendix 4). The anthesis stage fl4 (flower opening) occurred 21 to 32 days thereafter fl1 at both locations. Anthesis started between 0700h and 1300h, although not all flower buds opened at the same time on an inflorescence. At onset of anthesis, the petals curved inwards while the stamens were pressed to it and gradually, the filaments bent by placing the anther above the center of the flower. The filaments bent backward after exposing the anthers while the anthers presented yellow pollen grains in one to two hours after opening. Floral development of T. tetraptera is as shown in Plate 4.1. Flower opening on an inflorescence was completed between one to six days at FRIN, while it ranged from one to three days at NIHORT. The highest (60%) proportion of inflorescences at NIHORT completed anthesis on the second day, followed by the third day (30%), while the lowest (10%) anthesis was obtained on the first day (Figure 4.5a). However, the proportion of inflorescences that completed anthesis was highest (35%) on second day at FRIN, followed by third and fourth day (25%), while the lowest proportion (5%) was completed in one, five and six days (Figure 4.5b). The flowering period of an inflorescence was 29.5 \pm 1.57 days (Appendix 4). The opening of flower buds often begins from apex to base (acropetal) while sometimes from base to apex (basipetal) on the inflorescence (Plate 4.1c and 4.1d). Wilting of petals began 3.4 ± 0.49 days after anthesis (Appendix 5).

4.1.5. Fruiting phenology of *Tetrapleura tetraptera* trees

Fruit set on *T. tetraptera* inflorescence occured between April and June in the first fruiting cycle (Figure 4.6) while in the second cycle, it occured between November and December (Figure 4.7). The third cycle occured from February to August (Figure 4.8). Fruit initiation occurred 6.5 ± 0.51 days after wilting of petals (Plate 4.2a). At this stage, the fruit length was 0.7 ± 0.02 cm and flowers became discoloured and dehydrated. Fruit abortion on inflorescence was observed from 7 to 14 days after fruit set (Plate 4.2b). The trend of fruit length development in the species followed a sigmoid growth curve (Figure 4.9).





a. Young (fl1) and growing floral buds (Fl2) at FRIN

b: fl3: Developed buds at NIHORT

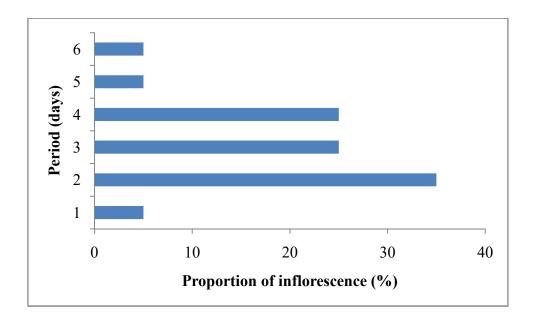


c. Acropetal anthesis

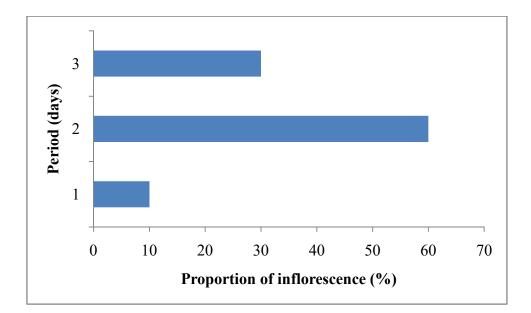


d. Basipetal anthesis



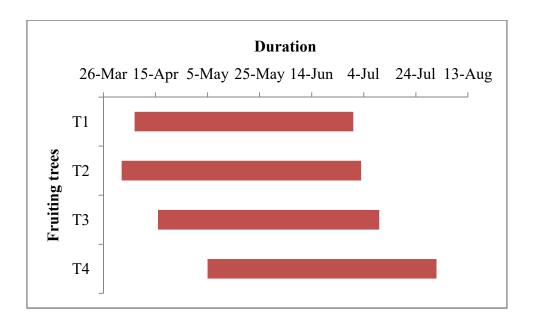


a. FRIN

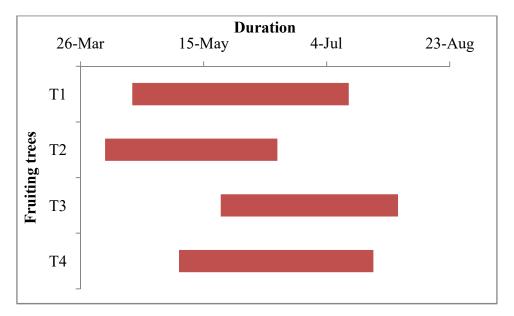


b. NIHORT

Figure 4.5. Daily rate of anthesis of *Tetrapleura tetraptera* inflorescence in FRIN and NIHORT, Ibadan



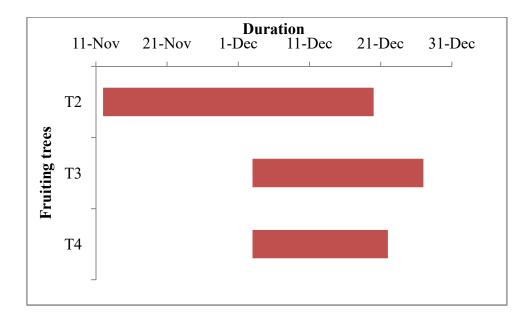
a. FRIN



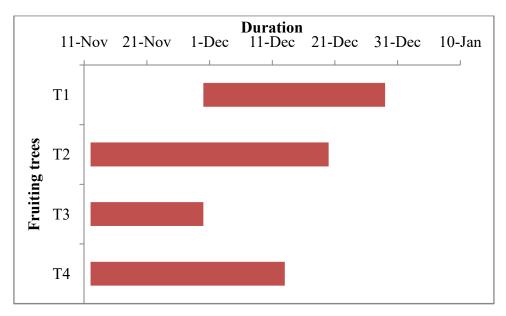
b. NIHORT

Figure 4.6. Onset and duration of fruit initiation of *Tetrapleura tetraptera* at FRIN (a) and NIHORT (b) in the first cycle in 2017

*T1, T2, T3 and T4 denotes fruiting trees in each location



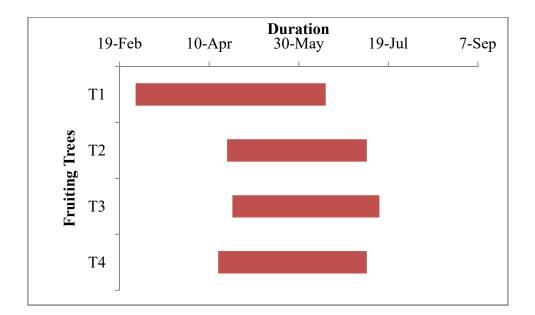
a. FRIN



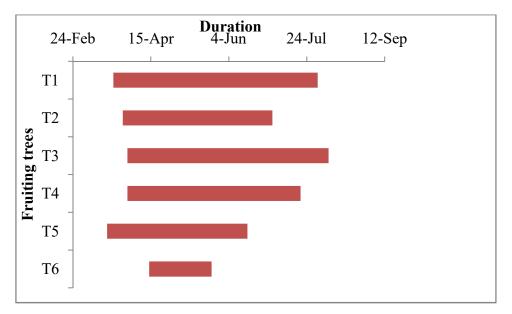
b. NIHORT

Figure 4.7. Onset and duration of fruit initiation of *Tetrapleura tetraptera* at FRIN (a) and NIHORT (b) in the second cycle in 2017

*T1, T2, T3 and T4 denotes fruiting trees in each location







b.NIHORT

Figure 4.8. Onset and duration of fruit initiation of *Tetrapleura tetraptera* at FRIN (a) and NIHORT (b) in the third cycle in 2018

*T1, T2, T3 and T4 denotes fruiting trees in each location

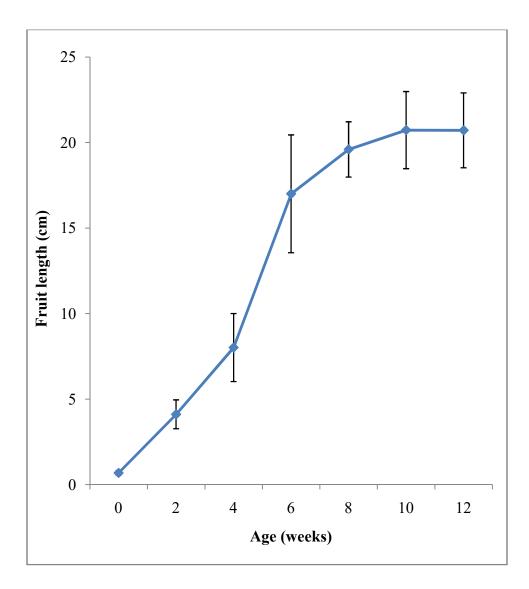


Figure 4.9. Fruit length (cm) of *Tetrapleura tetraptera* from fruit set to maturity at NIHORT (Ibadan) (n=25, mean ± sd)

There was progressive increase in fruit length until the 12^{th} week when the fruit length remained constant. Fruit length was 8.01 ± 0.39 cm, 28 days after fruit set while young fruits remained green in colour (Plate 4.2b). The fruit colour varies from green to brownish green at 42 days after fruit set with fruit length of 17 ± 0.69 cm (Plate 4.2). Fruit length measured 20.7 ± 0.45 cm at 70 days after fruit set while fruits began to turn brown as maturity progressed. The length of matured fruits ranged from 18 to 24.7 cm (94 days after fruit set) while the mean fruit length was 20.7 ± 0.43 cm. Matured fruits fall started at 104 days after fruit set.

4.1.6 Period of fruit maturation of *Tetrapleura tetraptera* at FRIN and NIHORT (Ibadan)

Fruiting cycles had significant effect on the period (days) of *T. tetraptera* fruit maturity (Appendices 5 and 6). However, there was no significant difference in the period of fruit maturity between the locations. The second fruiting cycle was significantly different from the first and the third fruiting cycle at both locations. The number of days taken to attain fruit maturity was significantly higher in the second fruiting cycle when compared to either the first or third cycle (Table 4.2). Period of fruit maturity in FRIN was highest (117 \pm 4.36 days) in second fruiting cycle compared to first (91 \pm 3.74 days) and third cycle (88 \pm 6.4 days). Similarly, the highest period of fruit maturity at NIHORT was recorded in the second fruiting cycle (125 \pm 4.57) compared to first (85 \pm 0.96) and third cycle (89 \pm 3.71).

4.1.7 Pattern of climatic variables at Forestry Research Institute of Nigeria (FRIN) Ibadan

The monthly rainfall ranged from 0 mm to 293.1 mm (Figure 4.10). The rainfall distribution pattern was bimodal with peak in June and July. Monthly minimum temperature ranged from 19.5°C and 28.1°C while monthly maximum temperature ranged from 25.3°C to 34.9°C. Highest minimum (24.4°C) and maximum temperature (34.9°C) in 2017 was observed in January and March respectively while that of 2018 was in March (Figure 4.10). The mean monthly relative humidity ranged from 50% to 86.7% (Figure 4.10).

Table 4.2. Duration of fruit maturity (days) of *Tetrapleura tetraptera* in FRIN andNIHORT, Ibadan (mean ±sd)

| Fruiting season | FRIN | NIHORT |
|-----------------|----------------------|----------------------|
| First | $91^{a} \pm 3.74$ | $85.25^{a} \pm 0.96$ |
| Second | $117.3^{b} \pm 4.36$ | $124.8^{b} \pm 4.57$ |
| Third | $88^{a} \pm 6.4$ | $88.83^{a} \pm 3.71$ |

*Means with same letters are not significantly different from each other



a. Fruit set (fr1) at 7days after anthesis



b. Fruits at 15 to 21 days after fruit set



c. Developing fruits at 28 days after fruit set



d. Matured fruit 90 days after fruit set

Plate 4.2: Pattern of fruit development in *Tetrapleura tetraptera* trees from NIHORT (Ibadan)

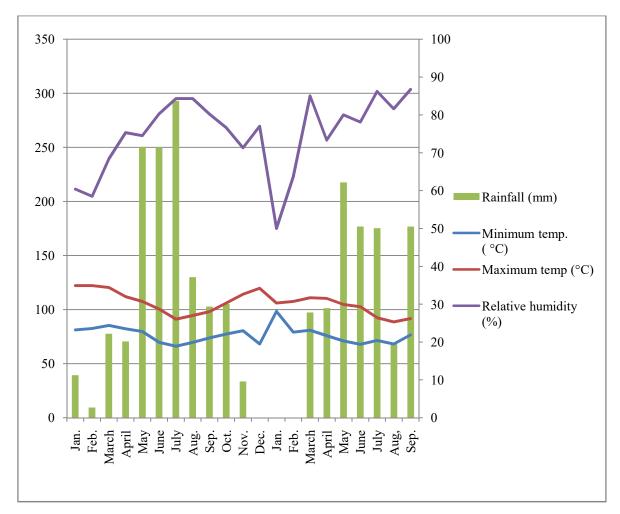


Figure 4.10. Mean monthly distribution of climatic variables in 2017/2018 at FRIN, Ibadan

4.1.8 Spearman's rank correlation between climatic variables and phenophase frequency

There was positive significant correlation between number of flowering trees and maximum monthly temperature (Table 4.3). Minimum monthly temperature and rainfall were not significantly correlated with frequency of flowering trees while relative humidity was negatively correlated with flowering frequency (Table 4.3). There was negative correlation between fruiting phenophase and temperature. However, minimum temperature was significantly correlated with fruiting phenophase while maximum was not. Significant positive correlation was also found between fruiting and monthly rainfall. Similarly, there was positively correlation between relative humidity and fruiting (Table 4.3).

4.2 Flower and inflorescence morphology of *Tetrapleura tetraptera*

4.2.1 Flower morphology

Flower buds were densely arranged on inflorescence in between the axils of the leaf petiole on the growing shoots. The inflorescence of *T. tetraptera* is a spike. Flowers are hermaphrodites, pentamerous and zygomorphic (bilaterally symetrical) with purplish-pink or creamy-yellow corrola. The androecium is composed of ten free stamens arranged in two whorls (five each) and fused at the base while the bilobed anthers are terminally located on the filaments presenting a gland at the apex (Plate 4.3a). The gynoecium is bottle-shaped and centrally located on the receptacle (Plate 4.3b). The ovary is superior, unilocular with marginal placentation while the style is slightly bent. The petals are convex-shaped and fused at the base to the receptacle below the gynoecium such that the corrolla form a bell shape. The stamens and style have low spatial separation (Table 4.4). The pedicel is hairy. There were no significant differences in the measured floral parts except for length of ovary that differed between the locations (Table 4.4). Ovary length was higher at NIHORT (4.65 ± 0.14 mm) than FRIN (4.48 ± 0.14 mm).

| | 1 | Flowering | Fruiting | |
|------------------------|---------|-----------|----------|---------|
| Climatic variables | FRIN | NIHORT | FRIN | NIHORT |
| Rainfall | 0.041 | -0.251 | 0.574* | 0.454* |
| Maximum temperature | 0.529* | 0.518* | -0.197 | -0.157 |
| Minimum temperature | 0.412 | 0.288 | -0.475* | -0.438* |
| Relative humidity | -0.602* | -0.224 | 0.323 | 0.451* |

Table 4.3. Spearman rank correlation coefficient (r_s) of climatic variables and phenophase frequency of *Tetrapleura tetraptera* trees

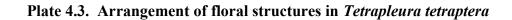
* Significant at P<0.05



a. A matured flower of *Tetrapleura tetraptera* (x40)



b. Marginal arrangement of ovules in the ovary (X40)



| Floral parameter | FRIN | NIHORT | P – value |
|----------------------|----------------|----------------|---------------------|
| Stamen length (mm) | 6.05 ± 0.67 | 6.45 ± 0.57 | 0.481 _{ns} |
| Style length (mm) | 3.75 ± 0.02 | 3.74 ± 0.04 | 1.000 _{ns} |
| Ovary length (mm) | 4.48 ± 0.14 | 4.65 ± 0.14 | 0.03* |
| Ovary diameter (mm) | 1.05 ± 0.05 | 1.06 ± 0.09 | 0.778_{ns} |
| Pedicel length (mm) | 3.25 ± 0.02 | 3.27 ± 0.03 | 0.621 _{ns} |
| Anther length (mm) | 1.17 ± 0.38 | 1.15 ± 0.35 | 0.972 _{ns} |
| Anther diameter (mm) | 0.68 ± 0.08 | 0.65 ± 0.04 | 0.501 _{ns} |
| Style width (mm) | 0.02 ± 0.005 | 0.02 ± 0.007 | 0.178_{ns} |
| Petal length (mm) | 4.45 ± 0.11 | 4.56 ± 0.19 | 0.43 _{ns} |
| Ovules per ovary | 18.8 ± 2.61 | 18.0 ± 1.3 | 0.799 _{ns} |
| Polyad:ovule | 1.07 ± 0.08 | 1.12 ± 0.08 | 0.169 _{ns} |

Table 4.4. Morphological characteristics of floral parts of *Tetrapleura tetraptera* from the study locations, Ibadan, Nigeria (n = 20, means \pm sd)

*significant at P < 0.05; ns – not significant P > 0.05

4.2.2 Pollen grains morphology/count of *Tetrapleura tetraptera*

There were no significant differences in the number of polyads per pollen grains of *T*. *tetraptera* (Table 4.5). The number of pollen grains per 10 μ l (N = 20 flowers) at FRIN was 310 \pm 35.4 while that of NIHORT was 304 \pm 23.4. The total number of pollen grains per flower at FRIN was higher (310,000 \pm 35,369.5) than NIHORT (304,000 \pm 21,283.8) and equivalent to 31,000 and 30,400 pollen grains per anther respectively (Table 4.5). The pollen grains were waxy and arranged asymmetrically and irregularly in a polyad (Plate 4.4). The number of pollen grains in a polyad of *T. tetraptera* was 20 \pm 2.31. The polar axis of pollen grains at FRIN was 16.4 \pm 0.35 μ m while the equatorial diameter was 14.7 \pm 0.7 μ m. Similarly, the polar axis of pollen grains at NIHORT was 17.7 \pm 3.18 μ m while the equatorial diameter was 16.2 \pm 1.77 μ m (Table 4.5). The apertural status of pollen grains from both locations was 3-porate while the shape was prolate-spheroidal (1.09-1.11 μ m).

4.2.3 Morphology of the inflorescence of *Tetrapleura tetraptera* from FRIN and NIHORT, Ibadan

There were no significant differences in the number of flowers per inflorescence of *T. tetraptera* (Table 4.6). The number of flowers per inflorescence of *T. tetraptera* at FRIN was 297 ± 27.3 while that of NIHORT was 294 ± 40.3 . Similarly, there was no significant difference in the length of an inflorescence of *T. tetraptera* between the locations (Table 4.6). The length of Inflorescence in NIHORT was slightly higher (9.3 \pm 2.85 cm) than FRIN (8.8 \pm 1.59 cm). However, there was a significant difference in the number of inflorescence per twig of *T. tetraptera*. The number of inflorescence per twig was higher (16.5 \pm 6.64) in FRIN than NIHORT (7.3 \pm 3.2) (Table 4.6).

| Parameters | NIHORT | FRIN |
|--------------------------------|--------------------|--------------------|
| Polyad per anther | 1,520 | 1,550 |
| Polyad per flower | 15,200 | 15,500 |
| Pollen grains/polyad | 20 ± 2.31 | 20 ± 2.31 |
| Polyad per 10 µl | 304 ± 23.4 | 310 ± 35.4 |
| Pollen grains/anther | 30,400 | 31,000 |
| Pollen grains/flower | 304,000 ± 21,283.8 | 310,000 ± 35,369.5 |
| Polar axis (µm) | 17.7 ± 3.18 | 16.4 ± 0.35 |
| Equatorial diameter (µm) | 16.2 ± 1.77 | 14.7 ± 0.7 |
| Polar axis/Equatorial diameter | 1.09 | 1.11 |
| Pollen shape | Prolate spheroidal | Prolate spheroidal |
| Apertural status | 3 – porate | 3 – porate |

Table 4.5. Polyad morphological features of *Tetrapleura tetraptera* from the study locations, Ibadan, Nigeria

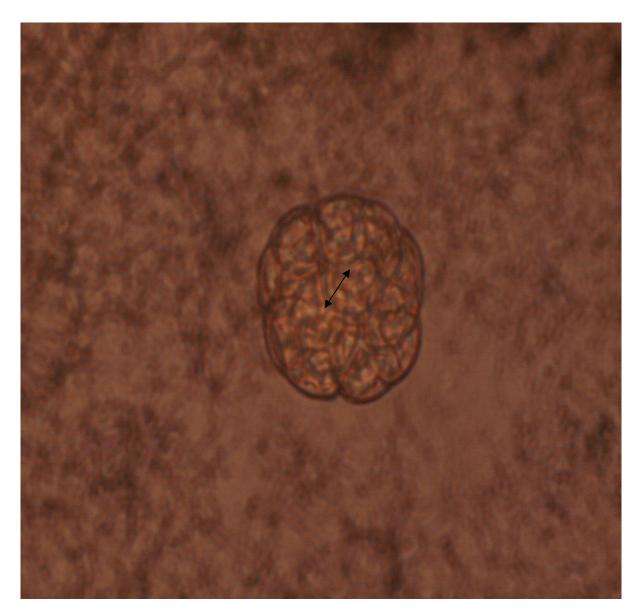


Plate 4.4. Pollen grains cluster in polyads of *Tetrapleura tetraptera* (x40) from NIHORT, Ibadan, Nigeria

| Variables | FRIN | NIHORT | T test | P – value |
|---|-------------------|-----------------|--------|---------------------|
| Number of flowers per inflorescence | 296.6 ± 37.30 | 294.2 ± 40.3 | 0.238 | 0.814 _{ns} |
| Number of inflorescence per twig | 16.3 ± 6.64 | 7.3 ± 3.2 | 8.514 | 0.000* |
| Length of inflorescence (cm) | 8.76 ± 1.59 | 9.31 ± 2.85 | -2.022 | 0.053 ^{ns} |

Table 4.6.Flowering structure and characteristics of *Tetrapleura tetraptera* atFRIN and NIHORT, Ibadan, Nigeria

*significant at P < 0.05; ns – not significant P > 0.05

4.2.4 Reproductive efficiency of *Tetrapleura tetraptera* trees from FRIN and NIHORT, Ibadan

There were no significant differences in the fruiting efficiency and number of matured fruits per inflorescence of *T. tetraptera* (Table 4.7). The fruiting efficiency per inflorescence at FRIN was higher (0.50 ± 0.21) than NIHORT (0.40 ± 0.11) . Similarly, the number of matured fruit per inflorescence at FRIN was higher (1.38 ± 0.53) than NIHORT (1.02 ± 0.29) (N=50). Fruit length and number of seeds were significantly higher at NIHORT than FRIN (Table 4.7). However, there was no significant difference in the fruit width which was higher at FRIN (5.62 ± 1.65 mm) than NIHORT (5.48 ± 0.53 mm).

The fruit set and abortion rate in *T. tetraptera* over the three fruiting cycles were similar in FRIN and NIHORT (Figure 4.11). The fruit set rate at FRIN in the first, second and third fruiting cycles were 11.1, 11.6 and 11.0% while NIHORT were 9.8, 12.3 and 11.2% respectively (Figure 4.11). The fruit abortion (drop) rate at FRIN was higher in the second cycle (98.6%) than first (96.6%) and third cycle (96.8%) (Figure 4.12). Similarly, fruit abortion rate at NIHORT was higher (99.4%) in the second cycle than first (97.6) and third cycle (97.9%) (Figure 4.12).

4.2.5 Distribution of insect visitors to *Tetrapleura tetraptera* trees at FRIN and NIHORT, Ibadan

A total of 912 insect visitors belonging to 15 species were recorded during the three days observation at the two locations (FRIN: 379; NIHORT: 533) (Appendix 8). Eight insect species were identified at FRIN while twelve species were identified at NIHORT. Hymenoptera was the most abundant insect order at FRIN (76%) and NIHORT (61%). Lepidoptera was the least abundant at FRIN (5%) while Coleoptera was the least abundant (1%) at NIHORT (Figure 4.13). The common insect visitors in the locations were *Apis mellifora*, *Bombus* sp., *Danaus chrysippus*, *Monomorium minimum*, *Orthetrum* sp., *Caliphora* sp. and *Sceliphron* sp. There was no significant difference in the periodic distribution of the insect visitors to *T. tetraptera* except for *Sceliphron sp.* and *Othetrum brachiale* (Appendix 9). Peak distributions of most insect visits were between 0700h to 1300h except *Sceliphron sp.* whose peak visit was 1500h (Figure 4.14). At FRIN, *Monomorium minimum* was the most abundant (6 ± 0.68) at each time period, followed by *Bombus* sp. (3 ± 1.41), *Vespus* sp. (3 ± 1.13) while the least abundant were *Apis* sp. (1 ± 1.3) and *Danaus chrysippus* (1 ± 0.77) (Figure 4.14).

| Variables | FRIN | NIHORT | T test | P – value |
|------------------|------------------|-----------------|--------|---------------------|
| Fruiting | 0.50 ± 0.21 | 0.40 ± 0.11 | -2.041 | 0.055 ^{ns} |
| efficiency | | | | |
| Number of fruits | 1.38 ± 0.53 | 1.02 ± 0.29 | 1.510 | 0.134 ^{ns} |
| per | | | | |
| inflorescence | | | | |
| Mature fruit | 17.01 ± 1.11 | 19.6 ± 1.65 | -4.719 | 0.000* |
| length (cm) | | | | |
| (df=19) | | | | |
| Mature fruit | 5.62 ± 1.65 | 5.48 ± 0.53 | 0.754 | 0.460 ^{ns} |
| width (cm) | | | | |
| (df=19) | | | | |
| Seeds per fruit | 12.5 ± 2.91 | 15.1 ± 2.9 | 2.850 | 0.01^{*} |

Table 4.7. Reproductive efficiency of *Tetrapleura tetraptera* at FRIN and NIHORT,Ibadan

*significant at P < 0.05; ns – not significant P > 0.05

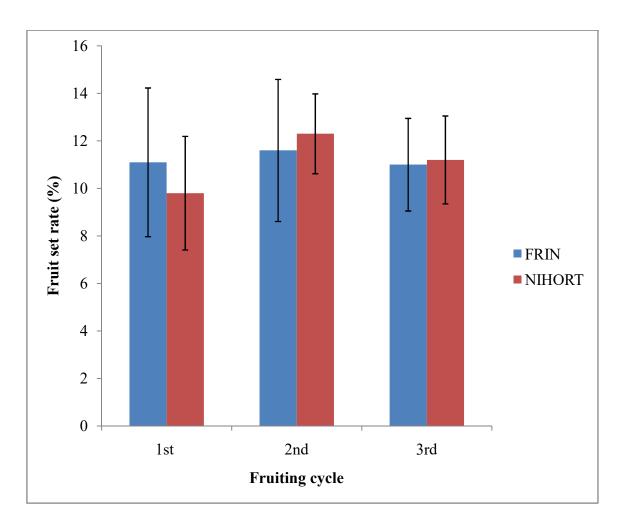


Figure 4.11. Fruit set rate (%) on inflorescence of *Tetrapleura tetraptera* (n=20, mean ±sd) at FRIN and NIHORT, Ibadan, Nigeria

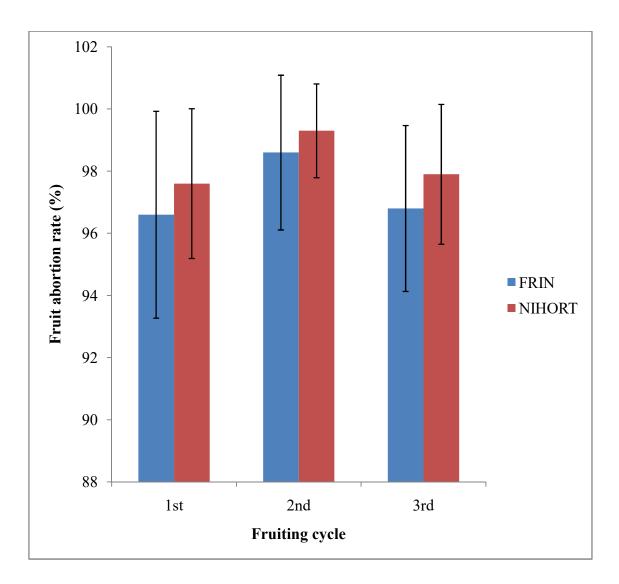
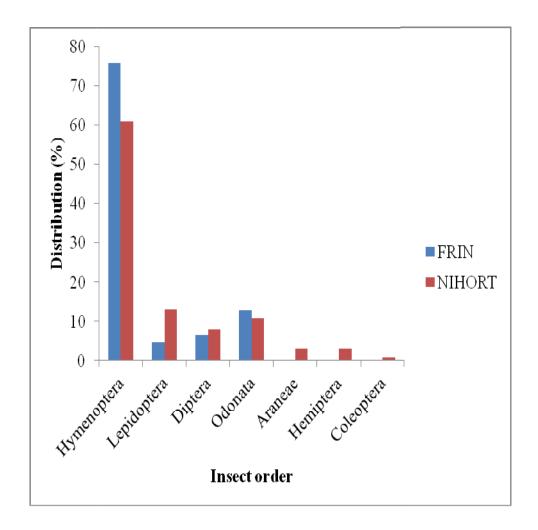
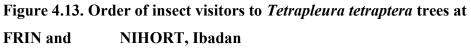


Figure 4.12. Fruit abortion rate (%) on inflorescence of *Tetrapleura tetraptera* (n=20, mean ±sd) at FRIN and NIHORT, Ibadan, Nigeria





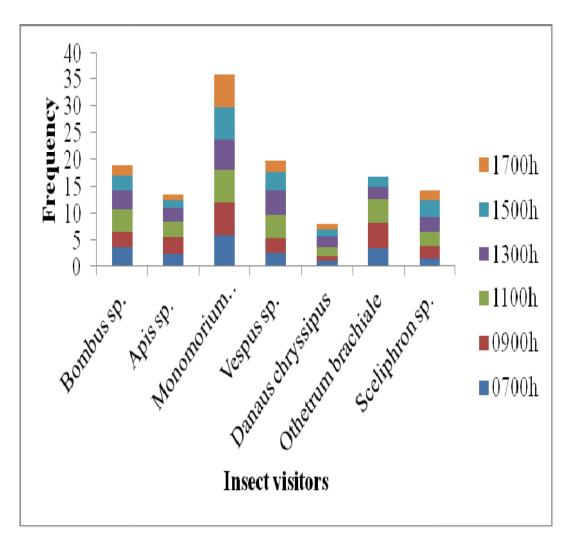


Figure 4.14. Frequency and period of visit of insect visitors to *Tetrapleura tetraptera* stands at FRIN in Ibadan

At NIHORT, *Monomorium minimum* had the highest visits (7 ± 2.34) , followed by *Othetrum brachiale* (4 ± 1.6) , *Bombus* sp. (3 ± 1.91) , *Sceliphron* sp. (3 ± 2.52) , while the least (1 ± 0.46) was *Brachypnoea* sp. (Figure 4.15). All insect visitors at NIHORT visited through out the observation period and showed peak visit at 1300h except for *Mylothris ocracea* and *Othetrum brachiale* that showed peak visit at 0900h (Figure 4.15).

4.2.6 Desciption of insect visitor's activities on *Tetrapleura tetraptera* trees

4.2.6.1 *Bombus* sp. (Bumble bee)

These insects landed on the inflorescence and inserted its mouth part on the dehisced anther to gather pollen, rubbing its abdominal segments on the pollen in the process. The abdomen presses against the stamens and the hair on the hind legs raked polyads from the anthers allowing the stigma to make contact with the anther repeatedly. They made quick and repeated visits between inflorescence intra and inter trees. *Bombus sp.* (bumble bee) made a buzzing sound by vibrating its wings while flying from one inflorescence to the other. The time spent per inflorescence was 31 ± 17.5 seconds (Table 4.8).

4.2.6.2 Apis sp. (Honey bees)

These insects moved round the inflorescence axis while feeding on the dehisced pollen and at the same time collecting pollen on its ventral region and legs. Foraging time on inflorescence was 116.4 ± 59 seconds (Table 4.8).

4.2.6.3 Sceliphron sp., Vespula vulgaris and Polistes belicosus (Wasps)

They exhibited similar foraging behaviour like the Bombus *sp.* by making frequent and inter-tree visits. Foraging time on inflorescence of *T. tetraptera* was highest (16.4 ± 4.72 seconds) in *Polistes belicosus*, followed by *Vespula vulgaris* (11.6 ± 6.5) while the lowest (10.6 ± 6.76) was *Sceliphron sp.* (Table 4.8).

4.2.6.4 *Calliphora* sp. (Fly)

They foraged on the inflorescence in the same way like *Apis* sp. and visited as many flowers as possible while making contact with the stigma. Their foraging time on flowers (inflorescence) was 122 ± 4.0 seconds (Table 4.8).

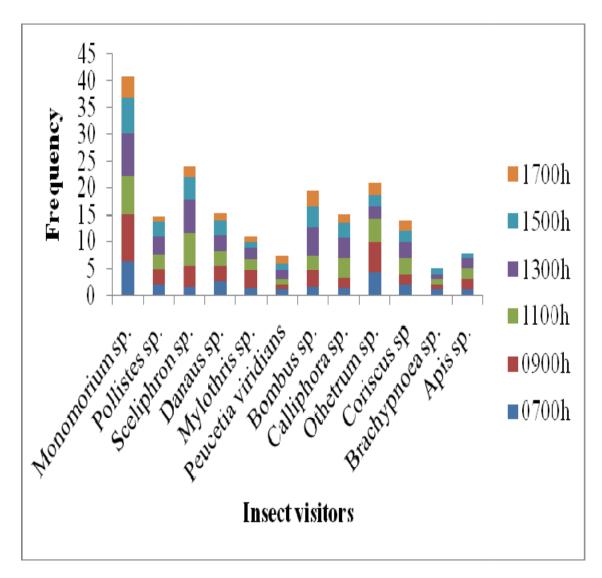


Figure 4.15. Frequency and period of visit of insect visitors to *Tetrapleura tetraptera* stands at NIHORT in Ibadan

| Order | Family | Species | Foraging time per inflorescence (s) |
|-------------|---------------|------------------------|--|
| Hymenoptera | Apidae | Bombus sp. | 31.00 ± 17.5 |
| | | Apis mellifera. | 116.4 ± 59.0 |
| | Sphecidae | Sceliphron sp | 10.60 ± 6.76 |
| | Vespidae | Polistes belicosus | 16.40 ± 4.72 |
| | | Vespula vulgaris | 11.60 ± 6.50 |
| | Formicidae | Monomorium minimum | $300.0\ \pm 0.00$ |
| Lepidoptera | Danaidae | Danaus chrysippus | 1.60 ± 0.55 |
| | Pieridae | Mylothris ocracea | $2.00\ \pm 0.71$ |
| Diptera | Calliphoridae | Caliphora sp. | 122.0 ± 4.00 |
| Odonata | Libelludae | Orthetrum brachiale | 1.60 ± 0.55 |
| Araneae | Oxyopidae | Peucetia viridians | 54.3 ± 8.50 |
| Hemiptera | Alydidae | Alydus eurinus | $13.60~\pm~1.15$ |
| Coleoptera | Chrysomelidae | Brachypnoea sp. | 204.8 ± 90.7 |

Table 4.8. Foraging time of insect visitors of *Tetrapleura tetraptera* at FRIN and NIHORT, Ibadan

4.2.6.5 *Monomorium mininimum* (Ants)

Ants foraged by moving from one flower to the other within an inflorescence while, having contact with the anthers and stigma during the process. They were resident within the inflorescence and therefore spent the highest (300 ± 0 seconds) foraging time on inflorescence of species.

4.2.6.6 Butterfly (*Danaus chrissipus* and *Mylothris ocracea*); dragon fly (*Othetrum brachiale*)

They moved frequently between inflorescences and made occassional contact with the anther. Foraging time of *Danaus chrissipus* on inflorescence was 1.6 ± 0.55 seconds while *Mylothris ocracea* 2 ± 0.71 seconds.

4.2.6.7 *Peucetia viridians* (green lynx spider)

They came to the inflorescence to prey on other insect visitors (e.g wasp). The time spent on foraging was 54.3 ± 8.5 seconds.

4.2.6.8 Alydus eurinus (bug) and Brachypnoea sp. (leaf beetles)

The bugs and leaf beetles moved within the inflorescence to feed on the petals and leaves. Alydus eurinus spent 13.6 ± 1.15 seconds on inflorescence while Brachypnoea sp. spent 204.8 ± 90.7 seconds.

4.2.7 Pollen load on insect visitors to *Tetrapleura tetraptera* trees at FRIN and NIHORT, Ibadan

The number of polyads on insect bodies varied from one insect visitor to another (Table 4.9). A total of thirteen insect visitors were caught while ten were confirmed for the presence of pollen grains that matched the description of *T. tetraptera* polyads. Insect species belonging to hymenoptera accounted for majority of polyads carrying insects recorded (90%). The highest number of polyads was found on *Bombus* sp. (28%) followed by *Apis mellifera* (16%) while the lowest was recorded on *Danaus chrissipus* with frequency of 1%.

| Order | Family | Species | Pollen load (%) |
|-------------|---------------|------------------------|-----------------|
| Hymenoptera | Apidae | Bombus sp. | 25 |
| | | Apis mellifera. | 14 |
| | Sphecidae | Sceliphron sp | 17 |
| | Vespidae | Polistes belicosus | 9 |
| | | Vespula vulgaris | 15 |
| | Formicidae | Monomorium minimum | 2 |
| Lepidoptera | Danaidae | Danaus chrysippus | 1 |
| | Pieridae | Mylothris ocracea | 0 |
| Diptera | Calliphoridae | Caliphora sp. | 12 |
| Odonata | Libelludae | Orthetrum brachiale | 0 |
| Araneae | Oxyopidae | Peucetia viridians | 0 |
| Hemiptera | Alydidae | Alydus eurinus | 2 |
| Coleoptera | Chrysomelidae | <i>Brachypnoea</i> sp. | 2 |

Table 4.9.Pollen load of insect visitors of *Tetrapleura tetraptera* at FRIN andNIHORT (Ibadan)

4.3 Morphological traits of fruits and seeds of *Tetrapleura tetraptera* from four sources in lowland rainforest zone of Nigeria

4.3.1 Weight of *Tetrapleura tetraptera* seeds from four different sources

Seed sources had a significant effect on the weight of 100-seeds of *T. tetraptera* (Appendix 11). Aponmu source had the highest $(16.16 \pm 0.08 \text{ g})$ seed weight, while the lowest $(10.9 \pm 0.28 \text{ g})$ was recorded in Mamu source (Table 4.10). The coefficient of variations within seed source for seed weight ranged from 0.51 to 1.57 % while intersource was 16.1% (Table 4.11).

4.3.2 Number of seeds per fruit in *Tetrapleura tetraptera* from four different sources

Number of seeds per fruit differed significantly among the four sources (Appendix 11). Ibadan and Mamu source did not differ significantly from one another but differed significantly from Iwo and Aponmu sources, which did not differ from each other. Aponmu source had the highest number of seeds per fruit (15.6 ± 2.44), followed by Iwo (15.2 ± 3.14) while Mamu and Ibadan had 10.9 ± 3.79 and 10.8 ± 3.94 respectively (Table 4.10). The highest coefficient of variation (30.55) among fruit traits was found in number of seeds per fruit (Table 4.11).

4.3.3 Fruit length in *Tetrapleura tetraptera* from four different sources

There were significant differences in fruit length of *T. tetraptera* from different sources (Appendix 11). Mamu source had the highest fruit length $(20.9 \pm 2.47 \text{ cm})$, followed by Aponmu $(20 \pm 1.89 \text{ cm})$, Iwo $(18.6 \pm 2.31 \text{ cm})$ while the lowest $(16.7 \pm 1.48 \text{ cm})$ was recorded in Ibadan (Table 4.10). The coefficient of variation among the sources was higher than within-source (Table 4.11).

4.3.4 Fruit width in *Tetrapleura tetraptera* from four different sources

Ibadan source had the highest fruit width (52.4 ± 5.99 mm), followed by Aponmu (48.1 ± 4.66 mm); Mamu (47.8 ± 4.33) while lowest value (37.2 ± 5.06 mm) was recorded in Iwo (Table 4.10). There were significant differences in the fruit width of *T. tetraptera* from the four sources (Appendix 11). The width of fruits from Aponmu (Ondo) was not

significantly different from that of Mamu (Ogun) while Ibadan (Oyo) was significantly different from Mamu, Aponmu and Iwo fruit widths (Table 4.10). The coefficient of variation for fruit width inter-source was higher than within-source (Table 4.11).

4.3.5 Correlation of *Tetarapleura tetraptera* fruit traits and geo-climatic variables

There was a positive significant strong correlation between seed weight and number of seeds per fruit (0.956) (Appendix 12). However, fruit length, width and number of seeds were not correlated. Seed weight was positively correlated with minimum altitude (0.966) and maximum altitude (0.997) of seed sources. Number of seeds was also significantly correlated with maximum altitude (0.965). There was no significant correlation between geo-climatic variables and fruit dimensions (length and width) (Appendix 12).

| Fruit Sources | Seed weight (g) | Fruit length (cm) | Fruit width (mm) | Seeds per fruit |
|------------------|----------------------|-------------------------|---------------------|----------------------|
| Aponmu (Ondo) | $16.16_{a}\pm 0.08$ | $20.0_a \pm 1.89$ | $48.1_b\pm4.66$ | $15.56_a \pm 2.44$ |
| Ibadan (Oyo) | $12.47_{b} \pm 0.20$ | $16.7_{b} \pm 1.48$ | $52.4_a \pm 5.99$ | $10.81_b \pm 3.94$ |
| Iwo (Osun) | $15.31_{c} \pm 0.19$ | $18.6_{\rm c} \pm 2.31$ | $37.2_{c} \pm 5.06$ | $15.16_{a} \pm 3.14$ |
| Mamu (Ogun) | $10.94_{d} \pm 0.28$ | $20.9_d\pm2.47$ | $47.8_{b} \pm 4.33$ | $10.98_{b} \pm 3.79$ |

Table 4.10Morphological traits of *Tetrapleura tetraptera* fruits from four sourcesin southwest Nigeria (n=150, mean \pm sd)

*Mean with same alphabet is not significantly different at P > 0.05

| Source | Seed weight (g) | Fruit length (cm) | Fruit width (mm) | No. of seeds |
|--------------|-----------------|----------------------|---------------------|--------------|
| Aponmu | 0.51 | 9.42 | 9.69 | 15.71 |
| Ibadan | 1.57 | 8.86 | 11.42 | 36.47 |
| Iwo | 1.22 | 12.41 | 13.58 | 20.71 |
| Mamu | 2.57 | 11.82 | 9.05 | 34.56 |
| Inter-Source | 16.09 | 13.71 | 16.23 | 30.55 |

Table 4.11. Intra and inter-source coefficient of variation (%) of fruit/seedmorphological traits of *Tetrapleura tetraptera* from four sources

4.4 Effect of seed sources on early growth of *Tetrapleura tetraptera* seedlings from four seed sources

4.4.1 Seedling height

Seedling height increased over time in the experiment across the four sources (Figure 4.16a). Seed source and age of seedlings significantly affected the seedling height (Appendix 13). Ibadan (Oyo) had the highest height ($61.8 \pm 12.6 \text{ cm}$) followed by Iwo ($61.6 \pm 14.0 \text{ cm}$); Aponmu ($60 \pm 13.2 \text{ cm}$) while the lowest height ($53.0 \pm 17.8 \text{ cm}$) was obtained in Mamu (Figure 4.16a). However, interaction effects of sources and time of experiment had no significant influence on seedling height (Appendix 13). The coefficient of variation within source was highest in Mamu (33.5%) followed by Iwo (25.5%), Aponmu (21.9%) while the lowest (20.4%) was Ibadan (Table 4.12).

4.4.2 Seedling collar diameter

Seedling collar diameter increased over the age of seedlings across the sources (Figure 4.16b). However, seed sources had no significant effect on collar diameter (Appendix 14). The interaction effect between source and seedling's age did not differ significantly. Aponmu had the highest collar diameter $(4.3 \pm 0.82 \text{ mm})$, followed by Mamu $(4.2 \pm 1.35 \text{ mm})$, Ibadan $(4.1 \pm 1.03 \text{ mm})$ and Iwo $(4.1 \pm 0.78 \text{ mm})$ (Figure 4.16b). Coefficient of variability was highest in Mamu (32.5%) followed by Ibadan (25.2%), Aponmu (19.3%) while the lowest (18.9%) was Iwo (Table 4.12).

4.4.3 Number of leaves of *Tetrapleura tetraptera* seedlings

Seed source and assessment time significantly influenced the number of leaves produced (Appendix 15). The interaction effects between the seed sources and seedling's age were also significantly different. Aponmu produced the highest number of leaves (16.3 ± 3.08) , followed by Iwo (14.8 ± 3.11) , Mamu (14.7 ± 3.22) , while Ibadan had the lowest (14.1 ± 2.86) (Figure 4.16c). The Coefficient of variability was highest within Mamu (22%), followed by Iwo (21%), Ibadan (20.4%) while the lowest (18.9%) was Aponmu (Table 4.12). Number of leaves was positively correlated with seedling height (0.740) and collar diameter (0.848) (Appendix 16).

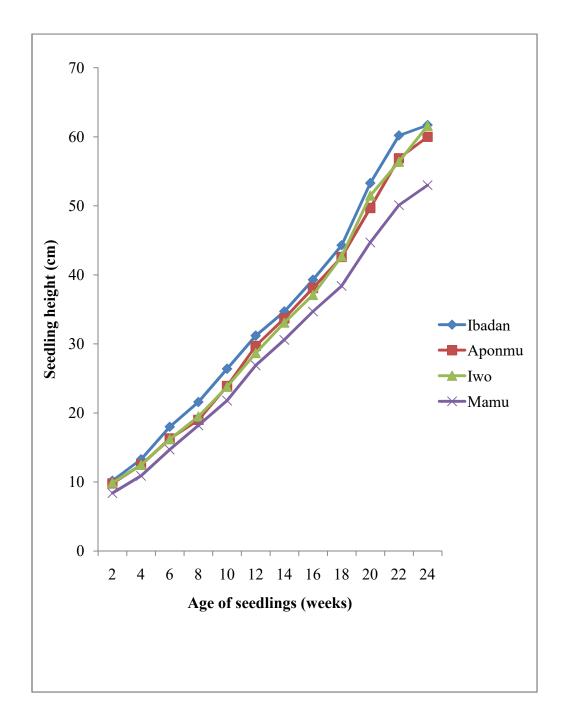


Figure 4.16a. Effect of seed sources on height (cm) of *Tetrapleura tetraptera* seedlings

| Source | Height (cm) | Collar diameter (mm) | Leaves |
|--------|-------------|-------------------------|--------|
| Aponmu | 21.9 | 19.3 | 18.9 |
| Ibadan | 20.4 | 25.2 | 20.4 |
| Iwo | 25.5 | 18.9 | 21.0 |
| Mamu | 33.5 | 32.5 | 22.0 |
| Source | 22.8 | 24.4 | 21.1 |

 Table 4.12. Coefficient of variation (%) for Tetrapleura tetraptera seedling character

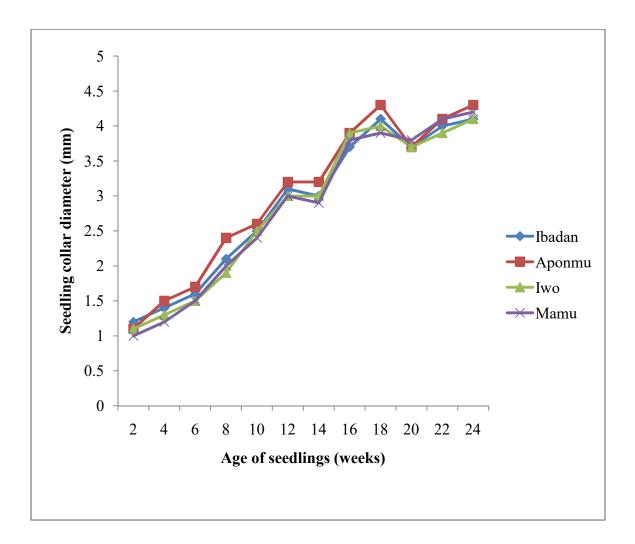


Figure 4.16b. Effect of seed sources on collar diameter (mm) of *Tetrapleura tetraptera* seedlings

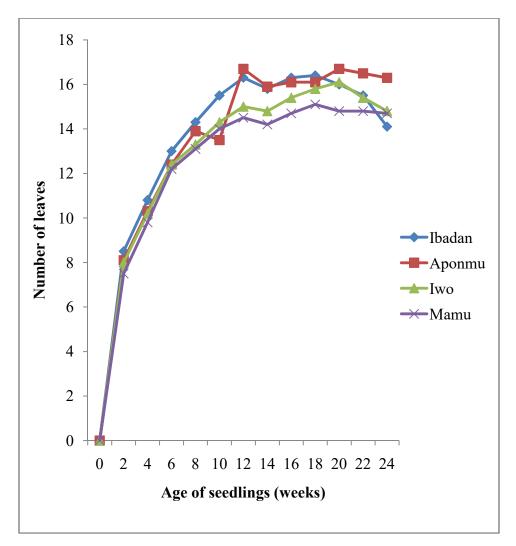


Figure 4.16c. Effect of seed sources on number of leaves of *Tetrapleura tetraptera* seedlings

4.4.4 Root dry weight of *Tetrapleura tetraptera* seedlings

The root dry weight of *T. tetraptera* seedlings from different sources increased with age of seedlings (Figure 4.17a). Seed sources and interaction between age of seedlings and source had no significant effect on root dry weight of *T. tetraptera* (Appendix 17). At the end of 24^{th} week of assessment, seedlings from Ibadan had the highest root dry weight (8.41 ± 6.67g) followed by Mamu (6.74 ± 3.46g), Iwo (6.36 ± 0.94g), while the lowest (5.13 ± 1.92g) was obtained from Aponmu (Figure 4.17a).

4.4.5 Shoot dry weight

Seed sources had no significant effect on the shoot dry weight while age of seedlings did (Appendix 18). Interactions between source and seedling's age did not show significant effect. The shoot dry weight increased with age of seedlings (Figure 4.17b). Aponmu had the highest shoot dry weight ($16.38 \pm 3.6g$), followed by Ibadan ($15.24 \pm 4.0g$), Mamu ($14.83 \pm 5.5g$) while the lowest ($13.68 \pm 2.5g$) was recorded in Iwo.

4.4.6 Biomass accumulation

Seedlings from Ibadan had the highest total biomass $(26.24 \pm 10.17g)$, followed by Mamu $(23.12 \pm 5.88g)$, Aponmu $(21.79 \pm 5.11g)$ while Iwo had the least $(20.3 \pm 3.5g)$ (Figure 4.17c). Seed sources had no significant effect on the biomass but age of seedlings had (Appendix 19). The interaction between source and harvest period was also not significant. The coefficient of variation (CV) within Ibadan was higher (38.8%) than overall source variation (27.8%) while the lowest (17.2%) was observed in Iwo (Appendix 21).

4.4.7 Relative growth rate (RGR) of seedlings

The relative growth rate (RGR) of *T. tetraptera* seedlings decreased with age of seedlings across the seed sources (Figure 4.18a). Mamu source had the highest initial RGR (2.41 g/month) at 2 to 4 months while Aponmu recorded the lowest initial RGR (1.88 g/month). Mamu recorded the highest value (0.86 g/month) at 12 months when the experiment was terminated, followed by Ibadan (0.84 g/month), Iwo (0.79 g/month) while Aponmu had the least (0.71 g/month).

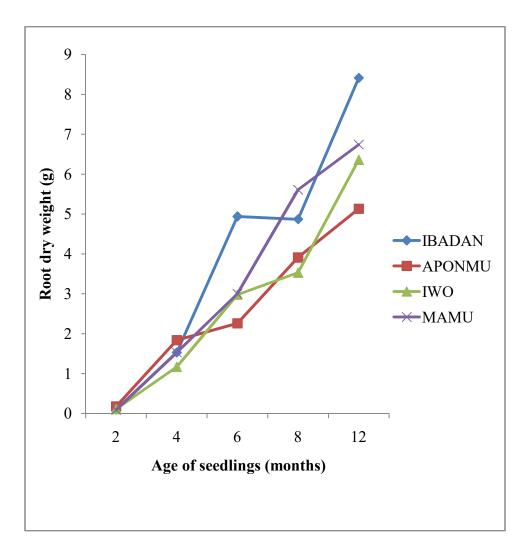


Figure 4.17a. Root dry weight (g) of *Tetrapleura tetraptera* seedlings from four seed sources

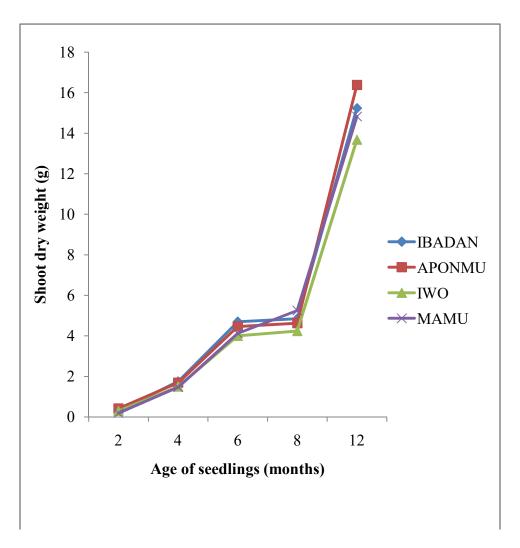


Figure 4.17b. Shoot dry weight (g) of *Tetrapleura tetraptera* seedlings from four seed sources

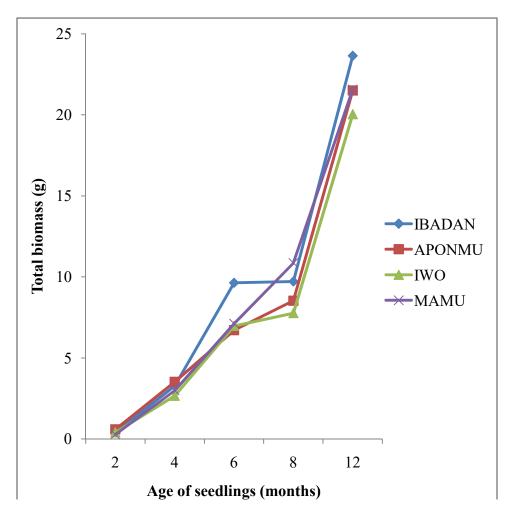


Figure 4.17c: Total biomass (g) of *Tetrapleura tetraptera* seedlings from four seed sources

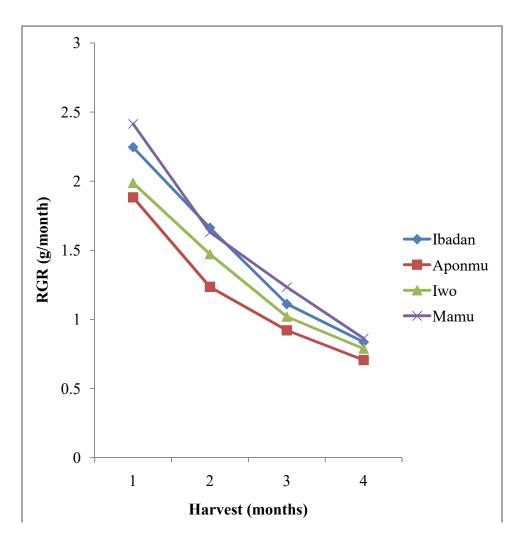


Figure 4.18a. Seedling's relative growth rate (RGR) from four seed sources in *Tetrapleura tetraptera*

4.4.8 Absolute growth rate (AGR) of seedlings

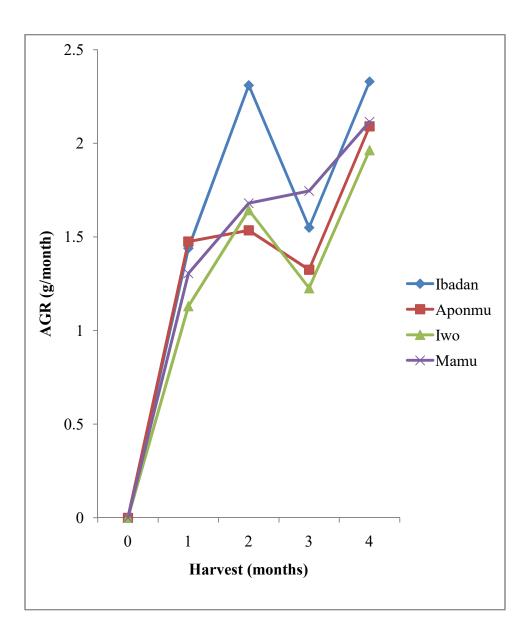
Aponmu had the highest initial AGR (1.48 g/month) at 4 months after germination, followed by Ibadan (1.44 g/month), Iwo (1.36 g/month) while the lowest (1.12 g/month) was Mamu (Figure 4.18b). The values of AGR were higher in the second harvest than the first while the fourth was higher than the third across the sources. The highest value (3.48 g/month) was obtained from Ibadan source at 12 months after germination while Mamu had the least (2.68 g/month).

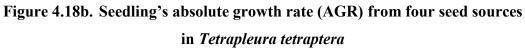
4.4.9 Net assimilation rate (NAR) in *Tetrapleura tetraptera* seedlings

Aponmu had the highest initial NAR (2.5×10^{-3} g/month) at 2 to 4 months while Iwo had the lowest (2.1×10^{-3} g/month) (Table 4.13a). The highest final NAR (3.7×10^{-3} g/month) was obtained from Ibadan, Iwo and Mamu had 3.6×10^{-3} g/month respectively while the least was Aponmu (3.4×10^{-3} g/month).

4.4.10 Seedling's Leaf area (cm²) in *Tetrapleura tetraptera*

Seed sources had significant differences on leaf area in the species (Appendix 24). Ibadan had the highest leaf area (726 ± 163.5 cm²), followed by Aponmu (686.4 ± 187.7 cm²), Mamu (686 ± 202 cm²) while the lowest (567.0 ± 132 cm²) was Iwo (Table 4.13b).





| Age interval (month) | | | | | |
|----------------------|------------------------|------------------------|------------------------|------------------------|--|
| Sources | 2 – 4 | 4 - 6 | 6 - 8 | 8-12 | |
| Aponmu | 2.5 x 10 ⁻³ | 2.3 x 10 ⁻³ | 2.4 x 10 ⁻³ | 3.4 x 10 ⁻³ | |
| Ibadan | 2.3 x 10 ⁻³ | 3.7 x 10 ⁻³ | 2.9 x 10 ⁻³ | 3.7 x 10 ⁻³ | |
| Iwo | 2.1 x 10 ⁻³ | 2.8×10^{-3} | 2.5 x 10 ⁻³ | $3.6 \ge 10^{-3}$ | |
| Mamu | 2.4 x 10 ⁻³ | 3.0 x 10 ⁻³ | 3.2 x 10 ⁻³ | 3.6 x 10 ⁻³ | |

Table 4.13a: Net assimilation rate (g/cm²/month) of *T. tetraptera* seedlings

Table 4.13b: Leaf area (cm²) of *T. tetraptera* seedlings (mean \pm sd)

| 4 6 3 ±111 792.0 ±1 | 8 14 567.6 | 12 686.4±187.9 |
|-------------------------------|----------------------|--------------------------|
| 3 ±111 792.0 ±1 | 14 567.6 | 686.4±187.9 |
| | | |
| | ±179.7 | |
| l ±59.7 699.6 | 5 554.4 ±52.8 | 3 726.0±163.5 |
| ±108.8 | 8 | |
| l ±91.2 660.0 |) 488.4 ±50.6 | 567.0±132 |
| ±109.9 | 9 | |
| 91.4 620.4 | t 567.6 ±99.9 | 686.0±202 |
| | 4 | |
| | ±109. 01.4 620.4 | ±109.9 |

4.5 Effect of concentration of growth regulators on rooting of *Tetrapleura tetraptera* stem cuttings

4.5.1 Survival of stem cuttings

There were no significant differences in the main and interaction effects of growth regulators and their concentration on cuttings survival (Appendix 26). Stem cuttings treated with IBA + NAA had the highest survival rates (83.3 ±15.1%), while the least survived ($66.7 \pm 24.2\%$) was recorded for those treated with NAA (Table 4.14). however, 80 % survival was recorded in cuttings treated with 0, 100 and 150 ppm while the lowest ($70 \pm 27.6\%$) was recorded at 200 ppm (Table 4.14).

4.5.2 Root sprout per stem cutting

Stem cuttings treated with coconut water had the highest number of roots (5 ± 2.65) followed by cuttings treated with IBA+ NAA (4.04 ± 1.78) while the lowest was recorded for those treated with NAA (3.33 ± 2.65) and IBA (3.33 ± 1.76) (Plates 4.5 and 4.6). There were no significant differences in the main and interaction effects of growth regulators and their concentrations on the number of roots per stem cutting (Appendix 25). Auxin application to stem cuttings at 150 ppm had the highest number of roots (4.04 ± 2.1) followed by 100 ppm (3.9 ± 1.82), while the lowest (2.75 ± 2.21) was 200 ppm (Table 4.14).

4.5.3 Stem cuttings root length

There were significant differences in the main and interaction effect of growth regulators and their concentrations on root length per cutting (Appendix 25). Stem cuttings treated with coconut water had the highest total root length (5.02 ± 1.61 cm) followed by the control (4.56 ± 2.23 cm), while the lowest (2.48 ± 1.61 cm) was recorded for those treated with NAA (Table 4.14). Control was significantly different from NAA and IBA+NAA while IBA was not (Table 4.14). IBA differed significantly from NAA while IBA+NAA did not. Coconut water differed significantly from IBA, NAA and IBA+NAA while control did not. In terms of concentration, control (0 ppm) had the highest effect on root length (4.56 ± 2.23 cm), followed by auxin applications at 150 ppm (3.96 ± 1.52)) while the least (2.6 ± 0.98) was 100 ppm. Auxin applied at 100 ppm was significantly different from 150 ppm while 200 ppm was not (Table 4.14).

| Growth regulators | Survival (%) | No of roots | Root length (cm) |
|------------------------|-----------------|-----------------|-----------------------|
| IBA | $80.0\pm\ 0.00$ | 3.33 ± 1.76 | $3.77^{abc} \pm 1.45$ |
| NAA | 66.7 ± 24.2 | 3.33 ± 2.65 | $2.48^{b} \pm 1.61$ |
| IBA+NAA | 83.3 ± 15.1 | 4.04 ± 1.78 | $3.34^{b}\pm1.50$ |
| CONTROL | 80.0 ± 21.9 | 3.87 ± 2.82 | $4.56^{cd}\pm2.27$ |
| COCONUT | 70.0 ± 10.9 | 5.00 ± 2.65 | $5.02^{d} \pm 1.61$ |
| WATER | | | |
| Concentration (PPM) | | | |
| 0 | 80.0 ± 21.9 | 3.87 ± 2.82 | $4.56^{b} \pm 2.27$ |
| 100 | 80.0 ± 0.00 | 3.92 ± 1.82 | $2.60^{a} \pm 0.98$ |
| 150 | 80.0 ± 12.7 | 4.04 ± 2.10 | $3.96^{b} \pm 1.52$ |
| 200 | 70.0 ± 27.6 | 2.75 ± 2.21 | $3.04^{ab}\pm1.90$ |
| | | | |

Table 4.14. Effect of growth regulators and their concentration on juvenile stem cuttings of *Tetrapleura tetraptera* (mean \pm sd)

There was no significant difference between auxin application at 150 ppm and 200 ppm on the length of root of *T. tetraptera*.

There were significant differences in the effect of growth regulators on the length of longest root per stem cuttings (Appendix 25). However, there were no significant differences in the concentrations of auxins. Interaction effects of growth regulators and their concentrations on the total root length was also not significant (Appendix 25). Control had the highest total root length root per cutting (6.84 ± 4.12 cm) followed by coconut water (6.8 ± 2.19 cm), IBA + NAA (4.75 ± 2.44 cm) while least (2.65 ± 2.14 cm) was recorded for NAA treated cuttings (Table 4.15). Stem cuttings treated with NAA was significantly different from other growth regulators. Stem cuttings treated with IBA and IBA + NAA were not significantly different but differed significantly from coconut water and control.

4.5.4 Number of shoots and leaves per stem cutting

There were no significant differences in the effect of growth regulators on the number of shoots per cutting (Appendix 25). The interaction of growth regulators and concentrations did not differ significantly. Naphthalene Acetic Acid (NAA) had the highest (2.12 ±1.58) number of shoots while the least (1.62 ± 1.25) was for control (Table 4.15). However, their concentrations significantly affected shoot emergence (Table 4.16). Auxin application at 150 ppm did not differ significantly from 100 ppm but differ significantly from 200 ppm (Table 4.16). There were significant differences in the effect of growth regulators on leaf production (Appendix 26). The interaction effect of growth regulators and their concentration was also significant (Appendix 30). Stem cuttings treated with IBA had the highest (3.83 ± 0.99) number of leaves while NAA had the lowest (2.65 ± 1.69) (Table 4.15). Stem cuttings treated with coconut water, control, IBA and IBA + NAA, was not significantly different from one another. However, NAA was significantly different from IBA, coconut water and control treatment (Table 4.15).

4.5.5 Shoot length per cuttings

Control treatment had the highest effect on shoot length (7.28 \pm 4.4cm), followed by coconut water (6.69 \pm 2.01cm), IBA (6.21 \pm 2.63cm) while the least (4.67 \pm 3.56cm) was NAA (Table 4.15). The longest shoot (7.28 \pm 4.40) was obtained from 0 ppm followed by 150 ppm (6.23 \pm 2.52cm), 100 ppm (5.42 \pm 2.59cm), while 200 ppm had the lowest (5.11 \pm 3.67cm).

There were no significant differences in the effect of growth regulators and concentrations on the length of shoot (Appendix 26). However, the interaction effects of growth regulators and concentrations were significantly different.

| Treatment | Length of longest root (cm) | No. of shoots | Shoot length (cm) | No. of leaves |
|------------------------|-----------------------------------|------------------|----------------------|---------------------|
| IBA | 4.70 ^a ± 1.91 | 1.87 ± 0.99 | 6.21 ± 2.63 | $3.83^{a} \pm 0.99$ |
| NAA | 2.65 ^b ± 2.14 | 2.12 ± 1.58 | 4.67 ± 3.56 | $2.65^{b} \pm 1.69$ |
| IBA + NAA | 4.75 ^a ± 2.44 | 1.87 ± 1.04 | 5.93 ± 2.52 | $3.41^{b} \pm 1.23$ |
| COCONUT WATER | 6.80 ^c ± 2.19 | 1.87 ± 1.30 | 6.69 ± 2.01 | $3.51^{a} \pm 1.02$ |
| CONTROL | 6.84 ^c ± 4.12 | 1.62 ±1.25 | 7.28 ± 4.40 | $3.51^{ab}\pm1.02$ |
| Concentration (PPM) | | | | |
| 0 | 6.84 ± 4.12 | 1.62 ± 1.25 | 7.28 ± 4.40 | 3.51 ± 1.02 |
| 100 | 3.69 ± 2.12 | 2.07 ± 0.94 | 5.42 ± 2.60 | 3.52 ± 1.09 |
| 150 | 4.77 ± 2.46 | 2.44 ± 1.42 | 6.23 ± 2.52 | 3.55 ± 1.19 |
| 200 | 3.64 ± 2.39 | 1.37 ± 1.03 | 5.11 ± 3.67 | 2.82 ± 1.76 |

 Table 4.15: Effect of growth regulators and concentration on juvenile stem cuttings of *Tetrapleura tetraptera*

| No. of shoots per cutting |
|---------------------------|
| $1.62_{a}\pm 1.25$ |
| $2.07_{ab}\pm\!0.94$ |
| $2.44_{b}\pm 1.42$ |
| $1.37_{a}\pm 1.03$ |
| |

Table 4.16. Effect of growth regulator concentrations on stem cuttings shoots inTetrapleura tetraptera (mean \pm sd)

Note: mean with same letters are not significantly different







- a. IBA at 100 PPM
- b. IBA at 150 PPM
- c. IBA at 200 PPM

I. IBA



a. NAA at 100 PPM

b. NAA at 150 PPM



c. NAA at 200 PPM

- II. NAA
- Plate 4.5 Effect of growth regulators (IBA and NAA) and their concentrations on stem cuttings rooting in *Tetrapleura tetraptera*



a. IBA+NAA100 ppm

b. IBA+NAA150 ppm



c. IBA+NAA 200 ppm



d. Coconut water at 25% dilution



e. Control (no hormone)

Plate 4.6. Effect of growth regulators (combined IBA and NAA), coconut water and their concentration on stem cuttings rooting in *Tetrapleura tetraptera*

4.6 Effect of seedling's source and cutting (nodal) positions on survival and rooting of *Tetrapleura tetraptera*

4.6.1 Survival of stem cuttings from different sources and nodal positions

There were significant differences in the main and interaction effect of sources and nodal position on survival of stem cuttings (Appendix 27). Cuttings from Mamu seedlings had the highest $(38.2 \pm 29.1\%)$ followed by Aponmu $(27.6 \pm 33.1\%)$, Iwo $(18 \pm 28.8\%)$ while the lowest $(12.4 \pm 22.95\%)$ was recorded for Ibadan (Figure 4.19). Stem cuttings obtained from the basal position of the stem had the highest survival rate $(56.3 \pm 29.1\%)$, followed by the middle position $(14 \pm 21.5\%)$ while the lowest $(1.8 \pm 7.3\%)$ was obtained from the apical nodal position (Figure 4.20). There was positive strong correlation between cutting's survival and growth variables of seedlings (Appendix 29).

4.6.2 Root sprout per stem cutting

There were significant differences in the main and interaction effect of sources and nodal positions on the number of emerged roots on juvenile stem cuttings of *T. tetraptera* (Appendix 27). The highest (1.52 ± 2.32) number of roots was recorded for cuttings obtained from Mamu seedlings, followed by Aponmu (0.72 ± 1.45) , Iwo $(0.58 \pm 1.38\%)$ while the lowest $(0.41 \pm 1.22\%)$ was recorded for Ibadan source (Figure 4.21). There were no significant differences between stem cuttings obtained from Iwo and Ibadan seedlings while Aponmu and Ibadan differed (Table 4.12). Mamu seedlings differed significantly from Aponmu, Iwo and Ibadan source but Aponmu and Iwo seedlings did not. Cutting positions on the stem had significant effect on the number of emerged roots (Plate 4.7). Number of roots (2.19 \pm 2.28) was highest in stem cuttings obtained from the basal position followed by the middle (0.20 ± 0.69), while the lowest (0.04 ± 0.3) was for the apical region (Figure 4.22).

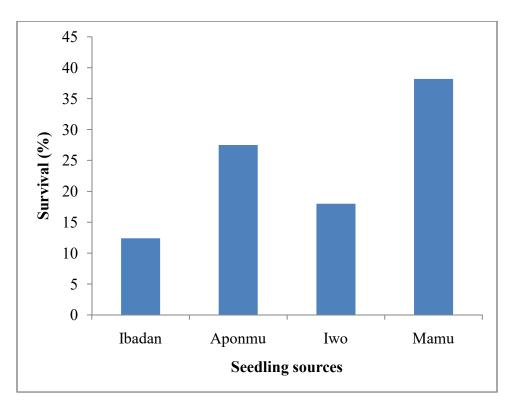


Figure 4.19. Effect of seedling sources on survival (%) of juvenile stem cuttings of *Tetrapleura tetraptera*

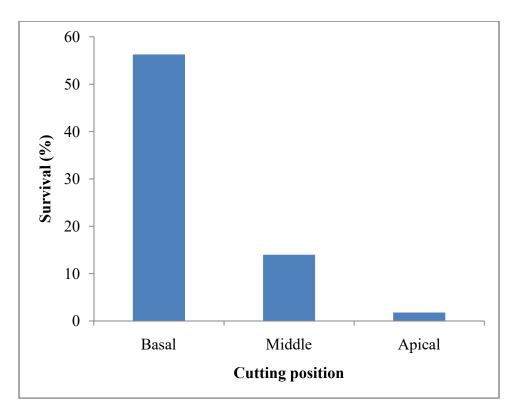


Figure 4.20. Effect of cutting (nodal) positions on survival (%) of juvenile stem cuttings of *Tetrapleura tetraptera*

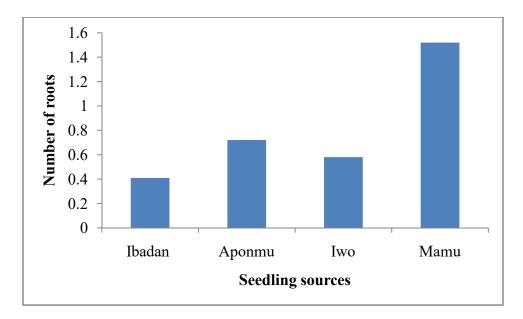


Figure 4.21. Number of roots per stem cutting of *Tetrapleura tetraptera* from four seedling sources

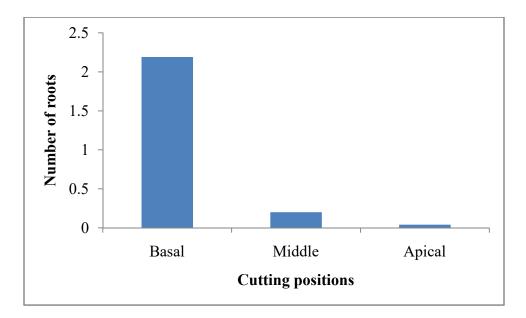


Figure 4.22. Number of roots per stem cutting of *Tetrapleura tetraptera* from three nodal positions



a. Apical nodal cuttings b. Middle nodal cuttings c. Basal nodal cuttings

Plate 4.7: Effect of cutting positions on rooting of *Tetrapleura tetraptera* stem cuttings

4.6.3 Root length per stem cutting

There were significant differences in the main and interaction effect of sources and cutting positions on root length per cutting (Appendix 27). The highest length of root $(1.56 \pm 2.16 \text{ cm})$ was recorded for Mamu seedlings followed by Aponmu $(1.17 \pm 2.22 \text{ cm})$, Iwo $(0.87 \pm 1.86 \text{ cm})$ while the least $(0.59 \pm 1.91 \text{ cm})$ was recorded for Ibadan (Table 4.17). Stem cuttings from the basal position had the longest root length $(2.76 \pm 2.63 \text{ cm})$, followed by the middle cutting position $(0.34 \pm 1.19 \text{ cm})$, and while the least $(0.03 \pm 0.29 \text{ cm})$ were obtained from the apical cutting positions. There were also significant differences in the main and interaction effects of source and cutting positions on the length of longest root per cutting (Appendix 27). Mamu had the highest $(2.05 \pm 3.04 \text{ cm})$ followed by Aponmu source $(1.25 \pm 2.49 \text{ cm})$, Iwo $(1.08 \pm 2.42 \text{ cm})$ while the lowest $(0.77 \pm 2.29 \text{ cm})$ was recorded for Ibadan source (Table 4.17). Stem cuttings from Mamu seedlings differed significantly from Aponmu, Iwo and Ibadan while Iwo and Aponmu cuttings did not differ (Table 4.17).

Cutting positions had significant effect on the length of longest root per cutting (Appendix 27). Stem cuttings from the base had the longest $(3.46 \pm 3.43 \text{ cm})$ followed by middle $(0.36 \pm 1.25 \text{ cm})$, while the apical position had the lowest $(0.03 \pm 0.28 \text{ cm})$ (Table 4.17). The interaction between cutting sources and positions on length of longest root per cutting was significant (Appendix 31). The apical, median and basal cutting positions on the seedlings were significantly different from one another (Table 4.17).

4.6.4 Number of shoots per stem cutting

There were significant differences in the main and interaction effects of seedling's source and cutting positions on the number of shoots per stem cutting of *T. tetraptera* (Appendix 28). Mamu had the highest (0.79 ± 1.11) , followed by Aponmu (0.58 ± 1.11) while the lowest (0.26 ± 0.74) was Ibadan (Table 4.18). Similarly, stem cuttings from the apical, middle and basal position on the seedlings were significantly different from one another (Table 4.18). Basal position had the highest (1.38 ± 1.47) number of shoots followed by middle position (0.18 ± 0.49) while the lowest (0.04 ± 0.28) were top positions on the stockplant (Table 4.18).

| | Root length (cm) | Length of longest root |
|-------------------|-------------------------|----------------------------|
| | | (cm) |
| Sources | | |
| Ibadan | $0.59^{a}\pm1.91$ | $0.77^{\mathrm{a}}\pm2.29$ |
| Aponmu | $1.17^{b} \pm 2.21$ | $1.25^{b} \pm 2.49$ |
| Iwo | $0.87^{\rm c}\pm1.86$ | $1.08^{b} \pm 2.42$ |
| Mamu | $1.52^{d} \pm 2.32$ | $2.05^{\circ} \pm 3.04$ |
| Cutting positions | | |
| Apical | $0.03^{a}\pm0.29$ | $0.03^a\pm0.28$ |
| Median | $0.34^{b}\pm1.19$ | $0.36^{b} \pm 1.25$ |
| Basal | $2.76^{\circ} \pm 2.63$ | $3.46^{\circ} \pm 3.43$ |
| | | |

Table 4.17. Root length per stem cutting of *Tetrapleura tetraptera* from different sources and cutting positions (mean \pm sd)

*Means with same alphabet are not significantly different (p > 0.05)

| Sources | No of shoots |
|-------------------|------------------------|
| Ibadan | $0.26^{a} \pm 0.74$ |
| Aponmu | $0.58^{b} \pm 1.11$ |
| Iwo | $0.50^{b} \pm 1.37$ |
| Mamu | $0.79^{c} \pm 1.12$ |
| Cutting positions | |
| Apical | $0.04^{\rm a}\pm 0.28$ |
| Median | $0.18^{b} \pm 0.49$ |
| Basal | $1.38^{c} \pm 1.47$ |
| | |

Table 4.18.Effect of seedling sources and cutting positions on number of shoot perstem cuttings of *Tetrapleura tetraptera* (mean ±sd)

*Means with same alphabet are not significantly different (p > 0.05)

4.6.5 Shoot length per stem cutting

There were significant differences in the main and interaction effects of sources and cutting positions on the shoot length per stem cutting (Appendix 28). Stem cuttings from Mamu seedlings was significantly different from Aponmu, Iwo and Ibadan (Table 4.19 and Plate 4.18). However, stem cuttings from Iwo and Ibadan seedlings were not significantly different from one another, but differed from Aponmu and Mamu. The longest $(2.59 \pm 3.4 \text{ cm})$ shoot per stem cuttings was recorded for Mamu, followed by Aponmu (2.11 ± 3.53cm) while the lowest $(1.15 \pm 3.07 \text{ cm})$ was Ibadan (Table 4.19).

Cutting positions differed significantly from one another (Table 4.19). Stem cuttings from the basal region of the seedling had the highest $(4.72 \pm 4.03 \text{ cm})$ shoot length, followed by median $(0.59 \pm 1.49 \text{ cm})$, while the lowest $(0.09 \pm 0.55 \text{ cm})$ was apical positions (Table 4.19).

4.6.6 Number of leaves per stem cutting

There were significant differences in the main and interaction effect of sources and cutting positions on the number of leaves per cutting (Appendix 28). The highest (1.45 ± 1.78) number of leaves were obtained from Mamu seedlings (cuttings), followed by Aponmu (1.18 ± 1.84) while the lowest (0.61 ± 1.59) was Ibadan (Table 4.19). Mamu seedlings were significantly different from Aponmu, Iwo and Ibadan while Iwo and Ibadan did not (Table 4.19).

Similarly, stem cuttings from the basal region of the seedling had the highest (2.54 ± 2.01) number of leaves followed by the middle (0.34 ± 0.85) while the lowest (0.07 ± 0.44) number of leaves was recorded for apical positions (Table 4.19). Stem cuttings from the apical, median and basal nodal positions on the seedling was significantly different from one another (Table 4.19).

| | Shoot length (cm) | Number of leaves | |
|-------------------|-------------------------|-------------------------|--|
| Sources | | | |
| Ibadan | $1.15^a \pm 3.07$ | $0.61^{a} \pm 1.59$ | |
| Aponmu | $2.11^{b} \pm 3.53$ | $1.18^{a} \pm 1.84$ | |
| Iwo | $1.36^{a}\pm2.74$ | $0.72^{a} \pm 1.41$ | |
| Mamu | $2.59^{\circ} \pm 3.40$ | $1.45^{b} \pm 1.78$ | |
| Cutting positions | | | |
| Apical | $0.09^{a}\pm0.55$ | $0.07^{a} \pm 0.44$ | |
| Median | $0.59^{b}\pm1.49$ | $0.34^{\rm b} \pm 0.85$ | |
| Basal | $4.72^{\circ} \pm 4.03$ | $2.54^{\circ} \pm 2.01$ | |

Table 4.19. Seedling sources and cutting positions on shoot length and number of leaves per stem cutting of *Tetrapleura tetraptera* (mean \pm sd)

*Means with same alphabet are not significantly different (p > 0.05)





B. Mamu



C. Ibadan



D. Aponmu

Plate 4.8. Rooted stem cuttings of *Tetrapleura tetraptera* from four different seedling sources

CHAPTER FIVE

5.0

DISCUSSION

5.1 Flowering and fruiting phenology of *Tetrappleura tetraptera*

5.1.1 Timing and duration of flowering of *Tetrapleura tetraptera*

The onset and duration of a phenophase are widely adopted phenomena for describing phenological patterns in tropical trees (Bawa et al., 2003). Flowering pattern in T. *tetraptera* was observed to be episodic as two cycles occurred in a year and three cycles during the assessment period (20 months). The flowering frequency in T. tetraptera was sub-annual (Newstrom et al., 1994). The coincidence of flowering with leaf flush (initiation) at the peak of dry season and/or at the onset of rainfall has also been reported for Spondias mombin (Adler and Kielpinski, 2000), Dalbergia latifolia, Pterocarpus marsupium (Sundarapandian et al., 2005), Shorea roxburgii (Singh and Kuswaha, 2006), Caesalpinia echinata (Borges et al., 2009) and Tamarindus indica (Fandohan et al., 2015). The positive significant correlation between flowering frequency and maximum temperature might indicate that maximum temperature was the proximate factor that triggered flowering activity in T. tetraptera as flowering was mostly initiated during period of high temperature (February-April and November-December). Temperature has also been reported as an important proximate flowering indicator for many tropical tree species (Nadarajan and Pujari, 2018). Flowering phenology requires a cue in which reduction in soil water is involved. Flower buds initiation became noticeable with expansion of inflorescences in T. tetraptera on each twig (new shoots) after rainfall. However, the simultaneous flowering initiation in the species with leaf flushing in all the cycles also indicated leaf flushing as an indirect endogenous control of flowering phenophase and not rainfall. This can be explained by the lack of correlation between rainfall and frequency of flowering trees per month. Studies have revealed that tree

species whose flowering is influenced by timing of leaf phenology at the onset of the rainy season may also be induced to flower in the event of significant rains in the dry season (Borchert et al., 2004; Borges et al., 2009). This characteristic reflects the flowering pattern of some tropical trees (Wei, 2016). Episodic flowering in T. tetraptera might have evolved over time to reduce reproductive failure from flunctuating population of pollinations (Bawa et al., 2003). The lack of significant correlation between flowering frequency and rainfall at the two locations indicated that flowering in T. tetraptera is not directly influenced by rainfall (water availability). This may be explained by the ability of trees to have extensive root system that penetrates deep down to obtain water for physiological activity. The negative significant correlation between flowering frequency and relative humidity indicated that flowering frequency increased when the humidity decreased. In addition, flowering in T. tetraptera indicated that change in moisture stress acts as trigger in flowering of the species because flowering events was initiated at period of low relative humidity. Such flowering behaviour has also been reported for Vitellaria paradoxa (Nguemo et al., 2014). Furthermore, trees with smaller dbh range (65-70 cm) flowered earlier (2 weeks -1 month) than trees with larger dbh (> 100 cm). These findings negate the assumption that tree size is positively related with the onset of flowering (Ollerton and Lack, 1998).

Tetrapleura tetraptera exhibited extended flowering pattern although the duration varied with timing or season of flowering. The maximum flowering period of *T. tetraptera* in the second flowering cycle was one month while first and third flowering cycle were each approximately four months. Flowering duration is affected by the length of favourable conditions for reproduction and the longer the flowering duration, the greater the chances of the flower being pollinated (Brito and Sazima, 2012). An extended duration of flowering may increase attraction to pollinators, their abundance and activities around the species, thereby enhancing reproduction success. Shorter flowering duration in *T. tetraptera* during the second flowering cycle could be due to short period of vegetative growth because flowering and leaf flushing occurs simultaneously or competition for resources between the vegetative and reproductive organs. The longer flowering duration exhibited in the first and third flowering cycle might also be attributed to sufficient

resources which support continuous shoot growth such that inflorescences continue to form on the axil of young leaves, thus ensuring the survival of the reproductive phase (Bullock and Soles-Magallanes, 1990). This is in agreement with Singh and Kushwaha (2006) who opined that production of non-photosynthetic tissues (floral parts) requires higher amount of energy. The lack of significant difference in the flowering patterns of the first and third cycle (episodes) sugests similarity in the climatic factors controlling flower induction in *T. tetraptera*. The similarity exhibited in flowering patterns at FRIN and NIHORT indicated that reproductively mature trees have synchronized flowering when triggered by appropriate environmental cues.

5.1.2 Duration of anthesis on inflorescence of *Tetrapleura tetraptera*

Anthesis on inflorescence of *T. tetraptera* was not simultaneous as previously reported for *Vitex doniana* (Sinebou *et al.*, 2016). Occasionally, anthesis may be completed in a day but generally lasted between two to six days. The pattern of flower buds opening (acropetal/basipetal) on inflorescence was probably due to the location of flower buds on the inflorescence within the tree crown. Flower buds exposed to sunlight initiated anthesis before the shaded flower buds (Borges *et al.*, 2009). This flowering pattern might have evolved to regulate pollen flow or enable insect pollinators to forage for pollen from one inflorescence to another within or between trees. This prevents self pollination and promotes cross pollination (Ewedje *et al.*, 2015). However, simultaneous opening of flowers on an inflorescence, in sexually compatible species, can cause intense daily insect foraging of parts thus promoting self pollination in sexually compatible species (Bawa, 1983).

5.1.3 Floral Synchrony and Flowering intensity in *Tetrapleura tetraptera*

Floral synchrony defines the degree of overlapping among individual flowering trees in a population (Auspurger, 1983). This is essential for out-crossing tree species with low density in order to ensure cross pollination within species (Sakai *et al.*, 1999). In this study, synchronization during the first and third cycles was higher (0.66-0.82) within and between the locations despite differences in the onset of flowering. The high synchronization could be explained by the long flowering duration (3 months). The higher the flowering duration, the higher the flowering synchronization index (Z) and vice-versa.

High flower synchronization exhibits large flower display which attracts high visitation rate by insects or pollinators. This leads to high reproductive success and ensures genetic diversity by facilitating pollen exchange among trees within and outside population (Kudo and Harder, 2005). The low synchrony (0.34-0.43) observed in the second cycle might be attributed to reduced flowering duration of *T. tetraptera* trees.

The flowering intensity indicates the quantity of plant resources designated to the reproductive process. The higher the amount of flowers produced by a tree, the higher the amount of pollen available for pollination. In this study, two major flowering peaks occurred during early rainy season (April to May) and a minor peak at the onset of dry season (October to November). This trend had been reported for most tropical trees (Morellato and Leitao-Filho, 1996).

5.1.4 Fruiting phenology of *Tetrapleura tetraptera*

Fruiting in the first and third cycle started in April and continued during the rainy season while fruiting in the second cycle started between November and December following the peak flowering time. In this study, fruiting phenophase was not seasonal as it occurred in the rainy season (May/June) and in the dry season (January). Tropical trees with similar fruiting patterns have been reported (Bhat and Murali, 2001; Singh and Kushwaha, 2006). First fruiting phenophase in T. tetraptera was completed during the late rainy season (September-October), while the second fruiting cycle was completed in the early rainy season (April). Schedule of fruit fall in the rainy period might have evolved to favour seed germination for subsequent seedling regeneration under natural conditions (Murali and Sukumar, 1994). Nevertheless, fruiting duration was significantly shorter in the second fruiting cycle (early dry season) when compared with the first/third fruiting cycle (rainy season). Rainfall has been attributed to be a major factor influencing the longer fruiting duration particularly during the first/third cycle (Morellato et al., 2000; Singh and Kushwaha, 2006). This is further explained by the significant positive correlation between fruit initiation and monthly rainfall (rs = 0.529 (FRIN); 0.518 (NIHORT)). Fruiting phenology in moist forests of Northeast Korea (Kikim and Yadava (2001) and Western Ghats also corresponded with rainy seasons (Sundarapandian et al., 2005). In the same vein, Fandohan et al., (2015) reported shorter fruiting duration of Tamarindus indica in the drier Sudan compared to Guinea Sudan linking rainfall amounts and duration to active phase of fruit development. The significant negative correlation between fruit initiation and monthly minimum temperature indicated that fruiting in *T. tetraptera* was influenced by minimum temperature. Fruiting phenology of tropical trees has been reported to be influenced by minimum temperature (Tutin and Fernandez, 1993; Nadarajan and Pujari, 2018). High rates of young fruits drop in *T. tetraptera* formed immediately after the peak flowering in the second fruiting cycle might explain the influence of temperature on fruit initiation. Lack of significant difference in fruiting phenology of the two locations suggests similar climatic conditions.

5.1.5 Fruits maturation in *Tetrapleura tetraptera* fruits

Seasons appeared to have effects on timing of fruit maturity in *T. tetraptera*. Fruits matured in approximately three months in the wet period and four months in the hot period. In this study, the longer period of fruit maturation in dry season (second cycle) could be associated with leaf senescence that may result from endogenous regulation of the sequence of resource sharing between the leaves and the developing fruits (Lechowicz, 1995). For instance, sharing of resources may occur when developing fruits utilize resources for shoot growth or resources translocated from leaves (Kozlowski, 1992).

5.2 Floral and inflorescence morphology of *Tetrapleura tetraptera*

The flowers are hermaphrodites with a zygomorphic symmetry. The dense arrangement of flowers on inflorescence and conspicuous colouration suggest insect-pollination syndrome (entomophilous). The floral display in addition to large pollen production on numerous small flowers on a spicate inflorescence might have evolved to attract pollinators from long distances to the species. This is characteristic of members of the sub-family Mimosoideae (Sornsathpornkul and Owens, 1998; Solomon-Raju *et al.*, 2006). The close arrangement of small flowers on an inflorescence may facilitate deposition of the anther of a flower on to the stigma of same or another flower within the same inflorescence. The inflorescence and the bell-shaped corolla serve as landing/pollination unit for the visiting insects. In addition, the centrally located androecium and gynoecium allowed pollinators free access to the sexual organs. These traits facilitate visiting of small insect such as flies,

wasps, ants and beetles which were observed to forage on the plant. Floral visitation by diverse small insects as observed in this study is consistent with tropical trees with small generalist flowers (Momose et al, 1998). For instance, these traits were also found in Acacia species (Sornsathpornkul and Owens, 1998), Pseudopiptadenia contorta and P. leptostachya (Pires and Freitas, 2008). Furthermore, floral display colours (purple-pink and creamy yellow) were strategies probably aimed at achieving inter plant movement within the population in order to achieve cross pollination. Colour changes in flowers encourage pollinators to move from plant to plant so as to reduce the pollination of stigma of the flower by anther of the same plant (Ida and Kudo, 2003). However, the low spatial separation between the style and stamen of the floral structure of T. tetraptera probably facilitated self pollination during foraging activities of insects especially with the stamens being slightly higher than the style. Invariably, the prior extension of filaments during anthesis before later elongation of stigma suggests the flower is protandrous (a condition that may eliminate self-pollination). The lack of significant difference in the floral morphological structures of T. tetraptera between the two locations suggested that the trait is genetically controlled as similar structures were observed at locations except for ovary length. Hence, the variation observed in ovary length suggests the trait was genetically controlled. The waxy pollen character as observed is adapted for pollen transport on insect. The arrangement of T. tetraptera pollen grains in polyads seemed to be an essential trait in ensuring reproductive success following a single pollination event (Erbar and Langlotz, 2005). The ratio of pollen grains in a T. tetraptera polyad to number of ovules in the flower was approximately one. The low ratios have been explained by the greater efficiency of fertilization provided in pollen fused in polyads thus indicating that a pollen-grain is available to fertilize an ovule in most cases (Tandon et al., 2003). A single compatible polyad may fertilize nearly all the ovules in an ovary therefore, ensuring maximum pollination (fertilization) success while eliminating the probability of pollen loss that occurs in monads. Otherwise, an incompatible polyad may result in low reproductive return (Cruden, 2000). The lower the pollen-ovule ratio in species with fused pollen grains, the more efficient the pollen delivery.

The number of inflorescence on twig at FRIN was higher (16.3 ± 6.64) than NIHORT (7.3 \pm 3.2). This was probably due to the size or age of trees which can be explained by the physiological status of the trees. The height of trees in FRIN ranged from 15.5 to 28.5m while NIHORT ranged from 7.8 to 12.5m. Ollerton and Lack (1998) attributed variation in flower morphology of *Lotus corniculatus* to tree size. Crown surface of tall trees in tropical forest appear to be more open to full sunlight whereas crown surface exposure has been reported to be positively associated with floral morphology (Pires *et al.*, 2014).

5.2.1 Insect visitors to *Tetrapleura tetraptera* at FRIN and NIHORT, Ibadan

Flowers of T. tetraptera appeared to be pollinated by insects (entomophilous) who were regular visitors from morning to late afternoon. The presence of similar species of insect visitors at both locations suggested their role as pollinators. Floral traits such as dense arrangement of small flowers on the inflorescence and conspicuous floral colours were typical of insect pollinating species (Faegri and Van der Pijl, 1979). The dense arrangements of flowers on the inflorescence suggested good landing platform by bees and flies. Tetrapleura tetraptera trees were visited by genenalist insects such as Hymenoptera, Diptera and Coleoptera. The main pollinators may be species from the orders: Hymenoptera (bees and wasps) and Diptera (flies). This is based on their interaction with the sexual organs, visiting frequency/abundance and presence of pollen grains that matched the description of T. tetraptera. These insect species made repeated contacts with the anther and stigma using the ventral side during their foraging processes on inflorescence. However, the limited time spent (15 to 30s) per flower and/or inflorescence, frequent visits and inter plant movement of bumble bees and wasps would probably ensure effective cross pollination as seen in *Terminalia pallida* (Solomon-Raju et al., 2012). Pollinator's movement enhance gene flow and pollen transfer thus contributing to high outcrossing rates in tropical trees (Ward et al., 2005). Bombus sp. was more frequent bee species while Apis sp. was limited. This appears that Bombus sp. was more involved in cross pollination. Bombus sp. has been regarded as effective pollinator of Mimosoideae sub family (Stone et al., 2003). The repeated visits of a pollinator to a flower increase seed set and genetic diversity among progeny (Sahli and Coner, 2007). The efficiency of *Apis* sp. in promoting cross pollination in *T. tetraptera* may be limited due to their foraging habit of visiting multiple flowers on inflorescence (Borges et al.,

2009). High polyad distribution on *Bombus* sp. and *Apis* sp. due to their hairy legs and foraging habits possibly enabled them to transport large amounts of pollen grains (Sornsathpornkul and Owens, 1999; Solomon-Raju et al., 2012; Gan et al., 2013; Sinebou et al., 2016). Similarly, polyad loads (12%) on the hairy legs of flies (Calliphora sp.) indicated their bodies were adapted to pollen transport and anther-stigma contact. However, their foraging behaviour which involved moving round the flowers along inflorescence axis during pollen feeding may have limited cross pollination between trees. The time spent in foraging (approximately 2 minutes) by *Calliphora* sp. within the inflorescence compared to bumble-bees and wasps (15 to 30s) may explained why pollination act of the *Calliphora sp.* may not be effective as the bumble bees. The high foraging time spent by ants (Monomorium minimum) and leaf beetle (Brachypnoea sp.) may have promoted self pollination due to movement within densely compacted flowers on T. tetraptera inflorescence. However, cross pollination between trees is needed to ensure genetic diversity (Sakai et al., 1999). The absence of the leaf beetles at FRIN site and their relatively low frequency at NIHORT along with their floral eating habits suggest they were not pollen thieves (Tandon et al., 2001; Pires and Freitas, 2008). Ants were the most abundant at each observation period and this suggest they were resident within the tree. However, they probably serve as guard for the flowers by repelling visits of other non rewarding insect visitor such as those that feed on floral parts (Frame and Durou, 2001). The longer time spent within an inflorescence and relatively low distribution of T. tetraptera polyads on ants suggests their bodies were not adapted for pollen carriage and possibly explained whey their pollinating activity may not be efficient (Faegri and Van Der Pijl, 1979). The repeated visits of the main insect pollinators during the observation period could be attributed to the asynchronous opening of flowers on most inflorescences which made pollen available at all times during the peak flowering period. Peak foraging visits by most insect pollinators (1100h-1300h) corresponded with the peak anthesis on inflorescence. This may be attributed to increase in density of flora resources mainly pollen (Momose et al., 1998). The butterfly species (Danaus chrysippus and Mylothris ocracea) had relatively low number of polyads on them. They are probably occasional pollinators because of their presence at the two locations. Tubular flower morphology has been noted to be characteristic of butterfly pollination (Momose et al., 1998) while T. *tetraptera* flower is bell-shaped. However, butterflies visits might be attributed to the brightly coloured flowers of *T. tetraptera*. *Alydus eurinus* (long-horned bug) and *Peucetia viridians* (green lynx spider) may not be considered as effective pollinator as the former which is a floral eater had relatively little pollen load (2%) while the latter predates on other visiting insects like wasps. Their low frequency distribution and lack of interplant movement further explained their ineffective role as pollinators in the species. However, pollination may likely occur during the feeding process due to dense arrangement of flowers on the inflorescence. *Orthetrum brachiale* (dragon fly) did not show any definite foraging behaviour on *T. tetraptera* flowers; although, it occurred at both locations. Hence, they could be classified as pollen thief's category. Trees often serve as a hosts or habitats to non- pollinating (Frame and Durou, 2001).

5.2.2 Reproductive efficiency of *Tetrapleura tetraptera* trees

The overall reproductive efficiency of *T. tetraptera* from FRIN (0.5 ± 0.2 %) and NIHORT (0.4 ± 0.1 %) in Ibadan (Oyo state) as observed in this study was greater than 0.05% reported in the same species from Edo state of Nigeria (Omokhua and Ukoimah, 2008). The low fruit set rate (approximately 10-11%) of *T. tetraptera* in open pollination is typical of most tropical tree species (Bawa *et al.*, 1983; Solomon-Raju *et al.*, 2012). However, this value was higher than that reported for *Terminalia pallida* (5.5%) and *Acacia caesia* (4.3%) (Solomon Raju *et al.*, 2012) but lower than that of *Tetracentron sinense* (18.43%) (Gan *et al.*, 2013), *Dacryodes edulis* (48%) (Makueti *et al.*, 2015) and *Pentadesma butyracea* (49%) (Ewedje *et al.*, 2015). The variations observed in the fruit set rate in *T. tetraptera* among the cycles could be attributed to variations in environmental conditions. Higher fruit set in the second cycle might be due to favourable temperature effect on pollinators and availability during the flowering period (dry season). Seasonal variation in flowering and fruiting events reflect the influence of proximate environmental index that trigger flowering and ultimate factors such as abundance of pollinators that select for particular reproductive phenology (Adler and Kielpinski, 2000).

Low fruit set rate in species of Fabaceae have been attibuted to self incompatibility (Tandon *et al.*, 2003). For instance, any pollination activity between a self incompatible polyad and a stigma usually result in reproductive failure. Low fruit set and high fruit

drop (abortion) rate in T. tetraptera may be attributed to inherent capacity of the tree to support the nutrient/energy needs of the numerous flowers on the inflorescence (Holland and De Angelis, 2002; Solomon-Raju et al., 2006). For instance, synchronous anthesis of large number of flowers on inflorescence of T. tetraptera may limit fruit set due to competition for nutrients and other resources. Also, early anthesis of successfully pollinated flowers on the inflorescence of T. tetraptera may restrict flowers pollinated later from gathering adequate resources to set fruit. Flowers or fruits that initiated growth earlier on same inflorescence tends to surpass other flowers or fruits growing later, a phenonomenon referred to as primogenous dominance (Bangerth, 2000). The slight increase in fruit abortion rate of T. tetraptera in the second cycle (dry season) fruiting may be explained by seasonal differences in environmental conditions (moisture and temperature) among the fruiting periods. Other causative factors of immature fruit abortion in fruit trees included adverse environmental conditions, pest and disease attack and competition between plant organs (Racsko et al., 2007). According to these authors, high temperature was noted to increase the rate of gynoecium abortion in plum. Resource sharing and competition between vegetative (shoot) and reproductive organs is another factor that may likely promote fruit abortion in T. tetraptera. The fruit drop rate recorded for T. tetraptera from Benin and Ekpoma in Edo state of Nigeria was higher (99.95%) than what was found in this study (Omokhua and Ukoimah, 2008). Naturally, fruiting efficiency is regulated in tropical fruit trees through physiological drop of young developing fruits because of associated dominance effect of adjoining fruit and/or nearby shoots (Bangerth, 2000). This phenomenon tends to optimize efficient use of tree resources for fruit growth, seed development and maturity in T. tetraptera (Kay, 2006; Sun et al., 2007).

Moreover, reproductive or pollination efficiency in tropical trees with naturally low fruit set rate is best evaluated by the number of seed set per fruit. In this study, the number of seeds per fruit in *T. tetraptera* from both locations confirmed the higher pollination efficiency found in pollen arranged in polyads. The significant variation between the numbers of seeds per fruit of *T. tetraptera* from FRIN (12.5 ± 2.9) and NIHORT ($15.1 \pm$ 2.9) could be explained by greater diversity and distribution of insect visitors to *T. tetraptera* trees at NIHORT than FRIN. The higher diversity and distribution of insect visitors at NIHORT may facilitate repeated visits of insects to an inflorescence, therefore, increasing seed set per fruit (Karron *et al.*, 2006). High synchronization of flowering at NIHORT (0.88) than FRIN (0.67) appeared to also facilitate higher pollination efficiency and/or maximum seed set. Display of high synchronization within and among tropical trees had been indicated as necessary to attract pollinators and facilitate pollen exchange within and between populations (Bawa *et al.*, 2003). Generally, high reproductive success in Mimosoideae results because a compactible polyad may pollinate all ovules in a flower and consequently optimizing seed set (Tandon *et al.*, 2001).

5.3 Morphological variation in fruit and seeds of *Tetrapleura tetraptera*

The morphological characters of fruits and seeds in T. tetraptera varied significantly from one source to another. Inter and intra-specific variations in the fruits and seeds morphology in T. tetraptera may be an indication of genetic and environmental effects (Assogbajo et al., 2010). The low coefficient of variation in fruit length (9.4 - 12.4%) and fruit width (9.1 - 13.6%) within source and the higher estimates of coefficient of variation in fruit length (13.7%) and width (16.2%) among the four sources suggested these variables were environmentally influenced (Wulff, 1995). Similar variations in fruit traits were observed for Chrysophyllum albidum (Dadegnon et al., 2015), Pentadesma butyracea (Ewedje et al., 2012) and Irvingia gabonensis (Leakey et al., 2000). Morphological traits in plant species have been noted to vary along climatic region and ecological gradients (Fredrick et al., 2015). The number of seeds per pod in T. tetraptera was the character with high inter-source variation, indicating this trait is important. This variation was higher (30.9%) than seed weight (16.1%), fruit length (13.7%) and fruit width (16.2%). Similarly, within-source variation for number of seeds was higher than source variation confirming that morphological traits of T. tetraptera is not only affected by environmental variations but also genetically controlled. For instance, within source variation of the number of seeds per pod from Ibadan (36.5%) and Mamu (34.6%) source was higher than the inter source variation (30.9%). Hence, selection of ascensions from Ibadan and Mamu sources would be beneficial to genetic improvement and fruit orchard for seed production (Sudrajat, 2016). The genetic control of morphometric traits of seeds of T. tetraptera has been recognized for tropical trees (Abraham et al., 2006). However, higher coefficient of variation (16.1%) in the seed weight among the sources compared with low coefficient of variation within sources (0.5 - 2.6%) also confirmed the influence of environmental variations on seed development (Singh and Bhatt, 2008). Across the four sources, seed weight of T. tetraptera varied eight times than fruit length and width and this may indicate that seed weight is more variable than fruit length and width (Shankar and Synrem, 2012). The highest seed weight and number of seeds per pod recorded by Aponmu (Ondo state) source suggests favourable environmental conditions within the source that may facilitate abundance and activities of floral visitors thus maximizing reproductive success by enhancing seed set and development. Influence of environmental factors during seed development has a major role in the seed weight determination (McAllister, 2005). Factors such as inadequate pollination, pollination by non-viable pollen, embryo degeneration affect seed production (Mamo et al., 2006). Availability of resources during the fruiting period has been attributed to variation in seed weight of Darlbergia sissoo (Singh and Bhat, 2008) and Magnolia officinalis (Shu et al., 2012). The positive strong correlation of number of seeds per pod and seed weight with seed longitudinal origin and altitude in this study is consistent with the established fact that these traits are affected by environmental factors. This is a selection criterion for superior phenotypes and highly adapted seed sources. Fruit dimensions (length and width) may not be employed as desirable traits for source selection because there was no correlation between the two characters as observed in the present study.

5.4 Seedling's growth and biomass assessment in *Tetrapleura tetraptera*

The increase in growth variables across the sources over time after planting might be attributed to significant positive correlation among the seedling growth characteristics. Evaluation of seed sources of tree species is aimed at measuring the extent of genetic variation in order to govern selection of superior phenotypes and well adapted accessions. In this study, the highest coefficient of variation in seedling's height (33.5%), collar diameter (32.5%) and number of leaves (22.0%) was obtained from Mamu source which indicated that high level of genetic variations existed within this source. The existence of these variations confirmed that genetic factors play a major role in influencing the growth characteristics (Shankar and Synrem, 2012). Height of *T. tetraptera* varied significantly within seed sources. Similar results were also reported for *Adansonia digitata*

(Assogbadjo et al., 2006) and Anthocephlus cadamba (Sudrajat, 2016). Meaningful improvement in the seedling's height, collar diameter and number of leaves could be achieved by selecting seedlings with superior phenotype within these sources. The highest coefficient of variation (39%) in final biomass accumulation found within Ibadan source suggests that seedlings from this source were highly variable in their biomass accumulation thus selection within this source would be beneficial for the species improvement. Meanwhile, the highest leaf area found in seedlings from Ibadan appeared to influence the highest absolute growth rate and total biomass accumulation obtained in T. tetraptera seedlings from this source (Fornah et al., 2017). The leaf is the photosynthetic site of a plant therefore, the number of leaves produced by a plant influences the photosynthetic rate and how other organs of the plant store food over time. Positive correlations found among growth variabled of T. tetraptera seedlings in this study implied that improvement in one character will correspond to improvement in another. Relative growth rate, absolute growth rate and total biomass of T. tetraptera seedlings followed a similar trend across the seed sources. This pattern might be attributed to the variation in net assimilation rate (NAR), influenced by leaf area (Shipley, 2006).

5.5 Stem cutting's rooting and growth regulators on *Tetrapleura tetraptera*

Stem cuttings taken from seedlings of *T. tetraptera* were amenable to vegetative propagation with or without growth regulator application. Auxins had been indicated to enourage adventitious root growth in stem cuttings of tropical trees by their potential to initiate lateral root primordial development and transportation of carbohydrates to the region of actively dividing cells (Hartmann *et al.*, 1997). For this experiment, growth regulators had no significant influence the survival and root formation even with varying concentrations in growth regulators. Other tropical trees like *Nauclea diderrichii* (Leakey *et al.*, 1990), *Milicia excelsa* (Ofori *et al.*, 1996), *Irvingia gabonensis* (Shiembo *et al.*, 1996), *Enantia chlorantha* (Gbadamosi and Oni, 2005) and *Buchhorlzia coriacea* (Akinyele, 2010) have also been documented for this observation. There were also records of lack of significant differences in the impact of growth regulators on the rooting of stem cuttings for *Switennia macrophylla* (Hossain *et al.*, 2004). The production of long root by cuttings that were not treated with growth regulators (control) might be an indication of

the presence of naturally occurring endogenous root promoting substances in the juvenile tissues of T. tetraptera cuttings. Successful rooting of stem-cuttings without growth regulators also suggests the presence of endogenous auxins at the time of cutting severance (Kebede et al., 2013). The presence of higher phenols in young tissues of scions was also attributed to higher survival and sprouting percentage noted on untreated cuttings of Jatropha curcas compared to the treated cuttings the species (Adekola and Akpan, 2010). Thus, exogenous application of auxin may be promotive, ineffective or inhibitory based on the extent of endogenous growth controlling substances on the cuttings, (Kesari et al., 2010). The concentration of IBA above and/or below 0.4 % has been reported to inhibit root formation in Milicia excelsa and Irvingia gabonensis (Ofori et al., 1996). Auxins concentration greater than those normally found in plant tissues can lead to tissue death in stem cuttings of some plant species (Hartmann et al., 2002). Coconut water gave a better performance than auxins as the highest number of roots was produced on T. tetraptera cuttings treated with coconut water. Previously, Usman and Akinyele (2015) reported higher sprouting percentage in coconut water-treated Massularia acuminata stem cuttings. Inaddition, coconut water output was further indicated by its stimulatory effect on the length of the root and the length of longest root in each T. tetraptera cutting. This role is also evident in stimulating rooting and development of tropical fruit trees (Trevisan et al., 2005; Okunlola, 2016; Dunsin et al., 2014). The presence of amino acid, myo-inositol, glucose, and phenyl urea microconstituents might explain the growth stimulating effect of coconut water (Agele et al., 2013).

5.6 Influence of stockplant sources and stem cutting positions on rooting of *Tetrapleura tetraptera*

Successful vegetative propagation of tropical trees using juvenile stem cuttings can be enhanced when interacting factors such as stockplant physiology and cutting positions are taken into consideration (Leakey, 2014). In this study, the rooting success may have been affected by stockplant source and position of shoots on the stockplant because of significant variation in all the measured variables of *T. tetraptera* stem cuttings. Therefore, adequate understanding of these factors may be essential for maximizing the success and cost effectiveness of macropropagation programme.

Stem cuttings obtained from Mamu seedlings had the highest rooting performances and differed significantly from the other sources. The considerable differences may be ascribed to superior inherent variation in the seedlings compared with other sources (Tchoundjeu and Leakey, 2001; Leakey, 2014). Mamu seedlings (stockplants) had the highest shoot dry weight which may be responsible for the outstanding performance in the rooting experiments. The positive correlation between seedling's growth variables and rooting variables in the present study also confirmed that stockplant's physiology was significantly associated with the rooting success of stem cuttings. Adaptation to environmental stress after the cuttings were severed from the stockplant may also be responsible for the variation in cutting's survival. Variation in rooting response of cuttings from different sources had also been reported for *Lovoa trichilioides* (Tchounjeu and Leakey, 2001).

The significant variation in the interaction effects of stockplant source and cutting positions also established the important impact of nodal positions on the rooting capacity of T. tetraptera. However, the rooting ability of cutting positions varied with tree species, ranging from apical to basal cutting positions. For instance, basal cutting position significantly enhanced rooting ability in the species stem cuttings when compared with apical and middle positions regardless of the sources of stockplant in this study. Similar results had been reported in rooting of *Khaya ivorensis* (Tchounjeu and Leakey, 2000) and Peltophorum pterocarpum (Saifuddin et al., 2013). The ability of basal cutting positions to enhance rooting significantly might be explained by bud dormancy in the region. In the present study, the dormant buds on the basal cuttings appeared to have enhanced carbohydrate retention for subsequent root initiations while the apical and middle cuttings flushed out shoots rapidly three to four days after setting, thus depleting the carbohydrate content needed to facilitate cutting's survival, root initiation and leaf maintenance in the region. This might explain the poor survival and death of cuttings observed in stem cuttings obtained from both apical and median positions since cellular activities during rooting require availability of carbohydrates in the stem (Kesari et al.,

2010). Inaddition, the basal cuttings of *T. tetraptera* shoots were larger in diameter than apical and middle cutting positions which suggested that cutting size could also be attributed to enhanced rooting success in this position. During rooting process of *T. tetraptera*, most cuttings propagated from apical and middle nodal positions dry out rapidly after shoot flushing three to four days after setting and this may indicate the depletion of carbohydrates and/or endogenous growth regulators that may be used for root initiation and subsequent leaf maintenance.

On the other hand, propagules from apical cutting positions had equally been reported to enhance rooting when compared with basal cutting positions in tropical trees such as *Lovoa trichilioides* (Tchounjeu and Leakey, 2001), *Triplochiton scleroxylon* (Leakey and Storeton-west, 1992), *Stevia* species (Beemnet and Solomon, 2012) and *Azadirachta indica* (Palanisamy and Kumar, 1997). High rooting success in apical cuttings of *Triplochiton scleroxylon* was explained by higher concentration of soluble carbohydrates in the region (Leakey and Storeton-west, 1992).

CHAPTER SIX

6.0. SUMMARY AND CONCLUSION

6.1. SUMMARY

Investigations were carried out on the reproductive phenology of *Tetrapleura tetraptera* in Forestry Research Institute of Nigeria (FRIN) and National Institute of Horticultural Research (NIHORT) in Ibadan, Oyo state of southwestern Nigeria; its flowering and fruiting patterns, floral morphology, insect visitors and reproductive efficiency, germplasm variations, early seedling growth and macro-propagation techniques between 2016 and 2018. Preliminary survey was carried out to ascertain the fruiting populations of *T. tetraptera* in southwestern Nigeria between July and November 2016. Hence, Ibadan, Mamu, Iwo and Aponmu source were selected.

Assessment of reproductive phenology of *T. tetraptera* was carried out on purposively selected matured trees from FRIN and NIHORT in Ibadan, Oyo state based on protection and availability. Onset and duration (days) of flower and fruit development were monitored from January 2017 to July 2018 to determine flowering intensity and synchrony. Relationship between climatic factors and flowering phenology were also investigated. Insect visitors to *T. tetraptera*, frequency of their visit, pollen load, floral/pollen morphology and fruiting efficiency were all assessed between 2017 and 2018.

Germplasm variations of *T. tetraptera* were conducted in 2016 across the four selected locations within southwestern Nigeria. One hundred and fifty matured fruits were randomly sampled in each of the locations to determine the fruit morphological variations that exist within the geographical distribution of *T. tetraptera* inorder to identify superior seed source for genetic improvement and contributes to species domestication.

The scarcity of information on growth rates, genetic variation and sources of superior planting stocks of *T. tetraptera* were identified as major constraint for its inclusion in agroforestry system.

Nursery experiments were carried out between 2017 and 2018 at the screen house of Forest Production and Products Department, University of Ibadan, Nigeria with a view to proffer solution to this.

Lastly, macropropagation experimens were performed in 2017 to test the amenability of *T. tetraptera* to vegetative propagation. This entails investigations on the impact of growth regulators on the rooting of *T. tetraptera* stem cuttings; nodal positions and stockplant's source.

The results showed that;

- 1. Flowering in *T. tetraptera* coincided with leaf flushing and pattern was subannual with two peaks (major and minor). First flowering occurred from February to July while second flowering from October to December respectively. Flowering was initiated at a period of high temperature and low relative humidity.
- Major flowering peak (FRIN (93.8 %) and NIHORT (81.3 %)) occured at the peak of dry season or onset of rainy season (April-May) while second peak (FRIN (56.3%) and NIHORT (45.8 %)) occurred in the early dry season (November to December). Flower buds commenced anthesis 29.5 ± 1.57 days after flower buds initiation.
- 3. Flowering in *T. tetraptera* was highly synchronized within and between locations in the first flowering cycle than second. Fruiting began approximately one month after flowering during the peak of flowering. Fruiting was not seasonal and maturity of fruits occurred from three to four months after setting depending on season.
- 4. Flower/inflorescence morphology revealed that flowers were hermaphrodites, pentamerous and zygomorphic. The number of flowers per inflorescence ranged from 294 ± 40.3 to 297 ± 27.3 . The polyad to ovule ratio ratio was estimated to be approximately one thus, eliminating the probability of pollen loss.
- 5. Investigation on insect visitors to *T. tetraptera* revealed that insect species from the Hymenoptera order were pollinators while insect species of the

Lepidoptera, Odonata, Araneae, Hemiptera and Coleoptera order were regarded as pollen thieves. *Bombus* sp. had the highest pollen load (25%) and thus regarded as most effective pollinators of *T. tetraptera*. The fruiting efficiency per inflorescence was low ranging from 0.25 to 0.99 %' while the number of matured fruit per inflorescence ranged from one to two. Fruit abortion rate was higher in the second fruiting season than the first.

- 6. Fruit/seed sources influenced the morphological traits of fruits and seeds of *T*. *tetraptera*. Aponmu (Ondo state) had the highest seed weight (16.16g ± 0.08 / 100 seeds) and number of seeds per pod (15.6 ± 2.44). Number of seeds per pod was the most variable among the fruit traits indicating that this character was the most superior for improvement.
- Early growth and biomass accumulation of *T. tetraptera* seedlings from the four seed sources revealed the pattern of genetic variation in the species. Hence, propagules from Mamu source were identified as most important for genetic improvement.
- 8. Vegetative propagation of *T. tetraptera* using stem cuttings was successful with or without growth regulators. Coconut water had the highest number of roots (5.00 ± 2.65) and root length $(5.02 \pm 1.61 \text{ cm})$.
- 9. Seedlings source and stem cutting positions of *T. tetraptera* are important factors that enhance rooting success in the species. Mamu seedlings had the best rooting performance. The basal cuttings also had the highest performance (cuttings survival (56.3 \pm 29.1%), number of roots (2.19 \pm 2.28), root length (2.76 \pm 2.63) and number of shoots (1.38 \pm 1.47)) while the apical cuttings had the lowest survival (1.8 \pm 7.3%), number of roots (0.04 \pm 0.3), root length (0.34 \pm 1.19) and number of shoots (0.04 \pm 0.28)

6.2 CONCLUSIONS

The research findings revealed that the pattern of reproductive phenology in *T. tetraptera* is associated with seasonal changes in temperature and relative humidity, an environmental cue that synchronize flowering events in the late dry season and early wet season. The episodic and synchronized flowering pattern eliminated the possibility of reproductive failure and ensures continued survival of the species. The synchronization within and between populations of *T. teraptera* was likely controlled

by similar environmental variables thus global climatic change might have strong implications on future reproductive success of the species.

Information on the floral traits, insect visitors and reproductive efficiency would be useful for planning breeding and conservation strategies. Fruiting efficiency in *T. tetraptera* seems to be inherent genetic characteristics and pollination efficiency may be better measured by the number of seeds set per fruit.

The study on morphological traits and early growth characteristics indicated variations within and between the seed sources. This would have implications in selection, improvement and management. The number of seeds per fruit had the highest variations among the fruit traits and thus regarded as an important morphological trait in *T. tetraptera* for germplasm collection and tree improvement. Fruit length and width were less variable trait. High level of genetic variation existed within the seed sources of *T. tetraptera* and selection of best phenotype within Mamu source will be beneficial for its improvement.

Vegetative propagation by stem cuttings provided prospect for mass multiplication of *T. tetraptera* using a low-technology high humidity propagator. Stem cuttings of *T.tetraptera* rooted easily with or without growth regulators. Coconut water could serve as cheap alternative growth regulator in mass production. However, attention must be given to the position of cuttings on the shoot and physiological condition of stockplant sources to facilitate high rooting success.

RECOMMENDATION

Based on the various output of this study, the following recommendations are made;

- 1. There is need to embark on long term evaluation of phenological patterns across distinct ecological zones of the species range to provide a better understanding of evolutionary process.
- 2. Controlled pollination experiments that would provide a clearer understanding of breeding systems in the species should be embarked upon for sustainable utilization.

- 3. Seeds from Mamu, Ibadan and Aponmu could be selected for genetic improvement of *T. tetraptera*
- 4. Stem cuttings arising from the basal position on the shoot is most preffered for maximum rooting success.

CONTRIBUTIONS TO KNOWLEDGE

- Tetrapleura tetraptera exhibited sub-annual (episodic) and synchronous flowering patterns with two peaks. Mature fruits can be collected for sowing in March and July/August, hence provides a guide in its breeding program. Temperature acts as a proximate cue for initiating flowering in *Tetrapleura tetraptera*.
- 2. *Tetrapleura tetraptera* flowers are insect pollinated (entomophilous) while also exist several pollon thieves. The polyad-ovule ratio increase seed set.
- 3. Seed sources influenced fruit, seed and seedling morphological traits of *T*. *tetraptera*, hence must be given adequate consideration in germplasm collection for improvement and domestication.
- 4. Macro-propagation via stem cuttings is a viable alternative in silvicultural management and conservation of *T. tetraptera* in view of diminishing population in its natural range.

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APPENDICES

| Location | | | | | |
|--------------------|-----------------|-----------------|--|--|--|
| Variables | FRIN | NIHORT | | | |
| DBH (cm) | 52.6 ± 3.28 | 38.3 ± 5.94 | | | |
| Height (m) | 22.0 ± 5.81 | 9.95 ± 2.16 | | | |
| Crown diameter (m) | 13.1 ± 0.38 | 14.0 ± 1.32 | | | |
| | | | | | |

Appendix 1. Morphometrics of *Tetrapleura tetraptera* trees

DBH; Diameter at breast height

| Appendix 2: | Flowering | duration | (days) | of | Т. | tetraptera | trees | among | the | flowering |
|-------------|-----------|----------|--------|----|----|------------|-------|-------|-----|-----------|
| seasons | | | | | | | | | | |

| Location/tree no. Flowering cycle | | | |
|-----------------------------------|-----|----|-----|
| FRIN | 1 | 2 | 3 |
| T1 | 95 | - | 108 |
| T2 | 107 | 60 | 78 |
| Τ3 | 113 | 39 | 82 |
| T4 | 102 | 47 | 83 |
| NIHORT | | | |
| T1 | 95 | 59 | 133 |
| T2 | 79 | 61 | 98 |
| Τ3 | 109 | 69 | 125 |
| T4 | 92 | 62 | 118 |
| T5 | - | - | 89 |
| Т6 | - | - | 77 |

| FRIN | First cycle | Second cycle | Third cycle |
|-----------|----------------------------|-----------------|-----------------|
| T1 | 0.86 | 0 | 0.34 |
| T2 | 0.82 | 0.47 | 0.74 |
| T3 | 0.8 | 0.66 | 0.73 |
| T4 | 0.78 | 0.60 | 0.74 |
| Mean ± sd | $\boldsymbol{0.82\pm0.03}$ | 0.43 ± 0.29 | 0.64 ± 0.27 |
| NIHORT | First cycle | Second cycle | Third cycle |
| T1 | 0.73 | 0.47 | 0.81 |
| T2 | 0.65 | 0.55 | 0.93 |
| Т3 | 0.51 | 0.49 | 0.73 |
| T4 | 0.73 | 0.54 | 0.84 |
| T5 | - | - | 1.02 |
| T6 | - | - | 0.97 |
| Mean ± sd | 0.66 ± 0.1 | 0.34 ± 0.27 | 0.88 ± 0.11 |

Appendix 3: Floral synchrony index within *T. tetraptera* trees and locations

FRIN - Forestry Research Institute of Nigeria, Ibadan

NIHORT - National Institute of Horticultural Research and Training, Ibadan

| Infloresence number | Developing buds (fl2) | Developed buds (fl3) | Anthesis (fl4) |
|---------------------|-----------------------|----------------------|----------------|
| 1 | 14 | 21 | 28 |
| 2 | 16 | 23 | 30 |
| 3 | 14 | 21 | 28 |
| 4 | 16 | 23 | 29 |
| 5 | 16 | 23 | 30 |
| 6 | 14 | 21 | 29 |
| 7 | 20 | 24 | 30 |
| 8 | 21 | 24 | 28 |
| 9 | 14 | 21 | 28 |
| 10 | 16 | 24 | 27 |
| 11 | 16 | 24 | 32 |
| 12 | 16 | 24 | 32 |
| 13 | 14 | 22 | 28 |
| 14 | 16 | 24 | 29 |
| 15 | 18 | 26 | 30 |
| 16 | 18 | 26 | 32 |
| 17 | 16 | 24 | 29 |
| 18 | 16 | 24 | 30 |
| 19 | 16 | 24 | 30 |
| 20 | 16 | 24 | 30 |
| Mean ± SD | 15.95 ± 2.01 | 22.85 ± 1.50 | 29.45 ± 1.57 |

Appendix 4: Floral development (days) of *Tetrapleura tetraptera*

| Source of variation | Df | Ms | F cal | P value |
|------------------------|----|--------|-------|---------|
| Fruiting cycle | 2 | 324.99 | 12.82 | 0.003* |
| Error | 8 | 25.34 | | |
| Total | 10 | | | |

Appendix 5: ANOVA of period of fruit maturation of *Tetrapleura tetraptera* in FRIN

*Significant at P < 0.05

Appendix 6: ANOVA of period of fruit maturation of *Tetrapleura tetraptera* at NIHORT

| Source of | Df | Ms | F cal | P value |
|-----------|----|--------|-------|---------|
| variation | | | | |
| Treatment | 2 | 445.44 | 36.48 | 0.00* |
| Error | 11 | 12.21 | | |
| Total | 13 | | | |
| | | | | |

*Significant at P < 0.05

| S/N | FRIN | NIHORT |
|------|------|--------|
| 1 | 7 | 6 |
| 2 | 6.3 | 8.2 |
| 4 | 9 | 9.2 |
| 5 | 11 | 17 |
| | 5 | 7.5 |
| 6 | 9 | 10 |
| 7 | 7 | 9.5 |
| 8 | 9.8 | 9.2 |
| 9 | 10.6 | 12 |
| 10 | 9.3 | 11 |
| 11 | 7.1 | 13 |
| 12 | 9.5 | 8.7 |
| 13 | 9 | 9 |
| 14 | 11 | 9.8 |
| 15 | 10 | 7.1 |
| 16 | 10.5 | 9.5 |
| 17 | 8.5 | 7.5 |
| 18 | 9.8 | 8.7 |
| 19 | 8 | 15 |
| 20 | 10.8 | 12.5 |
| 21 | 7.5 | 7.1 |
| 22 | 7.7 | 5 |
| 23 | 8.5 | 6.5 |
| 24 | 9 | 6 |
| 25 | 10 | 7.7 |
| Mean | 8.76 | 9.31 |

Appendix 7: Length of inflorescence of *Tetrapleura tetraptera* in the first flowering cycle

| Order | Family | Species | FRIN | NIHORT | Morning | Afternoon |
|-------------|---------------|-----------------------|------|--------|---------|-----------|
| Hymenoptera | Apidae | Bombus sp. | 56 | 36 | 47 | 45 |
| | | Apis mellifera. | 24 | 13 | 24 | 13 |
| | Sphecidae | Sceliphron sp | 42 | 63 | 42 | 61 |
| | Vespidae | Polistes belicosus | 0 | 39 | 19 | 20 |
| | | Vespula vulgaris | 59 | 0 | 29 | 30 |
| | Formicidae | Monomorium minimum | 106 | 122 | 120 | 108 |
| | | unidentified | 0 | 50 | 25 | 25 |
| Lepidoptera | Danaidae | Danaus chrysippus | 18 | 43 | 30 | 31 |
| | Pieridae | Mylothris ocracea | 0 | 28 | 18 | 10 |
| Diptera | Calliphoridae | Caliphora sp. | 25 | 45 | 21 | 24 |
| Odonata | Libelludae | Orthetrum brachiale | 49 | 61 | 81 | 14 |
| Araneae | Oxyopidae | Peucetia viridians | 0 | 14 | 5 | 6 |
| Hemiptera | Alydidae | Alydus eurinus | 0 | 14 | 7 | 7 |
| Coleoptera | Chrysomelidae | Brachypnoea sp. | 0 | 5 | 3 | 2 |
| | | Total | 379 | 533 | 471 | 396 |

Appendix 8: Frequency distribution of insect visitors of *Tetrapleura tetraptera*

| | | | Ti | me | | |
|-------------|---------------|------------------------|-----------------|----------------|------------------|---------------------|
| Order | Family | Species | 0700 – 1100h | 1200- 1700h | Mann- Whitney | P- value |
| | | | | | U test | |
| Hymenoptera | Apidae | Bombus sp. | 3 | 3 | 155 | 0.839 _{ns} |
| | | Apis mellifera. | 1 | 1 | 118 | 0.161 _{ns} |
| | Sphecidae | Sceliphron sp | 3 | 4 | 97 | 0.04* |
| | Vespidae | Polistes belicosus | 1 | 1 | 158 | 0.888_{ns} |
| | | Vespula vulgaris | 2 | 2 | 161 | 0.963 _{ns} |
| | Formicidae | Monomorium minimum | 7 | 6 | 110 | 0.097_{ns} |
| | | Orange ant | 2 | 2 | 162 | 1.000 _{ns} |
| Lepidoptera | Danaidae | Danaus chrysippus | 2 2 | 2 | | 0.988_{ns} |
| | Pieridae | Mylothris ocracea | 1 | 1 | 130 | 0.323_{ns} |
| Diptera | Calliphoridae | Caliphora sp. | 1 | 1 | 154 | 0.815_{ns} |
| Odonata | Libelludae | Orthetrum brachiale | 5 | 2 | 8 | 0.000* |
| Araneae | Oxyopidae | Peucetia viridians | 1 | 1 | 146 | 0.606 _{ns} |
| Hemiptera | Alydidae | Alydus eurinus | 1 | 1 | 162 | 1.000 _{ns} |
| Coleoptera | Chrysomelidae | Brachypnoea sp. | 1 | 1 | 153 | 0.791_{ns} |

Appendix 9: Mean frequency of periodic distribution of insect species on flowers of *T. tetraptera* in NIHORT and FRIN

Note: * Significant at P < 0.05; ns- not significant

| | | | Lo | cation | | |
|-------------|---------------|------------------------|------|--------|----------------------------|---------------------|
| Order | Family | Species | FRIN | NIHORT | Mann- Whitney U test | P- value |
| Hymenoptera | Apidae | Bombus sp. | 3 | 2 | 105 | 0.074_{ns} |
| | - | Apis mellifera. | 1 | 1 | 118 | 0.171_{ns} |
| | Sphecidae | Sceliphron sp | 3 | 4 | 118.5 | 0.171 _{ns} |
| | Vespidae | Polistes belicosus | 0 | 3 | 36 | 0.000* |
| | | Vespula vulgaris | 3 | 0 | 0 | 0.000* |
| | Formicidae | Monomorium minimum | 6 | 7 | 130 | 0.308 _{ns} |
| | | Orange ant | 0 | 3 | 0 | 0.000* |
| Lepidoptera | Danaidae | Danaus chrysippus | 1 | 3 | 46 | 0.000* |
| | Pieridae | Mylothris ocracea | 0 | 2 | 10.5 | 0.000* |
| Diptera | Calliphoridae | Caliphora sp. | 2 | 3 | 26.5 | 0.000* |
| Odonata | Libelludae | Orthetrum brachiale | 3 | 4 | 128 | 0.279_{ns} |
| Araneae | Oxyopidae | Peucetia viridians | 0 | 1 | 63 | 0.001* |
| Hemiptera | Alydidae | Alydus eurinus | 0 | 1 | 108 | 0.091_{ns} |
| Coleoptera | chrysomelidae | Brachypnoea sp. | 0 | 1 | 117 | 0.161 _{ns} |

Appendix 10: Mean frequency of occurrence of insect species in NIHORT and FRIN

*Significant at P < 0.05; ns – not significant at P > 0.05

| Seed weight | | | | |
|------------------|----------|---------|----------|----------|
| Source | of DF | MS | F- value | P- value |
| variation | | | | |
| Source | 3 | 17.7541 | 447.68 | 0.000* |
| Error | 8 | 0.0397 | | |
| Total | 11 | | | |
| Fruit length | | | | |
| Source | 3 | 510.234 | 118.93 | 0.000* |
| Error | 596 | 4.29 | | |
| Total | 599 | | | |
| Fruit width | | | | |
| Source | 3 | 6260.50 | 245.83 | 0.000* |
| Error | 596 | 25.47 | | |
| Total | 599 | | | |
| Seed number | per pod | | | |
| Source | 3 | 1000.76 | 87.45 | 0.000* |
| Error | 596 | 11.44 | | |
| Total | 599 | | | |
| * aignificant at | D < 0.05 | | | |

Appendix 11: Analysis of variance on the effect of source on morphological variation in fruits/seeds of *Tetrapleura tetraptera*

*significant at P < 0.05

| | Fruit | Seed weight | Seeds | Fruit width |
|------------------|--------|-------------|--------|-------------|
| | length | | | |
| Seed weight | -0.045 | | | |
| Number of seeds | 0.202 | 0.956* | | |
| Fruit width | -0.162 | -0.463 | -0.628 | |
| Min altitude | -0.277 | 0.966* | 0.849 | -0.311 |
| Max altitude | 0.022 | 0.997* | 0.965* | -0.447 |
| /lin temperature | -0.647 | 0.597 | 0.500 | -0.603 |
| Max temperature | -0.866 | 0.531 | 0.286 | -0.017 |
| Min longitude | 0.190 | 0.833 | 0.770 | 0.01 |
| Max longitude | 0.268 | 0.871 | 0.846 | -0.128 |
| Relative humidty | -0.168 | -0.480 | -0.644 | 1.000* |

Appendix 12: Correlation among the morphological traits of fruits and seed of *Tetrapleura tetraptera*

*Significant P < 0.05

| Source of variation | Df | Ms | F-val | P-val | |
|------------------------|------|---------|--------|--------------|---|
| Source | 3 | 2804.5 | 36.67 | 0.000^{*} | — |
| Time | 11 | 57106.8 | 746.76 | 0.000^{*} | |
| Source*Time | 33 | 87.8 | 1.15 | 0.258_{ns} | |
| Error | 2352 | 76.5 | | | |
| Total | 2399 | | | | |

Appendix 13: Analysis of variance (ANOVA) for effect of source on height of *Tetrapleura tetraptera*

*Significant at P<0.05; ns- not significant (P>0.05)

| Appendix 14: Analysis of variance (ANOVA) for effect of source on collar diameter of |
|--|
| Tetrapleura tetraptera |

| Source of variation | Df | Ms | F-val | P-val |
|------------------------|------|---------|--------|---------------------|
| Source | 3 | 5.316 | 10.42 | 0.000* |
| Time | 11 | 258.188 | 506.02 | 0.000^* |
| Source*Time | 33 | 0.335 | 0.66 | 0.934 ^{ns} |
| Error | 2352 | 0.510 | | |
| Total | 2399 | | | |

*Significant at P<0.05; ns- not significant (P>0.05)

| Source of variation | Df | Ms | F-val | P-val |
|------------------------|------|---------|--------|-------------|
| Source | 3 | 216.57 | 40.20 | 0.000^{*} |
| Time | 11 | 1255.43 | 233.04 | 0.000^* |
| Source*Time | 33 | 10.27 | 1.91 | 0.001* |
| Error | 2352 | 5.39 | | |
| Total | 2399 | | | |

Appendix 15: Analysis of variance (ANOVA) for effect of source on number of leaves of *Tetrapleura tetraptera*

*Significant at P<0.05; ns- not significant (P>0.05)

Appendix 16: Correlation among the fruits and seedling morphological traits of *Tetrapleura tetraptera*

| | Seed | Seedling | Collar | Leaves | Fruit | Fruit |
|----------|--------|----------|----------|--------|--------|--------|
| | weight | height | diameter | | length | width |
| Seedling | 0.683 | | | | | |
| height | | | | | | |
| Collar | 0.300 | 0.932* | | | | |
| diameter | | | | | | |
| Leaves | 0.704 | 0.740* | 0.848* | | | |
| Fruit | -0.045 | -0.786 | 0.713 | 0.561 | | |
| length | | | | | | |
| Fruit | -0.463 | -0.172 | 0.262 | -0.098 | -0.453 | |
| width | | | | | | |
| Number | 0.954* | 0.435 | 0.368 | 0.757 | 0.206 | -0.629 |
| of seeds | | | | | | |

*Significant P < 0.05

| Source of variation | Df | Ms | F-cal | P-val |
|------------------------|-----|---------|-------|---------------------|
| Source | 3 | 7.322 | 1.6 | 0.198* |
| Harvest | 4 | 115.059 | 25.16 | 0.000^{*} |
| Source*Harvest | 12 | 2.518 | 0.55 | 0.873 ^{ns} |
| Error | 60 | 4.573 | | |
| Total | 479 | | | |

Appendix 17: Analysis of variance for root dry weight of *Tetrapleura tetraptera* seedlings

*Significant at P<0.05; ns- not significant (P>0.05)

Appendix 18: Analysis of variance for shoot dry weight of *Tetrapleura tetraptera* seedlings

| Source of variation | Df | Ms | F-val | P-val |
|------------------------|-----|---------|-------|---------------------|
| Source | 3 | 1.73 | 0.37 | 0.777* |
| Harvest | 4 | 563.493 | 119.7 | 0.000* |
| Source*Harvest | 12 | 0.608 | 0.13 | 1.000 _{ns} |
| Error | 60 | 4.707 | | |
| Total | 479 | | | |

*Significant at P<0.05; ns- not significant (P>0.05)

| Source of | Df | Ms | F-val | P-val |
|----------------|-----|---------|--------|---------------------|
| variation | | | | |
| Source | 3 | 18.05 | 1.62 | 0.194 ^{ns} |
| Harvest | 4 | 1209.03 | 108.61 | 0.000^{*} |
| Source*Harvest | 12 | 5.62 | 0.5 | 0.904 ^{ns} |
| Error | 60 | 11.13 | | |
| Total | 479 | | | |

Appendix 19: Analysis of variance (ANOVA) for biomass accumulation of *Tetrapleura tetraptera* seedlings

*Significant at P<0.05; ns- not significant (P>0.05)

| Appendix 20: Total biomass (g) of <i>T. tetraptera</i> seedlings ac | cross the assessment period |
|---|-----------------------------|
| (mean ±sd) | |

| | | | Weeks | | |
|--------|-------------------------|-------------------|-----------------|--------------------|--------------------------|
| Source | 4 | 8 | 12 | 16 | 24 |
| Ibadan | $0.40_{a} \!\pm\! 0.01$ | $3.28\pm\!0.59$ | 9.64 ± 3.27 | $9.72\pm\!\!1.78$ | $26.2_a \pm 10.17$ |
| Aponmu | $0.60_{b}\pm 0.02$ | 3.53 ±1.15 | 6.72 ±2.11 | 8.54 ± 1.19 | 21.79 _a ±5.11 |
| Iwo | $0.41_{b}\pm 0.02$ | $2.67\pm\!\!0.41$ | 6.98 ± 2.59 | 7.77 ± 1.76 | $20.3_{a}\pm 3.50$ |
| Mamu | $0.28_{c}\pm 0.00$ | $3.00\pm\!\!0.31$ | 7.12 ± 2.72 | $10.86\pm\!\!2.70$ | $23.12_{a}\pm 5.88$ |
| | | | | | |

| | | Weeks | | | | |
|----------------|------|-------|------|------|------|--|
| Source/harvest | 4 | 8 | 12 | 16 | 24 | |
| Aponmu | 3.33 | 32.7 | 31.4 | 14.0 | 23.4 | |
| Ibadan | 3.54 | 18 | 33.9 | 24.9 | 38.8 | |
| Iwo | 5.97 | 15.4 | 37.1 | 18.3 | 17.2 | |
| Mamu | 0.00 | 10.2 | 38.2 | 22.7 | 25.4 | |
| Inter-source | 26.3 | 22.7 | 35.6 | 22.9 | 27.8 | |

Appendix 21: Coefficient of variation (%) in biomass accumulation of *Tetrapleura* tetraptera

Appendix 22: Relative growth rate of *Tetrapleura tetraptera* seedlings

| | | Relative growth rate (g/month) | | | | |
|---------|--------|--------------------------------|--------|--------|--|--|
| Sources | 2 – 4 | 4 -6 | 6 – 8 | 8 – 12 | | |
| Aponmu | 1.8827 | 0.5788 | 0.2916 | 0.3844 | | |
| Ibadan | 2.2458 | 1.0801 | 0.0153 | 0.4228 | | |
| Iwo | 1.9868 | 0.8764 | 0.1137 | 0.4392 | | |
| Mamu | 2.4137 | 0.8504 | 0.4359 | 0.4605 | | |

Appendix 23: Absolute growth rate of *T. tetraptera* seedlings

| Sources | | Absolute growt | h rate (g/month) | |
|---------|-------|----------------|------------------|--------|
| | 2 – 4 | 4 - 6 | 6 - 8 | 8 – 12 |
| Aponmu | 1.475 | 1.595 | 0.905 | 3.245 |
| Ibadan | 1.440 | 3.180 | 0.110 | 3.483 |
| Iwo | 1.120 | 2.155 | 0.395 | 3.068 |
| Mamu | 1.355 | 2.055 | 1.875 | 2.678 |

| 0.038* |
|--------|
| 0.000 |
| 0.889 |
| |
| |
| |

Appendix 24: Analysis of variance for leaf area of *Tetrapleura tetraptera*

| Variables | Df | Ms | F-cal | P value |
|--|-----|---------------------|-------|---------------------|
| | | No of roots | | |
| Hormone_type | 2 | 4.014 | 0.732 | 0.483_{ns} |
| Hormone concentrations | 2 | 12.181 | 2.220 | 0.113 _{ns} |
| Hormone_type * | 4 | 6.972 | 1.271 | 0.286_{ns} |
| Hormone_concentrations | | | | |
| Error | 109 | 5.486 | | |
| Total | 120 | | | |
| | | Root length | | |
| Hormone_type | 2 | 10.413 | 3.940 | 0.022* |
| Hormone concentrations | 2 | 11.660 | 4.412 | 0.014* |
| Hormone_type * Hormone_concentrations | 4 | 6.746 | 2.552 | 0.043* |
| Error | 109 | 2.643 | | |
| Total | 120 | | | |
| | Lei | igth of longest roo | ot | |
| Hormone_type | 2 | 34.250 | 4.828 | 0.010* |
| Hormone concentrations | 2 | 9.695 | 1.367 | 0.259_{ns} |
| Hormone_type * Hormone_concentrations | 4 | 8.400 | 1.184 | 0.322 _{ns} |
| Error | 109 | 7.093 | | |
| Total | 120 | | | |
| | | No of shoots | | |
| Hormone_type | 2 | 0.483 | 0.326 | 0.722_{ns} |
| Hormone concentrations | 2 | 7.097 | 4.786 | 0.010* |
| Hormone_type * Hormone concentrations | 4 | 0.847 | 0.571 | 0.684_{ns} |
| Error | 109 | 1.483 | | |
| Total | 120 | | | |

Appendix 25: Analysis of variance (ANOVA) results for number of roots, length of roots and length of longest root observed on juvenile stem cuttings of T. tetraptera after sixty days

Note: *significant at P < 0.05; ns = not significant at P < 0.05

| Variables | Df | Ms | F-cal | P value |
|---------------------------|-----|--------------------|-------|---------------------|
| | | Height of shoot | | |
| Hormone type | 2 | 16.213 | 1.816 | 0.168 _{ns} |
| Hormone concentrations | 2 | 8.700 | 0.974 | 0.381 _{ns} |
| Hormone type * | | | | |
| Hormone | 4 | 36.158 | 4.049 | 0.004* |
| concentrations | | | | |
| Error | 109 | 8.930 | | |
| Total | 120 | | | |
| | | No. of leaves | | |
| Hormone type | 2 | 8.506 | 4.707 | 0.011* |
| Hormone concentrations | 2 | 4.090 | 2.264 | 0.109 _{ns} |
| Hormone type * | | | | |
| Hormone | 4 | 4.572 | 2.530 | 0.045* |
| concentrations | | | | |
| Error | 109 | 1.807 | | |
| Total | 120 | | | |
| | Р | ercentage survival | | |
| Hormone types | 2 | 466.667 | 1.478 | 0.253 _{ns} |
| Hormone | r | 200.000 | 0 622 | 0.542 |
| concentrations | 2 | 200.000 | 0.633 | 0.542_{ns} |
| Hormone types * | 4 | 166 667 | 0.529 | 0 717 |
| concentrations | 4 | 166.667 | 0.528 | 0.717 _{ns} |
| Error | 19 | 315.789 | | |
| Total | 30 | | | |

Appendix 26: Analysis of variance (ANOVA) results for height of shoots, number of leaves and percentage survival of juvenile stem cuttings of *T. tetraptera*

Note: *significant at P < 0.05; ns = not significant at P > 0.05

| Source of | Df | Ms | F-cal | P-value |
|--------------------|------|-----------|---------|----------------|
| variation | | | | |
| Percentage | | | | |
| survival | | | | |
| Sources | 3 | 11532.963 | 35.005 | 0.000* |
| Cuttings_Positions | 2 | 98207.778 | 298.084 | 0.000* |
| SOURCES * | (| 2125 105 | C 401 | 0.000 |
| Cuttings_Positions | 6 | 2135.185 | 6.481 | 0.000* |
| Error | 348 | 329.464 | | |
| Total | 360 | | | |
| No of roots | | | | |
| Sources | 3 | 87.522 | 55.582 | 0.000* |
| Cuttings_Positions | 2 | 689.897 | 438.127 | 0.000* |
| SOURCES * | ſ | 44 720 | 20.407 | 0.000 |
| Cuttings Positions | 6 | 44.730 | 28.407 | 0.000* |
| Error | 1428 | 1.575 | | |
| Total | 1440 | | | |
| Root length | | | | |
| SOURCES | 3 | 60.901 | 23.360 | 0.000* |
| Cuttings Positions | 2 | 1073.503 | 411.772 | 0.000* |
| SOURCES * | 6 | 22 220 | 0.500 | 0.000 |
| Cuttings Positions | 6 | 22.339 | 8.569 | 0.000* |
| Error | 1428 | 2.607 | | |
| Total | 1440 | | | |
| Length of longest | | | | |
| root | | | | |
| SOURCES | 3 | 106.496 | 26.081 | 0.000* |
| Cuttings_Positions | 2 | 1718.529 | 420.864 | 0.000* |
| SOURCES * | C | 44 227 | 10.957 | 0.000 |
| Cuttings_Positions | 6 | 44.327 | 10.856 | 0.000* |
| Error | 1428 | 4.083 | | |
| Total | 1440 | | | |

Appendix 27. Analysis of variance of the effects of sources and cutting positions on the percentage survival, number of roots, length of root and length of longest root of juvenile stem cuttings of *Tetrapleura tetraptera*

Note: *significant at P < 0.05; ns = not significant at P < 0.05

| Source of | Df | Ms | F cal | P – value |
|--------------------|------|----------------|---------|-----------|
| variation | | Na af ab a sta | | |
| | | No of shoots | | |
| SOURCES | 3 | 17.290 | 22.386 | 0.000* |
| Cuttings_Positions | 2 | 260.696 | 337.548 | 0.000* |
| Sources * | C | 6 224 | 8.072 | 0.000 |
| Cuttings_Positions | 6 | 6.234 | 8.072 | 0.000* |
| Error | 1427 | 0.772 | | |
| Total | 1439 | | | |
| | | Shoot length | | |
| SOURCES | 3 | 159.699 | 27.551 | 0.000* |
| Cuttings_Positions | 2 | 3095.159 | 533.968 | 0.000* |
| Sources * | 6 | 27.750 | 6 51 4 | 0.000 |
| Cuttings_Positions | 6 | 37.759 | 6.514 | 0.000* |
| Error | 1428 | 5.797 | | |
| Total | 1440 | | | |
| | | No of leaves | | |
| SOURCES | 3 | 55.390 | 36.941 | 0.000* |
| Cuttings_Positions | 2 | 879.855 | 586.799 | 0.000* |
| Sources * | ~ | | - 026 | 0.000 |
| Cuttings_Positions | 6 | 11.750 | 7.836 | 0.000* |
| Error | 1428 | 1.499 | | |
| Total | 1440 | | | |

Appendix 28: Analysis of variance of the effects of sources and cutting positions on the number of shoots, length of shoots and number of leaves of juvenile stem cuttings of *Tetrapleura tetraptera*

Note: *significant at P < 0.05; ns = not significant at P < 0.05

| | AGR | RGR | Survival | Rt no. | Root lt | LLR | Sht no | Sht lt | Leaves |
|----------|--------|--------|----------|--------|---------|--------|--------|--------|--------|
| RGR | 0.976* | | | | | | | | |
| Survival | 0.989* | 0.997* | | | | | | | |
| Rt no. | 0.982* | 0.918 | 0.946* | | | | | | |
| Rt lt | 0.929 | 0.943 | 0.946* | 0.899 | | | | | |
| LLR | 0.99* | 0.947* | 0.968* | 0.993* | 0.944 | | | | |
| Sht no | 0.95* | 0.96* | 0.965* | 0.913 | 0.988* | 0.955* | | | |
| Sht lt | 0.97* | 0.99* | 0.992* | 0.908 | 0.909 | 0.932 | 0.93 | | |
| Leaves | 0.97* | 0.99* | 0.989* | 0.902 | 0.900 | 0.925 | 0.93 | 1.00* | |
| SDW | 0.67 | 0.56 | 0.59 | 0.712 | 0.356 | 0.636 | 0.403 | 0.607 | 0.611 |

Appendix 29: Pearson correlation coefficients (r) of rooting variables and stockplant growth characteristics of *Tetrapleura tetraptera*

*AGR- Absolute growth rate, RGR- Relative growth rate, Rt no- Number of roots, LLR-Length of longest root, Sht no- Shoot number.

| Treatment | Root length per cuttings (cm) | Length of longest root (cm) | No. of leaves |
|-----------|----------------------------------|--------------------------------|-------------------------|
| NAA | 2.48 ^a ±1.61 | 2.65 ^a ±2.14 | $2.65^{a}\pm 1.69$ |
| IBA+NAA | $3.34^{ab}\pm1.5$ | 4.75 ^b ±2.44 | $3.41^{ab}\pm 1.23$ |
| IBA | $3.77^{bc} \pm 1.45$ | $4.70^{b}\pm1.91$ | $3.83^{b}\pm 0.99$ |
| Control | 4.56 ^{cd} ±2.27 | $6.84^{\circ} \pm 4.12$ | 3.51 ^b ±1.82 |
| Coconut | $5.02^{d} \pm 1.61$ | $6.80^{\circ} \pm 2.19$ | $3.51^{b} \pm 1.02$ |

Appendix 30. Effect of growth regulators and concentration on juvenile stem cuttings of *Tetrapleura tetraptera*

Note: *significant at P < 0.05; ns = not significant at P > 0.05

| Appendix 31. Effect of growth regulator concentration on stem cuttings of <i>Tetrapleura</i> |
|--|
| tetraptera (mean ±sd) |

| Concentration | Root length per cutting (cm) |
|---------------|------------------------------|
| 0 ppm | $4.56^{a} \pm 2.27$ |
| 100 ppm | $2.60^{\circ} \pm 0.98$ |
| 150 ppm | $3.96^{ab} \pm 1.52$ |
| 200 ppm | $3.04^{bc} \pm 1.90$ |

Note: *significant at P < 0.05; ns = not significant at P > 0.05

| Sources | No of roots | Root length | Length of |
|-------------------|-----------------|--------------------|-----------------|
| | | (cm) | longest root |
| | | | (cm) |
| Aponmu | 0.62 ± 1.45 | 1.17 ± 2.21 | 1.25 ± 2.49 |
| Mamu | 1.52 ± 2.32 | 1.56 ± 2.16 | 2.05 ± 3.05 |
| Iwo | 0.58 ± 1.38 | 0.87 ± 1.86 | 1.08 ± 2.42 |
| Ibadan | 0.41 ± 1.22 | 0.59 ± 1.91 | 0.77 ± 2.29 |
| Cutting positions | | | |
| Upper | 0.04 ± 0.3 | 0.03 ± 0.29 | 0.03 ± 0.28 |
| Middle | 0.2 ± 0.69 | 0.34 ± 1.19 | 0.36 ± 1.25 |
| Basal | 2.19 ± 2.28 | 2.76 ± 2.63 | 3.46 ± 3.43 |
| | | | |

| Appendix 32. Effect of stockplant source and cutting position on juvenile stem cuttings | |
|---|--|
| of <i>Tetrapleura tetraptera</i> (mean ±sd) | |