PROPAGATION AND REGENERATION OF IMPORTANT INDIGENOUS TREE SPECIES IN KAKAMEGA FOREST KENYA

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DECLARATION AND RECOMMENDATION

Declaration

This research thesis is my original work and has not been presented for the award of degree in any University and that all the sources used herein have been acknowledged

Carolyne Busuru

NM11/2464/09

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Recommendation

This thesis has been submitted for examination with our approval as University Supervisors.

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This thesis is dedicated to my late sister Mildred and my inspirational mother Jane Busuru.

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ABSTRACT

The African cherry (*Prunus africana* (Hook.f.) Kalkm.) regenerates poorly both naturally and artificially because of its recalcitrant seed. There is therefore a great need to conserve this species to ensure it does not become extinct. This study evaluated regeneration potential of the African cherry, identified the appropriate stage of collecting seeds for propagation and suitable sowing media that gives optimum germination and sought to identify other tree species with potential commercial uses which could be used as alternative to P. *africana* and hence ease the exploitation pressure that it currently faces. An experiment was set to evaluate the regeneration potentials of *P. africana* Vis a Vis *Olea capensis* and *Croton megalocarpus*. It also identified the best stage of seed collection and sowing media that can give good germination results. The experimental design used was randomized complete block design (RCBD) in split plot arrangement with the main plots as the stage of seed collection while the subplots as the different media type replicated three times. Seeds were collected at two stages: when fruits are green and mature and when ripe. These seeds were prepared, germinated under the different media types and germination percent monitored. The media used included: {(soil: sand), (soil: sawdust), (sand: sawdust) in ratio 1:1}, {(soil only), (sand only), (sawdust only)} and the normal nursery media of (sand: soil) in the ratio 1:3 was used as a control. The study also identified the effects of site on natural regeneration where by a sample of 10 trees per species was identified and deliberate vegetative disturbance underneath was carried out, and regeneration monitored and compared to undisturbed sites. The data was collected on mean germination percent and statistical analyses conducted at 95% significant level. The results indicated that There was significant difference in the timing of the collection of seeds for *C. megalocarpus* ($F_{2, 60}$, f=24.47, P<0.001). A post-hoc test (Tukey test) showed that germination rate was lower in mature green seeds compared to the other two seed collection stages – mature ripe and stored seeds. There was significant seed timing effect (Chi-square test = 32.90, d. f= 2, $p < 0.001$) for *P. africana.* Germination rate was significantly lower in stored seeds compared to the other two seed collection stages i.e. mature green seeds and mature ripe. There was a significant 'medium' effect on the germination of *C. megalocarpus* ($F_{6, 62}$, f=4.84, p<0.001), *Prunus africana* (Chi-square test = 14.10, d. f= 6, p = 0.029) and *O. capensis* (Chi –square test = 18.33, d. f= 6, p = 0.005). Seeds in sand &sawdust 1:1 and sand had higher germination rate compared to those in sawdust. Soil sand 3:1 and soil had moderate germination rate. There was a significant site effect on *C. megalocarpus*, ($F_{1, 18}$, f=10.09, p=0.005), *P. africana* ($F_{1, 18}$, f=53.42, p=0.001) and O. *capensis* (W=155, p<0.001) among the three species. Disturbed sites had a higher wildling regeneration compared to non-disturbed site. From the results, it was concluded that the best seed for propagation of *P. africana* is seed freshly harvested; mature and ripe even without any pre-treatment, while *O. capensis* and *C. megalocarpus* seed need to be harvested and dried well before sowing. It is therefore recommended that *P. africana* seed should be sown using this medium-sand: sawdust 1:1 immediately after harvesting without pre-treatment for optimum germination per cent. The best medium recommended for optimum germination of seed of all the three species is sand: sawdust in the ratio 1:1. It is also recommended that more *P. africana* seedlings should be used to rehabilitate degraded areas of the natural forests due to its shade intolerance nature. Proper tending of the seedlings after germination should be advocated as well as protection of mature *P. africana* trees in the wild.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

ABBREVIATIONS AND ACRONYMS

ANOVA	Analysis of Variance
BPH	Benign Prostatic Hyperplasia
CITES	Convention on International Trade in Endangered Species of Fauna and Flora
ICRAF	International Centre for Research in Agroforestry
IUCN	International Union for Conservation of Nature (World Conservation Union)
KEFRI	Kenya Forestry Research Institute

CHAPTER ONE
INTRODUCTION

1.1 Background Information

The African cherry (*Prunus africana)* belongs to the subfamily Prunoideae of the Rosaceae family, it attains highest diversity in temperate regions (Clemente *et al.*, 2006). It is an evergreen hardwood tree with dark-brown longitudinal fissured bark and simple, thick, leathery, oval, leaves with pointed ends. It grows at 700-3000 m above sea level, up to a height of 40 m. It has creamy white flowers and produces black fleshy fruits resembling a cherry when ripe, which are eaten and dispersed by monkeys, birds and squirrels. Flowering occurs between November and February although sporadic flowering all over the year may be found in Kakamega Forest of Kenya (Orwa *et al.*, 2009). Therefore, seeds can be collected between March and August. It is long lived - up to 100 years. The unit for sowing is the depulped fruit (the stone). Germination normally takes place within 30-50 days (Orwa *et al.*, 2009). The seeds are desiccation sensitive (recalcitrant) and therefore only short-term storage is recommended in damp sawdust in a cool dry environment (Luke *et al.*, 2013).

In nature, a fruiting tree can produce thousands of seedlings (wildings) that can be collected and transplanted. However, it is often seen that the wildings do not transplant well. In Cameroon the species has been propagated from cuttings. Without the use of hormones, about 10% of the seedlings had rooted after three months (Hall *et al.,* 2000). This therefore indicates the difficulty in propagation. In terms of natural regeneration, this species shows low or sporadic recruitment (Ewusi *et al.*, 1992, Jimu L *et al.*, 2012).

Previous studies on the bark extracts of *P. africana* showed that they were effective in the treatment of benign prostatic hyperplasia (BPH) (Bombardelli and Morazzoni, 1997). In Kenya, *P. africana* is present in most forested areas above 1500m (Hall *et al*., 2000). Bark is harvested on privately owned forest land when it is converted to tea estates, resettlement lands, and other uses; harvest is forbidden from protected areas (Cunningham *et al*., 1997). The species is exported as dried bark, chipped bark, and timber. The collection of mature bark for this purpose has resulted in the species becoming endangered (IUCN, 2002). Harvesters remove too much of the bark in an unsustainable manner. Many trees are debarked up to the smallest branches and others felled with negative impact on the limited wild population of this tree species (Ndibi, 1996; Betti, 2008).

In the 1990s it was estimated that 35,000 trees were debarked annually (Cunningham and Mbenkum, 1993). Currently, *P. africana* bark is entirely collected from the wild, although attempts at cultivation are underway in Kenya (Dawson, 1997; Dawson *et al*., 2000).

From the beginning, the harvest has been known to be destructive (Ngengwe, 1996; Walter and Rakotonorina, 1995). By 1995, because of the growing international demand for the bark, it was included as an endangered species in Appendix II of CITES at the Ninth Conference of the Parties. It was also listed by IUCN as vulnerable (IUCN, 2002). Despite the protection afforded by these designations, however, *P. africana* remains Africa's most intensively exported medicinal plant species by volume (Cunningham *et al*., 2002).

Olea capensis is often a bushy shrub or a small to medium sized tree up to 10 m in height, but it may be much larger, occasionally reaching 40 m; occurring in bush, littoral scrub and evergreen forest. The fruits take about 6 months to ripen. Seed storage behaviour appears to be orthodox (Albrecht, 1993). Growth is reported fast in young plants but much slower in older ones. It is a shade-tolerant, pioneer species and a dominant forest tree (Orwa *et al.*, 2009).

Croton megalocarpus is a hardy and fast-growing tree that grows to 15-35 m with a distinctive layering of branches and a rather flat crown. It is a pioneer species and it is found growing in cleared parts of natural forests, forest margins or as a canopy tree. The species regenerates well through seedlings, and under favourable climatic conditions may sometimes become invasive (Maroyi, 2010). The seeds are extracted from the shell by cracking with a hammer or a stone. On average there are 1700 seeds/kg. (Maroyi, 2010) noted that after sowing, the seeds germinate within 35-45 days, attaining germination rates of 95% without any pretreatment.

1.2 Statement of the Problem

The three species; *Prunus africana*, *Olea capensis* and *Croton megalocarpus* are important multiple-use tree species with both local and international economic and medicinal value. The high demand has led to over-exploitation that has caused serious damage to wild populations leading to concerns on the long-term sustainability of harvesting and conservation of the tree species. Kakamega forest being the easternmost remnant of the Guinea-Congolian rain forest system is an important ecosystem that needs to be maintained. The African cherry (*Prunus africana*) regenerates poorly both artificially and naturally because of its recalcitrant seed and shade intolerant nature. However, high demand for *P. africana* has led to its over-exploitation due to its medicinal properties and its valuable timber. Currently, *P. africana* bark is entirely

collected from the wild, although attempts at cultivation are underway in Kenya. The species is therefore facing extinction threat and thus calls for conservation efforts. This is because neither the uncontrolled harvesting of *P. africana* populations, nor cultivation in people's farms as one of the alternatives, easy problems to solve. This is because the species takes over 15 years to bloom, bear fruit, and for the bark to attain an adequate thickness for harvesting. This study takes into consideration two other species for comparison purposes i.e. *O. capensis and C. megalocarpus.* These are multi-purpose trees and thus they have potential commercial importance which can be useful to ease the pressure that is currently faced by *P.africana*.

1.3 Research Objectives

1.3.1 Broad Objective

To document the regeneration potentials of *P. africana, Olea capensis and Croton megalocarpus* seeds in Kakamega forest and hence contribute to improved seed germination of the three species.

1.3.2 Specific Objectives

1. To determine the optimal timing of seed collection for propagation of *P. africana, O. capensis* and *C. megalocarpus.*
2. To determine the suitable media for optimum germination of *P. africana, O. capensis* and *C. megalocarpus* seeds in the nursery.
3. To compare propagation and regeneration of *P. africana* with that of *Olea capensis* and *Croton megalocarpus* in Kakamega forest.

1.4 Null Hypotheses

1. Optimal timing of seed collection has no effect on the germination potential for the seeds of the three species
2. There is no effect of sowing media on germination rate of the seeds.
3. There is no difference in propagation and regeneration of *Olea capensis, Croton megalocarpus and P. africana*.

1.5 Justification

Prunus africana is a hardwood multi-purpose tree. Its timber is excellent for a number of uses including making strong tool handles, making of bridge decks, window and door frames, and good for fuel wood. Its bark provides traditional medicine for both human and domestic animal ailments. It is also exported to Europe for preparation of drugs that treat BPH (Benign Prostatic

Hyperplasia), an increasingly common health problem in older men (Nyamai *et al.*, 2016). However, over-exploitation has led to listing of the species in appendix II of CITES as endangered. This is contrary to Kenya's journey towards prosperity which involves the building of a just and cohesive society, enjoying equitable social development in a clean and secure environment- Vision 2030. This species also plays an important role in the montane ecosystem and thus tree deaths from debarking affects the integrity of the forest and reduces food resources for rare birds (Cunningham and Mbenkum, 1993 and CITES, 1994). This study takes into consideration two other species for comparison purposes i.e. *O. capensis and C. megalocarpus.* This is both medium-sized multi-purpose trees and thus they have potential commercial importance. The bark leaves and roots of *C. megalocarpus* are useful in treatment of whooping cough, pneumonia and stomach problems. It also provides strong timber and building poles. The bark of *O. capensis* is useful in reduction of fever, treating venereal diseases and stomach problems. It also provides quality timber. The findings of this study therefore will contribute to new knowledge on propagation of *P. africana* and also help ease the exploitation pressure on *P. africana* as consumers will utilize alternative tree species. It is also hoped that the outcome of this study will contribute to a balance between exploitation, forest conservation and economic development. This will mean therefore sustainable utilization of the medicinal species.

1.6 Scope and limitation of the study

The study was done within the spatial dimensions of Isecheno forest station of kakamega south forest where *P. Africana, O. capensis* and *C. megalocarpus* is abundant. This study evaluated regeneration potential of the African cherry, identified the appropriate stage of collecting seeds for propagation and suitable sowing media that gives optimum germination and sought to identify other tree species with potential commercial uses which could be used as alternative to *P. africana and hence* ease the exploitation pressure that it currently faces. Therefore, experiments were set for *P. africana* vis avis *O. capensis* and *C. megalocarpus.* However, it should be noted that O. capensis did need seed in Kakamega forest during the year of study and therefore posed a challenge I the experiments that involved mature ripe and mature green seeds for propagation. Studies indicate that this species does not seed regularly, (Orwa *et al.*, 2009) observed that flowering of this species takes place only at irregular intervals of up to seven years in the late dry season. For experiments on the natural regeneration, the nature of disturbance underneath selected trees was clearance of vegetative material but no disturbance on the soil.

Only mean germination percent of the seedlings in the different media was studied and therefore, the study did not involve transplanting of the seedlings. Although seeds of *P. africana, O. capensis* and *C. megalocarpus* are dispersed in various ways, for instance they are eaten and dispersed by monkeys, birds and squirrels, this study limited itself to natural seed fall.

1.7 Definition of Terms

Benign Prostatic Hyperplasia - a condition that affects the prostate gland in men causing the flow of urine to be slower and less forceful.

Conservation - the act of using and protecting resources properly. Proper use of resources and strict compliance with the rules on conservation are the keys to protecting and conserving our limited resources.

Disturbance - forest disturbance is an event that causes changes in the structure and composition of the forest. In this proposal it is used to mean deliberate clearance of vegetation under the selected trees.

Enrichment Planting - is a technique for promoting artificial regeneration of forests in which seedlings of preferred timber trees are planted in the under-storey of existing logged-over forests and then given preferential treatment.

Hyperplasia - enlargement.

Propagation - the production of more plants by seeds, cuttings, grafting or other methods.

Prostate - a gland found between the bladder (where urine is stored) and the urethra (the tube urine passes through).

Prostate Cancer - a form of cancer that develops in the prostate.

Regeneration - the process of new trees growing again on land that was formerly wooded, whether naturally or as a result of planting schemes.

Vulnerable - Liable to injury and subject to be affected injuriously.

CHAPTER TWO
LITERATURE REVIEW

2.1 Ecology and Biology

2.1.1 *Prunus africana* (African cherry) – Family Rosaceae

Prunus africana is a member of the Rosaceae (subfamily Amygdaloideae), a family with its highest diversity in temperate regions (Cronquist, 1981). Although the relationships among the other subfamilies of Rosaceae and its purported sister groups are yet to be determined (Potter *et al*., 1999), the family is generally considered monophyletic (Judd *et al*., 1999). The genus *Prunus* is the largest member of the subfamily, and includes the commercially important cherry (*P. avium* L.), peach (*P. persica* L.), plum (*P. domestica* L.), and almond (*P. dulcis*. *P. africana* is the only member of the genus on the African continent and may be chemically distinctive.

Prunus africana is a medium–large canopy tree 30–40m in height. Young trees have smooth, reddish bark whereas older trees have dark, platy, resinous bark. Leaves are simple and alternate. They are evergreen but some fall prior to fruit development. Leaves, twigs, fruits, and bark emit a "cherry" odour when crushed, which is characteristic of the genus. The odor is due to cyanogenic glycosides (Fraser *et al*., 1996). The adult tree flowers between November and February, although sporadic inflorescences can be found all year round in some forests. The fruit develops within 4 to 6 months of pollination. The ellipsoidal fruit is about 7–8mm in width and 10–12mm in length and contains one or two small oval seeds (Friis 1992; Beentje 1994). There are 3400–6000 seeds per kilogram on average (Hong *et al*., 1998). The seed is spread by birds and this is shown by the sprouting seedlings under big trees on the forest floor.

Prunus africana has a wide distribution in Africa. It occurs in montane regions of central and southern Africa and the islands of Bioko, Sao-Tome, and Grande Comore (Kalkman, 1965). *P. africana* is most abundant in open areas along forest margins and in disturbed areas (Ndam, 1996) and is not shade-tolerant (Kiama and Kiyiapi, 2001). Ndam (1996) also found the most seedlings in forest gaps or fallow fields. This suggests that *P. africana* is a light-demanding, secondary-forest species. Recruitment is low or sporadic (Ewusi *et al*., 1992). Because of deforestation at lower elevations, *P. africana* is confined to distinct "forest islands" that differ genetically (Barker *et al*., 1994), with the Madagascar population being the most distinct (Martinelli *et al*., 1986). The tree occurs at altitudes between 1000 and 2500m in montane forests

(Sunderland and Tako, 1999). Distribution appears to be related to mean annual temperature and rainfall and/or cloud cover (Hall *et al*., 2000). Because of their relatively large areas of montane habitat, Cameroon and Madagascar contain the largest populations of the species. In Kenya, it is common in Mt. Kenya, Aberdares, Kakamega, and Cherangani Forests. It also occurs in Timboroa, Nandi, Tugen hills and western part of Mau Forest.

2.1.2 *Olea capensis* (Elgon teak) – Family Oleaceae

Olea capensis is often a bushy shrub or a small to medium sized tree up to 10 m in height, but it may be much larger, occasionally reaching 40 m; occurring in bush, littoral scrub and evergreen forest. Bark: light grey, becoming dark grey and vertically fissured with age; a characteristic blackish gum is exuded from bark wounds Fruits are ovoid, fleshy, up to 2 x 1 cm, when ripe they are somewhat succulent purplish drupes. Flowering in profusion apparently takes place only at irregular intervals of up to 7 years in the late dry season. The fruits take about 6 months to ripen. Seed storage behaviour appears to be orthodox. Growth is reported fast in young plants but much slower in older ones. Over the first 4 years a mean annual height of 1.1 m has been reported. It is a shade-tolerant, pioneer species and a dominant forest tree (Kiama and Kiyiapi, 2001).

2.1.3 *Croton megalocarpus* (Croton) – Family Euphorbiaceae

Croton megalocarpus is a hardy and fast-growing tree that grows to 15-35 m with a distinctive layering of branches and a rather flat crown. The bark is dark grey, rough, and crackling. In Kenya, seeds mature during October-November in central regions, and from January to March in Western regions. *C. megalocarpus* is a pioneer species and it is found growing in cleared parts of natural forests, forest margins or as a canopy tree. The species regenerates well through seedlings, and under favorable climatic conditions may sometimes become invasive. Young trees coppice well after pruning, but fruiting is unlikely with intensive pruning, such as in hedgerow management. In agroforestry systems, it is sometimes managed as scattered trees in farmland because of its open canopy and usefulness for mulching. The seeds are extracted from the shell by cracking with a hammer or a stone. On average there are 1700 seeds/kg. The seeds are dried in the sun to approximately 5-9% moisture content and thereafter can be stored up to 1 year at 3°C. After sowing, the seeds germinate within 35-45 days, attaining germination rates of 95% without any pretreatment (Maroyi, 2010).

2.1.4 Taxonomy and nomenclature of *Prunus africana*

Prunus africana is a member of the Rosaceae (subfamily Prunoideae), a family with its highest diversity in temperate regions (Clemente *et al.*, 2006). Although the relationships among the other subfamilies of Rosaceae and its purported sister groups are yet to be determined (Potter *et al*., 1999), the family is generally considered monophyletic (Judd *et al*., 1999). The genus *Prunus* is the largest member of the subfamily and includes the commercially important cherry (*P. avium* L.), peach (*P. persica* L.), plum (*P. domestica* L.), and almond (*P. dulcis*. *P. africana* is the only member of the genus on the African continent and may be chemically distinctive.

2.1.5 Botanical description

Prunus africana is a medium–large canopy tree 30–40m in height. Young trees have smooth, reddish bark whereas older trees have dark, platy, resinous bark. Leaves are simple and alternate. They are evergreen but some fall prior to fruit development. Leaves, twigs, fruits, and bark emit a "cherry" odour when crushed, which is characteristic of the genus. The odor is due to cyanogenic glycosides (Fraser *et al*., 1996).

2.1.6 Distribution and habitat

P. africana has a wide distribution in Africa. It occurs in montane regions of central and southern Africa and the islands of Bioko, Sao-Tome, and Grande Comore (Kalkman, 1965). *P. africana* is most abundant in open areas along forest margins and in disturbed areas (Ndam, 1996) and is not shade-tolerant (Kiama and Kiyiapi, 2001). Ndam (1996) also found the most seedlings in forest gaps or fallow fields. This suggests that *P. africana* is a light-demanding, secondary-forest species. Recruitment is low or sporadic (Ewusi *et al*., 1992). Because of deforestation at lower elevations, *P. africana* is confined to distinct "forest islands" that differ genetically (Barker *et al*., 1994), with the Madagascar population being the most distinct (Martinelli *et al*., 1986). The tree occurs at altitudes between 1000 and 2500m in montane forests (Sunderland and Tako, 1999). Distribution appears to be related to mean annual temperature and rainfall and/or cloud cover (Hall *et al*., 2000). Because of their relatively large areas of montane habitat, Cameroon and Madagascar contain the largest populations of the species. In Kenya, it is common in Mt. Kenya, Aberdares, Kakamega, and Cherangani Forests. It also occurs in Timboroa, Nandi, Tugen hills and western part of Mau Forest.

2.1.7 Fruit and Seed Physiology and Phenology

The adult tree flowers between November and February, although sporadic inflorescences can be found all year round in some forests. The fruit develops within 4 to 6 months of pollination. The ellipsoidal fruit is about 7–8mm in width and 10–12mm in length and contains one or two small oval seeds (Beentje 1994). At maturity the fruits are dark red or reddish-brown. The seeds are oval and delicate There are 3400–6000 seeds per kilogram on average (Hong *et al*., 1998). The seed is spread by birds and this is shown by the sprouting seedlings under big trees on the forest floor.

2.1.8 Regeneration, sowing and germination

The unit for sowing is the depulped fruit (the stone). Germination normally takes place within 30-50 days (Orwa *et al.*, 2009). In nature, a fruiting tree can produce thousands of seedlings (wildings) that can be collected and transplanted. However, it is often seen that the wildings do not transplant well. In Cameroon the species has been propagated from cuttings. Without the use of hormones, about 10% of the seedlings had rooted after three months (Hall *et al.,* 2000). This therefore indicates the difficulty in propagation. In terms of natural regeneration, this species shows low or sporadic recruitment (Ewusi *et al.*, 1992)

2.2 Uses

Prunus africana trees are an important part of the montane ecosystem. It should therefore be noted that tree deaths from bark stripping affects the integrity of the forest and reduces food resources for rare birds (Cunningham & Mbenkum, 1993 in CITES proposal, 1994). *P. africana* is a very useful tree both at local and global markets. It takes fifteen years for the bark to develop the active ingredients necessary for medicinal use (Longo, 1981). Traditional medicinal uses of the bark include the treatment of stomach aches, urinary and bladder infections, chest pain, malaria, and kidney disease. It also provides durable timber for different household purposes.

2.2.1. Timber

The durable timber of *P. africana* makes it a favoured of wood for household purposes, the type of use being dependent on tree diameter and form (straightness) and local lifestyle. Small trees are a source of axe and hoe handles (Kom area, NW Cameroon) (Nsom and Dick, 1992) and grinding pestles (Bwindi forest, Uganda) (Cunningham, 1992). In western Uganda, large *P. africana* trees are a popular source of “beer boats” for making banana beer (Cunningham, 1992).

In southern Africa the wood has been used for timber and wagon making (Palmer and Pitman, 1972).

2.2.2 Medicinal Uses

Prunus africana is widely used in traditional medicine in southern, east and central Africa (Jeanrenaud, 1991). The bark is not only used by traditional healers, but also by local people collecting their own medicinal extracts, including for use as a purgative for cattle (Kalkman, 1965). In the Mount Cameroon area, for example, 88% of people collect traditional medicines of which *P. africana* was the fourth most popular medicinal plant species amongst people interviewed around Mount Cameroon and was collected by 14% of households surveyed (Jeanrenaud, 1991). Similarly, it is an important medicine in the Ijim montane forest area, where it is used to treat malaria, stomach ache and fever (Nsom and Dick, 1992).

Prostate gland hypertrophy and the closely related but more serious condition, benign prostatic hyperplasia (BPH) are common diseases affecting older men worldwide. They are expected to become more common amongst the ageing male population of Western Europe and the USA. It is now expected that one out of every two men in western countries will live longer than 80 years, with the result that 88% have the chance of developing histologic evidence of BPH. In the USA, for example, a 40-year-old man has at least a 10% chance of needing surgery for BPH, and in a recent survey in Scotland, 30.2% of a random sample (492) of otherwise healthy men aged between 40 and 79 years had prostatic enlargement and symptoms of BPH. Treatments for this disorder include surgery, balloon dilation, hyperthermia (using urethral probes), phytotherapy and pharmaceuticals containing anti-androgens and 5-alpha reductase inhibitors. Although surgery is common and effective, it is expensive, can cause impotence and is potentially dangerous, with a 1-3% post-operative mortality. For this reason, medical therapy and phytotherapy are popular alternatives.

Nearly 30 years ago, *P. africana* bark extracts were identified and patented as active in the treatment of benign prostatic hypertrophy (Debat, 1966). Bark extracts contain fatty acids, sterols and pentacyclic terpenoids (Uberti *et al.*, 1990). The sitosterol glucoside content of *P. africana* bark is 11 mg 100g-1 bark (Longo and Tira, 1981). Extracts of *P. africana* bark have been shown to be effective on rats (Thieblot *et al.*, 1977) and in recent clinical trials conducted in Austria (Barlet *et al.*, 1990). Capsules containing bark extracts have been marketed in Europe (mainly Austria, France, Italy and Switzerland) for over 20 years. By contrast with an earlier, small

clinical trial which showed no significant difference between patients treated with "Tadenan" and a placebo (Donkervoort *et al.*, 1977), the recent, detailed clinical trial conducted by (Barlet *et al*.,1990) showed that 66% of patients treated with "Tadenan", containing *P. africana* bark extract, had improved urine flow compared to 31% with the placebo. Five patients out of the 263 patients involved in the study showed gastro- intestinal side effects.

2.2.3 Volume and Economic Value of *P. africana* Bark Trade

At least four European companies have interests in *P. africana* bark for medicinal purposes: Laboratoires Debat (France) and its subsidiary company Plantecam Medicam in Cameroon; Madaus (Germany, Spain); Prosynthese (France); Inverni della Beffa and Indena Spa (Italy). Bark is bought for 150-170 CFA kg-1 (US$ 0.6-0.7) in Cameroon and for 11 French francs kg-1 (US$ 2) from Kenya. Capsules containing the bark extract are marketed in Europe, a 15-tablet box costing US$ 7-8. The market value of this trade has been roughly estimated at US$ 150 million a year (Hamilton, 1992). The Italian companies import bark extract from Madagascar and the other European companies import processed or unprocessed bark from Cameroon, Kenya, Uganda and Zaire. Extract in tablets or capsules are marketed under two main trade names: "Tadenan", produced by Laboratoires Debat (France) and "Pygenil" produced by Indena Spa (Italy).

2.2.4 History of Exploitation of *Prunus africana*

For about 42 years, the African cherry has been used in the treatment of benign prostatic hyperplasia and other disorders (Richard *et al*., 2017). The bark, from which the treatment is derived, is entirely wild-collected. The major exporters of bark include Cameroon, Madagascar, Equatorial Guinea, and Kenya (Ingram *et al*., 2009). Groupe Fournier of France and Indena of Italy produce 86% of the world's bark extract, both for their own products and for the free market. Worldwide exports of dried bark in 2000 have been estimated at 1350–1525 metric tons per year, down from its peak of 3225 tons in 1997. Bark extracts (6370–7225 kg per year) are worth an estimated $4.36 million per year (Bodeker *et al*., 2014). In 2000, Plantecam, the largest bark exporter in Africa, closed its extraction factory in Cameroon, due to complex ecological, social, and economic factors. Wild-collection is no longer sustainable (and probably never was) where harvest seriously affects morbidity and mortality rates of harvested populations. Prunus can be considered at least as an endangered plant species in Cameroon according to population reduction as outlined in the IUCN check list for Non-Detriment Findings (IUCN, 2001). This

explains the ban pronounced on October 2007 by the European Commission on Cameroon's Prunus. The Prunus ban impacts both the economic operators and the local people for whom Prunus represents an important non-timber forest product. Cameroon was proposed for ban as there are concerns that some provisions regarding the sustainable harvesting of Prunus barks are not being fully met, (Betti and Ambara, 2011).

In Kenya, *P. africana* is present in most forested areas above 1500m (Hall *et al*., 2000). Bark is harvested on privately owned forest land when it is converted to tea estates, resettlement lands, and other uses; harvest is forbidden from protected areas (Cunningham *et al*., 1997). The species is exported as dried bark, chipped bark, and timber. Harvest began in 1995 with the export of 150 tons of bark. Export peaked at 500 tons in 1998 and about 300 tons were shipped in 2000. Only one exporter (Jonathan Leakey) has been involved in the *Prunus africana* trade in Kenya and he ships dried or chipped bark to Prosynthese (a subsidiary of Groupe Fournier of France).

The CITES management authority for Kenya objected to the continued harvest without a Detriment Study and halted the harvest at the end of 2002. Cultivation trials have been conducted, but large-scale plantations are not yet in production (Barker *et al*., 1994; Dawson, 1997). Mature trees are also exploited for their timber. Following harvest of mature trees for local and export timber products (Nzilani, 2001) examined the Kakamega and South Nandi forests in western Kenya. She found few saplings and young trees, suggesting poor recruitment resulting from the removal of mature trees.

2.2.5 Conservation Efforts of *P. africana*

Alternatives to wild-collection of *P. africana* must be investigated, including the implementation and enforcement of conservation measures as well as efforts to cultivate the species. Several aspects of conservation have therefore been considered as follows:

2.2.6 Cultivation

Over 35 years of wild-collections have led to serious resource depletion that appears to threaten the resource. Cultivation may be one mechanism to protect the resource baceause It is light demanding and responds well to cultivation (Hall *et al*., 2000; Vivien and Faure, 1985, Fraser *et al*., 1996; Tchouto, 1996). The Conservation through Cultivation Programme at the Limbe Botanic Garden in Cameroon and the International Centre for Research in Agroforestry (ICRAF) cultivation experiments in Kenya are leading cultivation efforts. (Cunningham and Mbenkum, 1993) recommended *P. africana* plantations at low altitude, especially in areas of abandoned oil

palm stands. However, trees at low altitudes are more likely to become infested with wood borers and fungal pathogens, seriously reducing timber value (Cunningham *et al*., 2002). Available land at higher altitudes is scarce, at least in Cameroon; nearly all lands outside of forest preserves have been converted to farm crops or pastures.

2.2.7 Enrichment plantings

Enrichment plantings have long been recommended as a way to replace the trees lost to harvest. In 1972, the Cameroon Office National des Eaux et For^ets (ONADEF) established an enrichment plantation in Ntingue near Dschang. Unfortunately, these forest reserve trees reportedly were recently debarked (Cunningham *et al*., 2002). The Forest Department of Kenya has started enrichment plantings for timber production (Dawson *et al*., 2000). No enrichment plantings are known for Madagascar but, considering that the species is genetically unique (Dawson and Powell, 1999), enrichment plantings may be critical where harvesting has removed mature trees.

Vegetative techniques, such as grafting and cuttings (Leakey, 1994) may be used in this case to speed the recovery in denuded Madagascar forests as well as other heavily harvested areas, such as the Bamenda Highlands of Cameroon. Where enrichment plantings occur, they should include opening the canopy and clearing the competitive undergrowth around reproductive trees (Ndam, 1996). However, plantings must not occur to the detriment of other forest species. With the large market potential for future bark sales, it could be tempting to manage a natural forest for *P. africana* production (Cunningham *et al*., 2002) conducted an economic analysis of enrichment plantings in Cameroon. Based on the historical annual bark volumes processed at Plantecam (1923 tons per year), they calculated that a 12-year-old stand on 820 ha with 1363 trees planted per hectare would supply enough bark for a 12-year rotation.

2.2.8 Ex situ conservation practices

Barker *et al*., (1994) and Dawson and Powell, (1999) showed the genetic uniqueness of each country's populations. Even within an area, the species demonstrates differences (Cunningham *et al*., 2002) discusses three different varieties in Cameroon reported by harvesters. Secure field gene banks have been recommended (Cunningham and Mbenkum, 1993; Cunningham *et al*., 2002) as a mechanism to conserve the different genetic strains. Further, the harvest of populations of *P. crassifolia*, possibly a separate species endemic to the Kivu region of the Democratic Republic of Congo, may cause severe population decline before science has a chance

to examine it. Thus, it is critical to establish secure field gene banks for established *P. africana* and *P. crassifolia* genotypes (Cunningham *et al*., 2002). The felling or girdling of large reproductive trees reduces seed production and possible recruitment (Stewart, 2001) documented the large numbers of seeds produced by large trees and her model simulations showed the importance of conserving large trees. A reduction in seed and animal dispersal may further isolate the montane populations. Storage of seed as a conservation strategy does not appear to be possible (Sunderland and Nkefor, 1996) demonstrated the recalcitrant nature of *P. africana* seed, which fails to germinate if stored for more than 18 months.

2.2.9 Convention of International Trade in Endangered Species (CITES) of Flora and Fauna

CITES is the principal international conservation strategy for the protection of internationally traded endangered species. In 1994 (effective in 1995), *P. africana* was included in Appendix II as an endangered species. Listing specifies that trade in wild and cultivated material must be licensed in exporting and importing countries. Difficulties in identification of the various *P. africana* products have been cited as a problem in its effective implementation, especially in importing countries (Cunningham *et al*., 1997). This has led to under-reporting of the trade. In addition, some countries lack independent scientific CITES Authorities, allowing exports to occur without a valid non-detriment finding. (Cunningham *et al*., 1997) made several recommendations to improve CITES oversight.

2.3 Elgon teak (*Olea capensis*) – Family Oleaceae

Olea capensis is often a bushy shrub or a small to medium sized tree up to 10 m in height, but it may be much larger, occasionally reaching 40 m; occurring in bush, littoral scrub and evergreen forest. Bark: light grey, becoming dark grey and vertically fissured with age; a characteristic blackish gum is exuded from bark wounds Fruits are ovoid, fleshy, up to 2 x 1 cm, when ripe they are somewhat succulent purplish drupes. Flowering in profusion apparently takes place only at irregular intervals of up to 7 years in the late dry season (Orwa *et al.*, 2009). The fruits take about 6 months to ripen. Seed storage behaviour appears to be orthodox (Albrecht, 1993). Growth is reported fast in young plants but much slower in older ones. Over the first 4 years a mean annual height of 1.1 m has been reported. It is a shade-tolerant, pioneer species and a dominant forest tree.

2.3.1 Uses of *O. capensis*

Medicinal Value - Bark is chewed or boiled to make tea that helps in reducing fever; treating venereal diseases and stomach problems.

Timber – The wood makes a fine, high quality timber. It has dark brown heartwood and is attractively figured, fine-grained, hard and heavy. It has been widely used in railway sleepers, wagon woods, bridge construction and for flooring blocks. It can also produce beautiful furniture (Neuwinger, 2000).

Fuel - A viable species for fuel wood from which excellent charcoal can be made.

Fodder - *O. capensis* is a useful fodder tree.

2.3.2 Croton (*Croton megalocarpus*) – Family Euphorbiaceae

Croton megalocarpus is a hardy and fast-growing tree that grows to 15-35 m with a distinctive layering of branches and a rather flat crown. The bark is dark grey, rough, and crackling. In Kenya, seeds mature during October-November in central regions, and from January to March in Western regions. *C. megalocarpus* is a pioneer species and it is found growing in cleared parts of natural forests, forest margins or as a canopy tree. The species regenerates well through seedlings, and under favourable climatic conditions may sometimes become invasive (Maroyi, 2010). Young trees coppice well after pruning, but fruiting is unlikely with intensive pruning, such as in hedgerow management. In agroforestry systems, it is sometimes managed as scattered trees in farmland because of its open canopy and usefulness for mulching. The seeds are extracted from the shell by cracking with a hammer or a stone. On average there are 1700 seeds/kg. The seeds are dried in the sun to approximately 5-9% moisture content and thereafter can be stored up to 1 year at 3°C. (Maroyi, 2010) noted that after sowing, the seeds germinate within 35-45 days, attaining germination rates of 95% without any pretreatment.

2.3.3 Uses of C. megalocarpus

Medicinal value - The bark is boiled to make tea which is taken for treatment of whooping cough and a remedy for worms. The leaves, roots, and bark are used to treat stomach problems and pneumonia.

Fodder - The seed finely ground is incorporated in poultry feeds, as its protein content is high (50%).

Fuel - Well-dried nuts are reportedly used in some areas together with charcoal in cooking stoves. The tree is also utilized for firewood.

Apiculture - This species produces a dark-ambered honey with strong flavour.

Timber - Wood is of medium weight, hard, termite-resistant, and strong; it is used for timber and building poles.

Shade - C. *megalocarpus* forms a flat crown and has horizontal layers of branches, which make it useful in providing light shade and serving as a windbreak.

Soil improver - Leaves have high levels of nitrogen and phosphorus and serve as a source of mulch, for instance, in coffee plantations.

Ornamental - Its conspicuous flowers make it suitable as an ornamental.

Boundary - As the species is not browsed by livestock; it is often used as a live hedge (Salatino *et al.*, 2007).

2.4 Research gaps on the Propagation and regeneration of *P. africana, O. capensis* and *C. megalocarpus*

Most studies on the propagation and regeneration of *P.africana, O.capensis* and *C.megalocarpus* have focused on the harvested, treated and stored seed. Less attention has been given to the stage of maturity for harvesting the seed for propagation. For instance, (Luke *et al*., 2013, Sacande *et al.*, 2004., Sunderland and Nkefor, 1996) demonstrated the recalcitrant nature of *P. africana* seed. On the other hand, (Orwa *et al.*, 2009) demonstrated the orthodox seed behaviour of *C. megalocarpus*. (Orwa *et al.*, 2009, Albrecht, 1993) noted that seed storage behaviour of *O. capensis* appears to be orthodox. Different studies have also been done to identify the ideal medium for seed germination but few have focused on the different ratio of mixing different medium types.

Studies indicate that the natural regeneration for *P. africana* species is poor (Jimu *et al*., 2012) and recruitment is low or sporadic (Ewusi *et al*., 1992). This is due to the fact that the species is not shade tolerant. The same was observed by (Stewart, 2003. Shanley *et al.*, 2002. Dawson *et al.*, 2000. However, several authors noted that the species is light demanding. (Hall et al., 2000; Vivien and Faure, 1985, Fraser *et al*., 1996; Tchouto, 1996) (Maroyi, 2010) noted that *C. megalocarpus* regenerates well and under favourable conditions may sometimes become invasive. On the other hand, *O. capensis* trees showed good regeneration and establishment away from adult trees. (Tsingalia and Nyongesa, 2010) noted too that *O.capensis* in Kakamega forest does not show any evidence of natural regeneration under the parent trees. It is a shade-tolerant, pioneer species and a dominant forest tree (Orwa *et al*., 2009). Despite these numerous research,

stage of seed maturity for propagation has been understudied. There has also been limited studies on the medium used for propagation and the site conditions for regeneration of these species.

2.5 Conceptual Framework of the study

The optimal timing of seed collection for propagation impacts on the germination results of a specific species. The results of germination are affected by the type of medium used for propagation. The natural regeneration of seedlings in the wild is dependent on the site conditions in a specific area. The conceptual framework (Figure 1) identifies and suggests the timing of seed collection, suitable media for germination of *Prunus africana*, *Croton megalocarpus* and *Olea capensis* that can improve the seed germination results. It also identifies the site conditions that affect the natural regeneration of the three species which is in affected by the quantity of light that penetrates the sites.

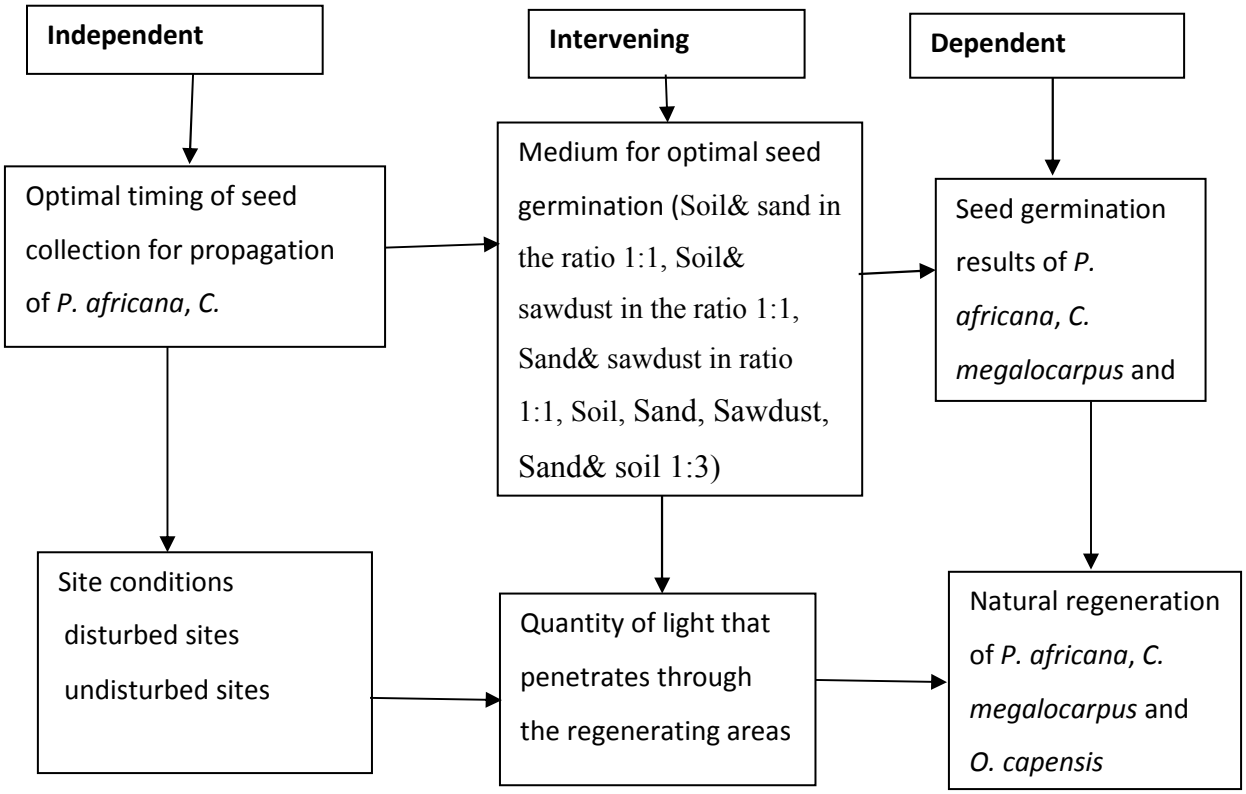

Figure 1 : Conceptual Framework for the study in Kakamega Forest

CHAPTER THREE
MATERIALS AND METHODS

3.1 Study Area

The study was carried out within the spatial dimensions of Isecheno Forest Station of Kakamega South Forest (Figure 2). Kakamega Forest is the easternmost remnant of the Guinea– Congolian rain forest system that once stretched virtually unbroken across much of equatorial and West Africa. The forest is situated in Western Kenya off Eldoret- Webuye road and about 8 kms from Kakamega town. It is located at GPS coordinates (Latitude: 0° 17' 18.00" N and Longitude 34° 51' 13.19" E) at an altitude of 1,400-2,300 meters and encompasses an area of 240 sq. Kms. Although Kakamega Forest is dominated by central African lowland species, it also contains highland species. This mix of lowland and highland species gives Kakamega Forest a unique variety of flora and fauna. Kakamega Forest possesses this assemblage of species because of its elevation and its proximity to the Nandi Forests which are east and south.

The rock formations are overlaid by a layer of clay-loam soils. The soils are dependent on the decomposition and reincorporation of dead organic matter. Fertility of the soils has dropped as wood has been increasingly removed by illegal loggers and local families. Kakamega Forest receives approximately 2000 mm of rain per year. This is nearly twice the rainfall of most temperate forests. Rainfall is heaviest in April and May (during the "long rains"), with a slightly drier June and a second peak of rain roughly in September to November (the "short rains"). January and February are the driest months. The temperature is fairly constant throughout the year, with mean daily minimums of about 11° C and mean daily maximums of about 26° C.

This reserve is an area that is famous for its abundance of bird and butterfly life. Up to 20% of all Kenyan plant and animal species occur only here, including 75% of all butterflies. In addition there are over 350 species of birds that reside in the forest. Several primate species are also present such as the Black and White Colobus Monkey, Blue Monkey, Olive Baboon, Red-tailed Monkey, and the rare De Brazza's Monkeys. Plants, and especially orchids, are found in extremely high diversity. Over 380 species of plant have been identified in Kakamega Forest.

Though the forest is listed as covering 240 km^2, indigenous forest accounts for only 100 km^2 with the remainder being composed of plantations, tea fields, and grasslands (Brooks *et al*., 1999; Wass, 1995).

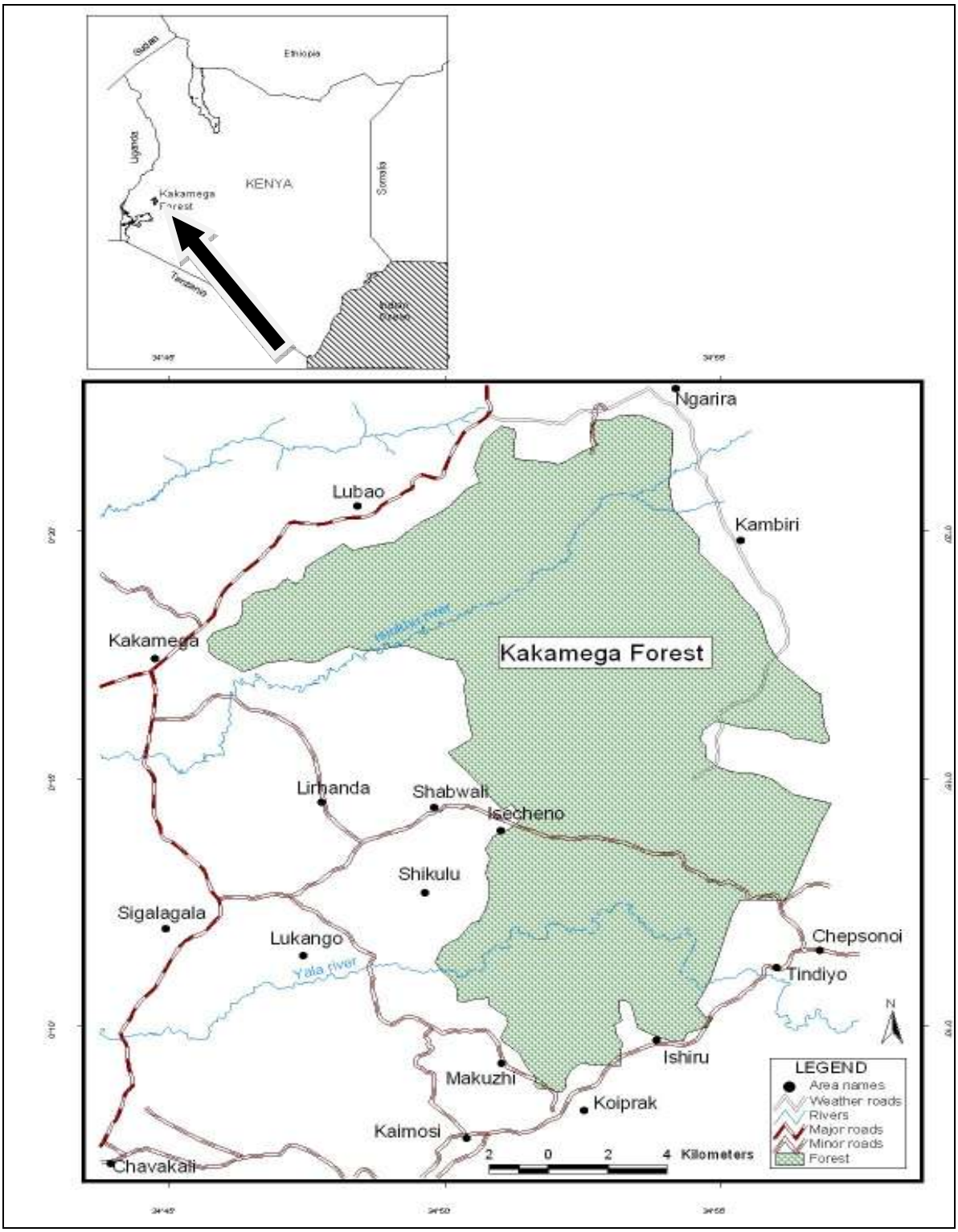

Figure 2: Location of the study site within Kenya (Source- Maina, G.M, 2011)

3.2 Artificial Regeneration for *P. africana* and *C. megalocarpus*.

3.2.1 Collection of Seed Material

Fruits of *P. africana and C. megalocarpus* were harvested from Kakamega forest with the help of KEFRI staff. The fruits harvested were classified as mature and green as well as those that were mature and ripe. A sample of 20 trees i.e. 10 trees per species were systematically selected for this exercise. Seed extraction was carried out for *C. megalocarpus* seeds before sowing. Although seeds of Olea capensis were also to be sown, the trees did not seed during the study period. Similarly, (Orwa *et al.*, 2009) observed that flowering of this species takes place only at irregular intervals of up to seven years in the late dry season.

Plate 1: *Prunus africana* seeds

Plate 2: Mature ripe seeds of *Prunus africana*

Plate 3: Green mature seeds of *Prunus africana*

Plate 4: Stored seeds of *Prunus africana*

Plate 5: Stored seeds of *Olea capensis*

3.2.2 The timing of seed collection for propagation in the different medium types

The seeds germinated were seeds from mature green fruits, those from ripe fruits and their germination potential were compared to that of harvested, processed and stored seeds. The experimental design was randomized complete block design (RCBD) in split plot arrangement with the main plots as the stage of seed collection while the subplots as the different media type replicated three times.

RCBD model: $Y_{ij} = \mu + A_i + B_j + \sum_{ij}$

$i = 1……7$

$j = 1…… 3$

Y_{ij} = observation of the i^{th} treatment and j^{th} block

μ = overall mean

A_i = effect of the ith media

B_j = effect of the jth seed stage

$\sum_{ij}$ = Random error component

The seeds were sown at a depth of twice their diameter on well- prepared seedbeds (explanation given below under experimental layout), containing different media types as mentioned below. A uniform number of seeds were sown as follows: 100 seeds per media, i.e., 1 x 1metre, 10 lines for each media and 10 seeds per line. The seeds were then covered lightly and light mulch spread over to keep them moist then watering was done regularly i.e. twice daily- in the morning and evening until seed emergence. Mulch was then removed immediately germination started. Germination percent was then recorded starting from emergence, then weekly for a period of five weeks where no more sprouting could be observed.

The media that was used included the following:

Treatments: Soil & sand in the ratio 1:1 (1wheelbarrow of forest soil to 1 wheelbarrow of fine sand), Soil & sawdust in the ratio 1:1, Sand & sawdust in ratio 1:1, Soil, Sand, Sawdust,

Control: Sand & soil in the ratio1:3

Plate 6: Preparation of sowing media

Germination beds were prepared through the following procedure: Germination beds were raised 15 cm above the ground level by the support of bricks. This was to prevent edges from being eroded away during watering. The size of the beds was 1m wide by 7m long. After demarcation, the beds were dug 30cm deep to remove ground roots, stones were placed inside then sand was spread on top. The 7m long beds were divided into seven potions of $1m^2$ so that each accommodates the different sowing media mixed in different ratio as indicated in plate 6 above. Each bed was then replicated 3 times. Table 1 below shows different media combinations that were used during seed propagation.

Table 1: The different media combinations that were used during seed propagation

Medium	Soil & sand	Soil & sawdust	Sand & sawdust	Sand & soil	Soil	Sand	Sawdust
Ratio	1:1	1:1	1:1	1:3			

The same experiment was repeated to make 7 germination beds as indicated below:

Table 2: Germination beds used for propagation of seed

Germination bed	**Species**	**Seed maturity stage**
1	*P. africana*	Mature green
2	*P. africana*	Mature ripe
3	*P. africana*	Stored seeds
4	*C. megalocarpus*	Mature green
5	*C. megalocarpus*	Mature ripe
6	*C. megalocarpus*	Stored seeds
7	*O. capensis*	Stored seeds

3.3 Natural Regeneration of *P. africana, O. capensis* and *C. megalocarpus*

Purposive sampling was used to select trees in Kakamega forest with the help of the Forester, Kakamega forest tour guides and forest rangers. A sample of 10 trees per species was used. The trees were marked as follows;

Table 3: Arrangement of the sample trees under disturbed conditions

Species	Tree number	site
P.africana	PD1, PD2, PD3, PD4, PD5, PD6, PD7, PD8, PD9, PD10.	Pakasinga Rondo Kisaina
C.megalocarpus	CD1, CD2, CD3, CD4, CD5, CD6, CD7, CD8, CD9, CD10.	Kisaina Musine circuit
O. capensis	OD1, OD2, OD3, OD4, OD5, OD6, OD7, OD8, OD9, OD10.	Ma Mutere trail Pakasinga

P - *P.africana*, C - *C.megalocarpus,* O - *O. capensis,* D – Disturbed

At the onset of rains, deliberate disturbance under these trees was done which involved removal of the vegetative matter underneath. The extent of clearing under the selected trees was a uniform radius of ten meters. Data was collected for a period of 13 weeks from the date of disturbance. Mean regeneration was monitored and compared to another sample of the same under undisturbed conditions.

Table 4: Arrangement of the sample trees under undisturbed conditions

Species	Tree number	site
P.africana	PUD1, PUD2, PUD3, PUD4 PUD5, PUD6, PUD7, PUD8 PUD9, PUD10	Ma Mutere trail Pakasinga Kisaina
C.megalocarpus	CUD1 CUD2, CUD3, CUD4, CUD5 CUD6, CUD7, CUD8 CUD9, CUD10	Ma Mutere trail Musine circuit Kisaina Rondo
O. capensis	OUD1, OUD2, OUD3 PUD4, OUD5, OUD6 OUD7,OUD8,OUD9, OUD10	Ma Mutere trail Pakasinga Kisaina

P - *P.africana,* C - *C.megalocarpus,* O - *O. capensis,* UD - undisturbed

Regeneration potential of *O. capensis* and *C. megalocarpus* was monitored and compared with that of *P. africana*. These two species have medicinal value and potential commercial use and thus if well regenerating, they can help ease the pressure that is currently on *P. africana*. Therefore, this exercise was done to minimize the risks as *P. africana* is being threatened to extinction.

3.4 Data analysis

Summary of data analysis is provided in Table 5. Analysis was carried out using Minitab and Excel. Descriptive statistics are presented in terms of means and standard deviation. Inferential test was also conducted where data was subjected to tests of normality and homogeneity of variance and where these assumptions were not met, they were log transformed. Comparisons were conducted using parametric and non-parametric tests (Analysis of Variance, Chi-square test, moods median tests and Mann-Whitney tests). Post hoc tests (Tukey tests) were also conducted to identify treatments that were significantly different.

Table 5: Data analysis conducted on each objective

Objective	**Data analysis tools**
Objective 1: To determine the optimal timing of seed collection for propagation of *P. africana, O. capensis* and *C. megalocarpus*	One-way ANOVA, moods median test, Chi-square test
Objective 2- To determine the suitable media for optimum germination of *P. africana, O. capensis* and *C. megalocarpus* seeds in the nursery	one-way ANOVA, moods median test, Chi-square test
Objective 3 - To compare propagation and regeneration of *P.africana* with that of *Olea capensis* and *Croton megalocarpus* in Kakamega forest.	one-way ANOVA, mann-whitney test

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Effects of seed maturity stage on germination

The seeds of the species harvested at different stages of maturity and sown responded differently in the different media types. It took 28 days for *P.africana* seed that were harvested when mature green to germinate in all the media types and recorded germination rates of 97%. It took *C.megalocarpus* seed 18 days to emerge and attained 52% germination rate. Seeds harvested when mature ripe, it took 25 days for *P. africana* seed to sprout in all the media types attaining germination rate of 97% while *C. megalocarpus* seed took 15 days to sprout with final mean germination rate of 95%. Stored seeds took 30 days for *P. africana* seed to sprout with a mean germination rate of 13%. On the other hand, *C.megalocarpus* seed took 15 days to sprout in all the media types with 95% germination rate. *O. capensis* seed took 46 days to sprout in all the media types with 90% germination rate (Fig. 4).

There was significant difference in the timing of the collection of seeds for *C. megalocarpus* ($F_{2, 60}$, f=24.47, P<0.001). A post-hoc test (Tukey test) showed that germination rate was lower in mature green seeds compared to the other two seed collection stages – mature ripe and stored seeds (Fig. 3). There was significant seed timing effect (Chi-square test = 32.90, d.f= 2, p < 0.001) for *P. africana.* Germination rate was significantly lower in stored seeds compared to the other two seed collection stages i.e mature green seeds and mature ripe (Fig. 4).

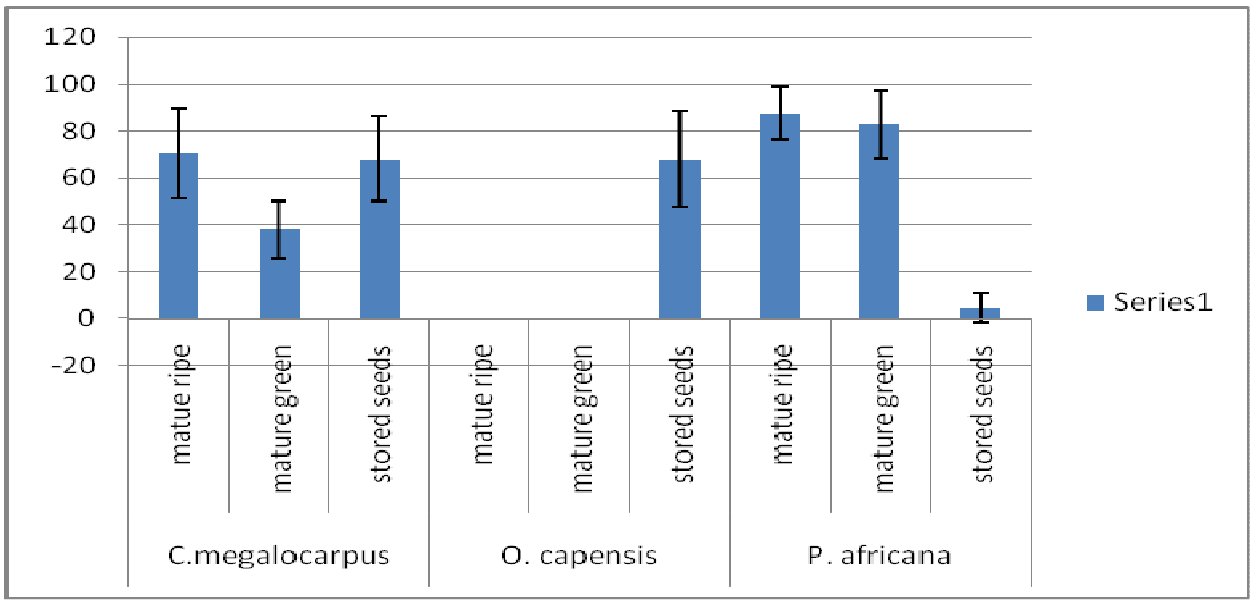

Figure 3: Germination rates for the species at different stages of seed maturity

Freshly harvested seed of *P. africana* germinates better than stored seed because the seed loses viability upon storage. Similarly, (Luke *et al.*, 2013, Sacande *et al.*, 2004., Sunderland and Nkefor, 1996) demonstrated the recalcitrant nature of *P. africana* seed. This study therefore is in agreement that this seed is indeed recalcitrant. On the other hand, good germination results of C. megalocarpus were attained from stored seed because of its orthodox seed behaviour (Orwa *et al.*, 2009). (Maroyi, 2010) noted that after sowing, *C. megalocarpus* seeds germinate within 35-45 days contrary to the results of this study whereby the seed germinated within 15- 18 days. (Orwa *et al.*, 2009, Albrecht, 1993) noted that seed storage behaviour of *O. capensis* appears to be orthodox. This was similarly noted because *O. capensis* stored seed attained germination rates of 90% from stored seeds.

4.2 Effect of media type on germination

Germination rates showed different results in the different 'medium' types. Generally, sawdust gave low germination rates while sawdust & sand (1:1) gave high germination rates. There was a significant 'medium' effect on the germination of *C. megalocarpus* ($F_{6,62}$, f=4.84, p<0.001), *Prunus africana* (Chi- square test = 14.10, d.f= 6, p = 0.029) and *O. capensis* (Chi –square test = 18.33, d.f= 6, p = 0.005). Seeds in sand &sawdust 1:1 and sand had higher germination rate compared to those in sawdust. Soil & sand 3:1 and soil had moderate germination rate (figure 4)

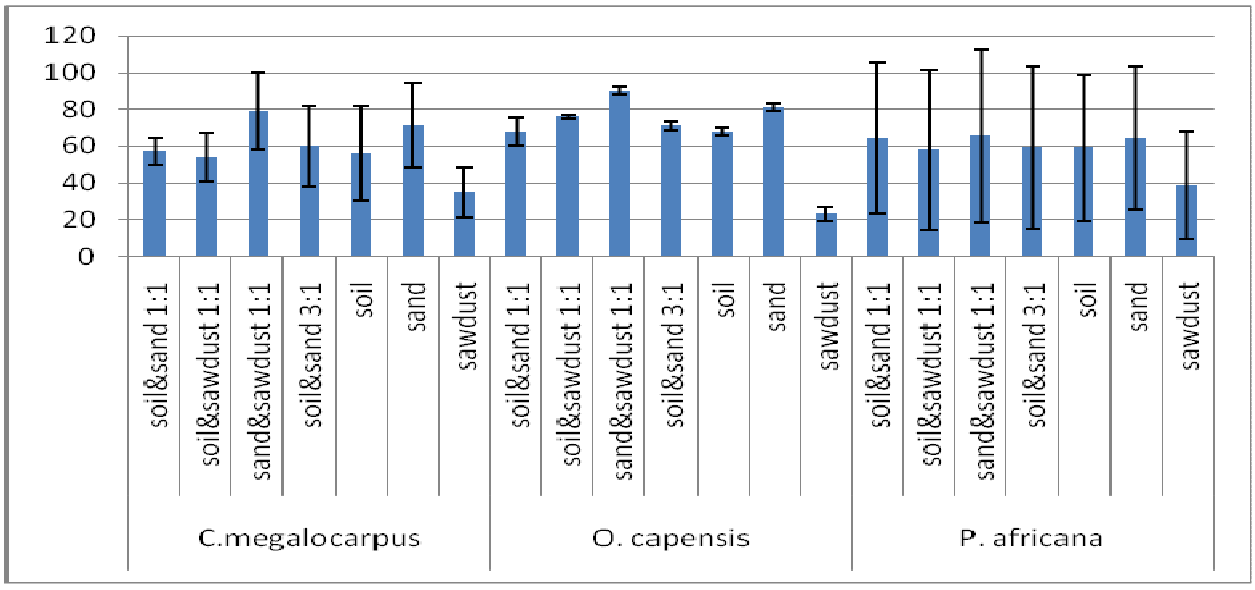

Figure 4: Germination rates per species in different media

The 'medium' that gave better germination results is sand: sawdust 1:1 while 'medium' sawdust gave poor germination results. This is contrary to (Tchoundjeu *et al.*, 2002) who indicated that the percentage of cuttings that rooted was significantly greater in sawdust compared to a mixture of sand and sawdust. Sand hastens germination through provision of heat. It is also free from contamination. Therefore, when mixed with sawdust, both play a role in that sand retains heat while sawdust retains moisture thus providing conducive environment necessary for germination.

4.3 Effects of site on natural regeneration

It was noted that a lot of wildings for the three species emerged in the forest. However, the rectruitment is poor especially for *P.africana*. However, *C.megalocarpus* showed good natural regeneration and *O.capensis* trees showed good regeneration and establishment away from adult trees. There was a significant site effect on C. megalocarpus, ($F_{1,\ 18}$, f=10.09, p=0.005), P.africana ($F_{1,18}$, f=53.42, p=0.001) and O. capensis (W=155, p<0.001) among the three species. Disturbed sites had a higher wildling regeneration compared to non-disturbed site (figure 5).

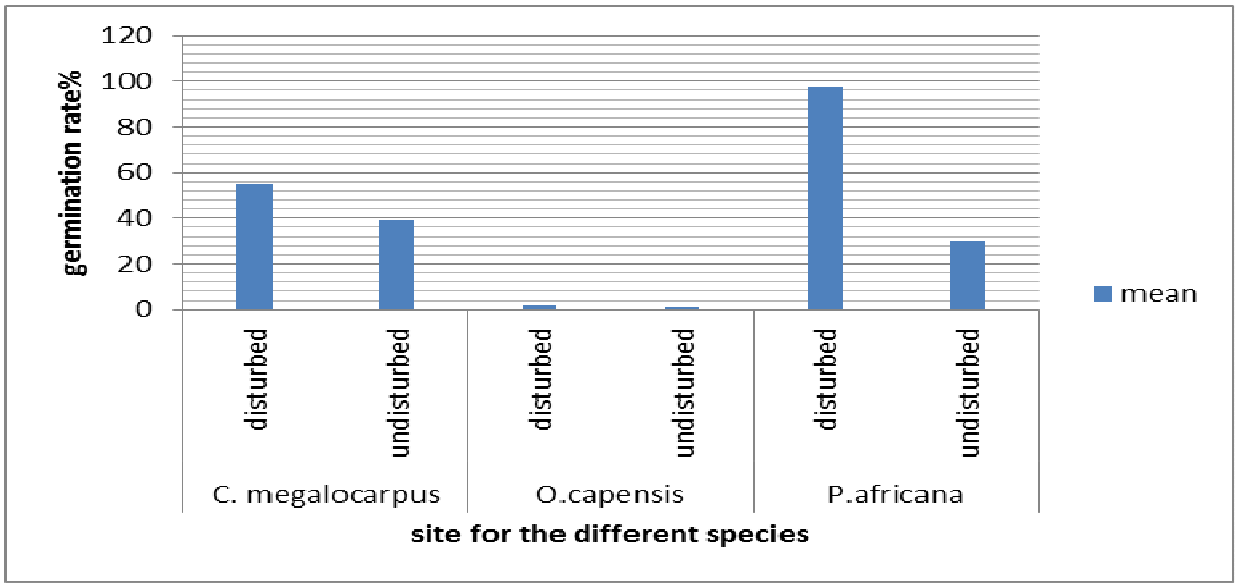

Figure 5: Regeneration rates for species under disturbed and undisturbed conditions

A lot of wildings for the three species emerged in the forest due to the natural seed fall from the mature trees and the favourable conditions for germination during the time of study i.e the rainy season. The disturbed sites indicated higher wildings as compared to the undisturbed sites as the clearing opened up provision for enough penetration of light for the sprouting of the seeds.

However, the natural regeneration for *P. africana* species is poor (Jimu *et al.*, 2012) and recruitment is low or sporadic (Ewusi *et al*., 1992). This is due to the fact that the species is not shade tolerant The same was observed by (Stewart, 2003. Shanley *et al.*, 2002. Dawson *et al.*, 2000.

Hall *et al*., (2000); Vivien and Faure, 1985, Fraser *et al*., 1996; Tchouto, 1996) who noted that the species is light demanding. However, *C. megalocarpus* showed good natural regeneration due to their shade tolerance characteristic. (Maroyi, 2010) similarly noted that *C. megalocarpus* regenerates well and under favourable conditions may sometimes become invasive. On the other hand, *O. capensis* trees showed good regeneration and establishment away from adult trees. (Tsingalia and Nyongesa, 2010) noted too that *O. capensis* in Kakamega forest does not show any evidence of natural regeneration under the parent trees. It is a shade-tolerant, pioneer species and a dominant forest tree (Orwa *et al.*, 2009).

CHAPTER FIVE
CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

Based on the evaluations carried out, *P.africana* exhibits good germination percent when propagated from seed freshly harvested when mature and ripe even without any pre-treatment. This is contrary to *C. megalocarpus* seed which does not germinate well when it is propagated from freshly harvested seed. Both *O. capensis* and *C. megalorcapus* germinate well from stored seed. The best medium for optimum germination of *O. capensis, P. africana* and *C. megalocarpus* is sand: sawdust in the ratio 1:1 whereas the medium that gives poor germination results is sawdust only. *P. africana* is not shade tolerant and although wildings are in plenty as observed in Kakamega forest, their recruitment is poor. However, *C. megalocarpus* and *O. capensis* are good natural regenerating species.

5.2 Recommendations

Based on the findings of the study, the following recommendations are made:

1. *P.africana* seed should be sown immediately after harvesting for optimum germination percent. On the other hand, *C. megalocarpus* seed should be harvested as fruits, dried thoroughly, extracted and sown. They can still be stored well and sown later.
2. Since *C. megalocarpus* is a fast growing species with its germination percent over 95%, it is recommended that local communities should be informed about its multi-purpose nature so as to ease the pressure currently on *P. africana*.
3. Since *O. capensis* does not seed yearly, it is recommended that the moment it seeds, a lot of it should be propagated in the nursery and wildings collected for seedling production.
4. The best medium recommended for optimum germination of seed of all the three species is sand: sawdust in the ratio 1:1.
5. Since natural regeneration of *P. africana* is poor inside the forest, it is recommended that wildings should be collected in large numbers, raised in the nurseries and used to rehabilitate degraded areas of the natural forests due to their shade intolerance nature.

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APPENDICES

Appendix A. Turkey tests on effect of seed stage on *C. megalocarpus*

Appendix A.1 Individual 95% CIs for Mean Based on Pooled Standard Deviation

Level	N	Mean	StDev	----+---------+---------+---------+-----
mature ripe	21	70.57	19.31	(-----*-----)
mature green	21	38.00	11.83	(-----*-----)
stored seeds	21	68.14	18.30	(-----*-----)
				----+---------+---------+---------+-----
				36 48 60 72

Pooled St Dev = 16.81

Tukey 95% Simultaneous Confidence Intervals

All Pairwise Comparisons among Levels of SEED STAGE (*C. megalocarpus*)

Individual confidence level = 98.07%

Appendix A. 2: SEED STAGE = mature ripe subtracted from:

SEED STAGE	Lower	Center	Upper	--------+---------+---------+---------+-
Mature green	-45.04	-32.57	-20.10	(----*----)
Stored seeds	-14.90	-2.43	10.04	(----*----)
				--------+---------+---------+---------+-
				-25 0 25 50

Appendix A.3 SEED STAGE = mature green subtracted from:

SEED STAGE	Lower	Center	Upper	--------+---------+---------+---------+-
Stored seeds	17.67	30.14	42.61	(----*----)
				--------+---------+---------+---------+-
				-25 0 25 50

Appendix B. Mood Median Test: SURVIVAL versus SEED STAGE (*P. africana*)

Mood median test for SURVIVAL_1

Chi-Square = 32.90 DF = 2 P = 0.000

Individual 95.0% CIs

SEED STAGE_1 N<= N> Median Q3-Q1 +---------+---------+---------+------

Mature ripe	3	18	90.0	7.5	(*)

Mature green 8 13 87.0 17.0 (--*-)
Stored seeds 21 0 3.0 8.5 (*)
+---------+---------+---------+------
0 30 60 90
Overall median = 81.0

Appendix C.ANOVA of media on *C. megalocarpus*

Appendix C.1 medium code used for data analysis

medium	**code**
Soil & sand 1:1	A
Soil & sawdust 1:1	B
Sand & sawdust 1:1	C
Soil & sand 3:1	D
soil	E
sand	F
sawdust	G

Appendix C.2 ANOVA of media on *C.megalocarpus*

Source DF SS MS F P
MEDIUM 6 10503 1751 4.84 0.000
Error 56 20274 362
Total 62 30777
S = 19.03 R-Sq = 34.13% R-Sq (adj) = 27.07%

Appendix C.3 Turkey tests on media **(***C. megalocarpus*)

Individual 95% CIs for Mean Based on
Pooled StDev
Level N Mean StDev ---------+---------+---------+---------+

A 9 57.00 7.35 (------*-----)
B 9 54.00 13.32 (-----*-----)
C 9 79.00 20.62 (------*-----)
D 9 60.00 21.94 (-----*-----)
E 9 56.00 26.03 (-----*-----)
F 9 71.33 23.08 (------*-----)
G 9 35.00 13.63 (------*-----)
---------+---------+---------+---------+
40 60 80 100
Pooled StDev = 19.03
Tukey 95% Simultaneous Confidence Intervals
All Pairwise Comparisons among Levels of MEDIUM
Individual confidence level = 99.66%

Appendix C.4 MEDIUM = A subtracted from:

MEDIUM Lower Center Upper --------+---------+---------+---------+-
B -30.40 -3.00 24.40 (------*------)
C -5.40 22.00 49.40 (------*-----)
D -24.40 3.00 30.40 (------*------)
E -28.40 -1.00 26.40 (------*------)
F -13.07 14.33 41.73 (------*-----)
G -49.40 -22.00 5.40 (------*-----)
--------+---------+---------+---------+-
-40 0 40 80

Appendix C.5 MEDIUM = B subtracted from:

MEDIUM Lower Center Upper --------+---------+---------+---------+-
C -2.40 25.00 52.40 (------*------)
D -21.40 6.00 33.40 (------*-----)

E	-25.40	2.00	29.40	(------*-----)
F	-10.07	17.33	44.73	(------*------)
G	-46.40	-19.00	8.40	(------*------)
	--------+---------+---------+---------+-			
	-40	0	40	80

Appendix C.6 MEDIUM = C subtracted from:

MEDIUM	Lower	Center	Upper	--------+---------+---------+---------+-
D	-46.40	-19.00	8.40	(------*------)
E	-50.40	-23.00	4.40	(------*------)
F	-35.07	-7.67	19.73	(------*------)
G	-71.40	-44.00	-16.60	(------*------)
	--------+---------+---------+---------+-			
	-40	0	40	80

Appendix C.7 MEDIUM = D subtracted from:

MEDIUM	Lower	Center	Upper	--------+---------+---------+---------+-
E	-31.40	-4.00	23.40	(------*------)
F	-16.07	11.33	38.73	(------*------)
G	-52.40	-25.00	2.40	(------*------)
	--------+---------+---------+---------+-			
	-40	0	40	80

Appendix C.8 MEDIUM = E subtracted from:

MEDIUM	Lower	Center	Upper	--------+---------+---------+---------+-
F	-12.07	15.33	42.73	(------*------)
G	-48.40	-21.00	6.40	(------*------)
	--------+---------+---------+---------+-			
	-40	0	40	80

Appendix C.9 MEDIUM = F subtracted from:

MEDIUM Lower Center Upper --------+---------+---------+---------+-

G -63.73 -36.33 -8.93 (------*------)

--------+---------+---------+---------+-

-40 0 40 80

Appendix D . Mood Median Test: SURVIVAL_1 versus MEDIUM_1 (*P. africana*)

Chi-Square = 14.10 DF = 6 P = 0.029

Individual 95.0% CIs

MEDIUM_1 N<= N> Median Q3-Q1 +---------+---------+---------+------

A 3 6 90.0 83.0 (-------------------------*)

B 5 4 80.0 91.5 (-------------------------*---)

C 3 6 96.0 94.5 (----------------------------*)

D 3 6 87.0 90.5 (--------------------------*)

E 6 3 77.0 84.5 (----------------------*----)

F 3 6 87.0 74.0 (----------------------*-)

G 9 0 53.0 61.5 (-----------------*--)

+---------+---------+---------+------

0 30 60 90

Overall median = 81.0

Appendix E. Mood Median Test: SURVIVAL_1_1 versus MEDIUM_1_1(*O. capensis*)

Mood median test for SURVIVAL_1_1

Chi-Square = 18.33 DF = 6 P = 0.005

Individual 95.0% CIs

MEDIUM_1_1 N<= N> Median Q3-Q1 +---------+---------+---------+------

A	2	1	69.0	15.0	(----*--)
B	0	3	76.0	2.0	*)
C	0	3	90.0	4.0	(*)
D	3	0	70.0	5.0	*-)
E	3	0	68.0	4.0	(*)

F 0 3 81.0 4.0 (*)
G 3 0 22.0 7.0 (*--)
+---------+---------+---------+------
20 40 60 80
Overall median = 74.0
* NOTE * Levels with < 6 observations have confidence < 95.0%

Appendix F: One-way ANOVA: WILDINGS versus GROUND (*c. megalocarpus*)

Source DF SS MS F P
GROUND 1 1217 1217 10.09 0.005
Error 18 2171 121
Total 19 3388
S = 10.98 R-Sq = 35.91% R-Sq(adj) = 32.35%

Appendix F.1: Individual 95% CIs for Mean Based on Pooled St. Dev

Level N Mean StDev +---------+---------+---------+---------
disturbed 10 55.10 12.41 (--------*--------)
undisturbed 10 39.50 9.34 (--------*--------)
+---------+---------+---------+---------
32.0 40.0 48.0 56.0
Pooled St Dev = 10.98

Appendix G: One-way ANOVA: WILDINGS_1_1 versus GROUND_1_1 (*p. africana*)

Source	DF	SS	MS	F	P
GROUND_1_1	1	22916	22916	53.42	0.000
Error	18	7722	429		
Total	19	30639			

S = 20.71 R-Sq = 74.80% R-Sq(adj) = 73.40%

Appendix G.1: Individual 95% CIs For Mean Based on Pooled St. Dev

Level	N	Mean	StDev	----+---------+---------+---------+-----
disturbed	10	97.50	23.33	(-----*-----)
undisturbed	10	29.80	17.71	(-----*----)
				----+---------+---------+---------+-----
				25 50 75 100

Appendix H: Mann-Whitney Test and CI: pWILDINGS_1_1, pWILDINGS_1_1_1 *(O. capensis*)

	N	Median
Disturbed	10	98.50
Undisturbed	10	31.50

Point estimate for ETA1-ETA2 is 68.50

95.5 Percent CI for ETA1-ETA2 is (46.01,88.00)

W = 155.0

Test of ETA1 = ETA2 vs ETA1 not = ETA2 is significant at 0.0002