KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY

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EFFECTS OF DESICCANTS, PACKAGING MATERIALS AND STORAGE PERIODS ON SEED QUALITY AND LONGEVITY DYNAMICS OF THREE INDIGENEOUS FOREST TREE SPECIES

A THESIS SUBMITTED TO THE SCHOOL OF GRADUATE STUDIES, KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY, IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF MASTER OF PHILOSOPHY (M. Phil. SEED SCIENCE AND TECHNOLOGY) DEGREE

BY

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DEDICATION

I dedicate this work to my wife, Mrs Juliet Tandoh, my children Maame Araba Tandoh and Paa Kwesi Tandoh who in diverse ways have made this dream a reality.

DECLARATION

I hereby declare that this work being submitted is my own original research work and that it has neither in part nor in whole been used for any degree elsewhere. All other works cited is duly acknowledged.

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ABSTRACT

This study was carried out to determine the effects of seed desiccants, packaging materials and storage periods on seed quality and longevity dynamics of three very important indigenous forest tree species. The experimental period was December, 2015 to June, 2016. Seed collection was done at the Bobiri Forest Reserve. The desiccation experiment was set up using a Randomized Complete Block Design (RCBD) with three (3) replications for each of the three species. The seed storage experiment was set up using 3 x 6 factorial arrangements in

Completely Randomized Design (CRD) with three replications for each of the three species. Seed storage experiment was conducted using six packaging materials (jute, nylon, paper, ziplock bag, airtight bottle and no packaging) with three storage periods (no storage, three months storage and six months storage). Germination percentage, seed vigour, 1000 seed weight, moisture content, seed health analysis, carbohydrate, protein and oil contents were assessed before storage, three and six months after storage. The study revealed that the beads dried the seeds of *Pericopsis elata* within 2 days, 3 days to dry *Sterculia rhinopetala* seeds and 12 days to *dry G. cedrata*, which was much faster than the rest of the desiccants without any deleterious effect on seed quality and longevity. *Pericopsis elata* and *Sterculia rhinopetala* showed orthodox seed storage behavior by surviving drying to a lower moisture content hence improving storability when packaged in airtight bottle (3.5%) and ziplock bags (3.5%) storage periods increased. The seed carbohydrates, proteins and oils for *Pericopsis elata* and *Sterculia rhinopetala* were maintained in the airtight packaging materials thereby improving seed storability. The dormancy of *Sterculia rhinopetala* was released by using GA3 with a germination percentage of 96.33%. Moist saw dust significantly extended the life span of the

Guarea cedrata seeds for at least 21 days. A total number of nine fungi isolates were identified during storage. The seed viability equations predicted that *P. elata* and *S. rhinopetala* seeds could be stored for 200 years and 177 years, respectively, after six months of storage. Seed viability dropped significantly (155 days for *P. elata* and 79 days for *S. rhinopetala*) after the accelerated aging test performed on the six-months old seeds. The study concluded that *P. elata* and *S. rhinopetala* could best be dried with beads at a faster rate and subsequently stored for a longer period using airtight packaging materials whilst *G. cedrata,* could be stored in a moist saw dust.

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TABLE OF CONTENT

CONTENTS **PAGE**

DEDICATION ..... **ii**
DECLARATION ..... **iii**
ABSTRACT ..... **iv**
ACKNOWLEDGEMENT ..... **vi**
LIST OF TABLES ..... **xiii**
ABBREVIATIONS AND ACRONYMS ..... **x**
CHAPTER ONE **1**
1.0 INTRODUCTION **1**
CHAPTER TWO **4**
2.0 LITERATURE REVIEW **4**
2.1 *PERICOPSIS ELATA* 4
2.1.1 Origin and Uses 4
2.2 *STERCULIA RHINOPETALA* (K. SCHUM) 4
2.2.1 Description and uses 4
2.3 *GUAREA CEDRATA* (A. Chev. Pellegr.) 5
2.3.1 Description and uses 5
2.4 CATEGORIES OF SEEDS 5
2.4.1 Orthodox seeds (Desiccation-tolerant) 5
2.4.2 Recalcitrant (Desiccation-sensitive seeds) 6
2.4.3 Intermediate Seeds 6
2.5 SEED DESICCATION 7
2.5.1 Seed desiccation tolerance and its scientific basis 7
2.5.2 Properties of seed desiccants 7
2.5.3 Desiccant drying systems 9
2.6 PACKAGING MATERIALS 10

2.6.1 Properties of seed packaging materials 11
2.7 SEED QUALITY 11
2.7.1 Components of Seed Quality 12
2.7.2 Genetic Purity 12
2.7.3 Physical Purity 12
2.7.4 Germination Capacity and Viability 13
2.7.5 Seed Vigour 13
2.7.5.1 Electrical Conductivity test 14
2.7.5.2 Accelerated Aging test 15
2.7.6 Moisture Content 15
2.7.7 Seed Health 16
2.7.8 Seed Storability 16
2.7.8.1 Moist storage of recalcitrant seeds 17
2.7.9 Effect of Ambient Storage on Seed Quality 18
2.7.10 Storage effects on internal food reserves 18
2.7.10.1 Carbohydrate 19
2.7.10.2 Protein 19
2.7.10.3 Lipids 20
2.7.11 Seed moisture content 21
2.9 SEED LONGEVITY 21
2.9.1 Deterioration-related damages 22
2.10 CLASSES OF SEED DORMANCY 23
2.10.1 Physiological dormancy 24
2.10.2 Morphological dormancy (MD) 24
2.10.3 Morpho-physiological dormancy 24
2.10.4 Physical dormancy 25
2.10.5 Combinational dormancy 25
2.11 DORMANCY-BREAKING ACTIVITIES 25
2.11.1 Hydro-priming 25
2.11.2 Seed scarification 25
2.11.3 Use of plant growth hormones 26

CHAPTER THREE **27**
3.0 MATERIALS AND METHODS **27**
3.1 Location of Seed Collection 27
3.2 Laboratory Experiments 27
3.2.1 Seed Desiccation Experiment 28
3.2.1.1 Seed Desiccants for the experiment 28
3.2.1.2 Zeolite beads 28
3.2.1.3 Charcoal 28
3.2.1.4 Rice 28
3.2.1.6 Parameters taken in this experiment 29
3.2.2 Seed Storage Experiment 29
3.2.3 Moist storage of *G. cedrata* seeds 29
3.2.4 Accelerated Ageing test 29
3.2.5 Dormancy breaking experiment of *S. rhinopetala* seeds 30
3.3.10 Using the seed viability equation to predict longevity of stored seeds 35
Explaining the terms used in the viability equation and viability modelling 35
4.1 Seed initial quality characteristics **37**
4.2 Seed initial proximate composition 38
4.3 Number of days taken for seeds to attain dryness 38
4.4 Effects of desiccants on proximate composition, vigour and 1000 seed weight of the seed 41
species 41
4. 5 Effects of desiccants on germination (viability) of *G. cedrata* after desiccation 43
4.6 Ambient conditions of storage 43
4.7 PROXIMATE, PHYSICAL AND HEALTH COMPOSITION OF *P. ELATA* SEEDS ... 44
4.7.1 Effects of Packaging material and storage periods on carbohydrate content of *P. elata* 44
seeds 44
REFERENCES **86**
APPENDICES **100**

LIST OF TABLES

Table 4.1 Initial seed quality characteristics of *G. cedrata, S. rhinopetala* and *P. elata* after seed collection. 40

Table 4.2: The initial proximate composition of the three species after seed collection. 41

Table 4.3 Number of days taken for seeds to attain dryness 43

Table 4.4 Effects of desiccants on the 1000 seed weight of the three seed species 45

Table 4.5 showing the relative humidity and temperature for the storage experiment 46

Table 4.6 Effects of storage and packaging materials on protein and carbohydrate *of P. elata* ... 48

Table 4.8 Effects of storage periods on the protein content of *P. elata* seeds 50

Table 4.9. Effects of packaging materials and storage periods on the Germination percentage of *P. elata* seeds 51

Table 4.10 Effects of packaging materials and storage periods on the moisture content of *P. elata* seeds 52

Table 4.11 Effects of packaging materials and storage periods on the vigour of *P. elata* seeds .. 54

Table 4.12 Effects of packaging materials and storage periods on the vigour index of *P. elata* seeds 55

4.13 Effects of packaging materials and storage periods on the thousand seed weight of *P. elata* seeds 57

Table 4.14 Effect of packaging material and storage periods frequency of occurrence of *Aspergillus niger* on *P. elata* seeds 58

Table 4.15 Effect of packaging material and storage periods number of seeds infected with *Fusarium moniliforme* 59

Table 4.16 Effect of packaging material and storage periods on percent incidence of *Penicillium*

spp. on *P. elata* seeds 60

Table 4.17 Estimate of seed longevity after 6 months storage and accelerated aging of *P. elata* seeds 61

Table 4.18 Effects of packaging materials and storage periods on carbohydrate content of *S. rhinopetala* seeds 63

Table 4.19 Effects of packaging materials and storage periods on oil content of *S. rhinopetala* seeds 64

Table 4.20 Effects of storage periods on the protein content of *S. rhinopetala* seeds 65

Table 4.21 Effects of packaging materials and storage periods on the germination percentage of *S. rhinopetala* seeds 66

Table 4.22 Effects of packaging materials and storage periods on the Moisture content of *S. rhinopetala* seeds 67

Table 4.23 Effects of packaging materials and storage periods on the vigour of *S. rhinopetala* seeds 68

Table 4.24 Effects of packaging materials and storage periods on the vigour index of *S. rhinopetala* seeds 69

Table 4.25 Effects of packaging materials and storage periods on the 1000 seed weight of *S. rhinopetala seeds* 70

Table 4.26 Effects of dormancy breaking techniques on number of days to emergence germination capacity of *S. rhinopetala* seeds 71

Table 4.27 Effects of packaging materials and storage periods interaction on percent incidence of *Aspergillus flavus* 72

Table 4.28 Effects of packaging materials and storage periods interaction on number of seeds infected with *Botridiplodia theobrome* 73

Table 4.29 Effects of packaging materials and storage periods interaction on number of seed infected with *Aspergillus niger* 74

Table 4.30 Estimate of seed longevity after 6 months storage and accelerated aging of *S. rhinopetala* seeds 75

Table 4.31 Effects of storage periods on moisture content, germination percentage and vigour of *G. cedrata* 76

Table 4.32: Effects of moist storage on the longevity of *G. cedrata* seeds. 77

ABBREVIATIONS AND ACRONYMS

AOAC	Association of Official Analytical Chemists
FAO	Food and Agriculture Organization
IPGRI	International Plant Genetic Resources Institute
ISTA	International Seed Testing Association
ITTO	International Tropical Timber Organization
IUCN	International Union for Conservation of Nature
MSSE	South Eastern *Moist* Semi -*deciduous* zone
NTSC	National Tree Seed Center
SCBD	Secretariat for Convention on Biological Diversity
SID	Seed Information Database
UNEP	United Nations Environmental Programme
WCMC	World Conservation Monitoring Centre

CHAPTER ONE

1.0 INTRODUCTION

Globally, the significance of tropical forests is now well understood by scientists, politicians and people of all races (Wagner and Cobbinah 1993; Goldsmith 1998; Verweij 2002). Tree planting is undoubtedly, known to be an effective measure to protect the climate and mitigate climate change. Trees sequester the greenhouse gas carbon dioxide, counteract soil erosion and desertification (Grainger 1993). Trees act as "carbon sinks" and absorb carbon from the air and change it into plant material. That means that to a large extent, planting trees reduces greenhouse gases in the atmosphere and significantly limit the rate at which the ozone layer is depleted. Goods and services obtained from forests sustain human life. Many forests serve as valuable biodiversity reservoirs. They maintain the fertility and stability of agricultural lands, protecting the natural watershed, serve as homes for countless wildlife, and habitats for some cultures and communities. To be more precise, forests are natural assets of huge importance (SCBD, 2009).

At an alarming rate, however, forests are disappearing nearly one percent a year in sub-Saharan Africa (FAO, 2003), regardless of the many reforestation and conservation activities. Apart from the major widespread environmental problems which include global warming, flooding, food insecurity and loss of biological diversity, there is also widespread concern for the 1.5 to 2 billion people who depend on trees for livestock fodder, fruits, local construction, cooking and heating fuel (Bonner, 1992). The World Conservation Union (IUCN, 2008) UNEP WCMC (2001) estimated that about 2000 tropical tree species in Africa are considered to fall into the categories of being „near threatened" to „critically endangered".

High quality tree seeds are needed to support both reforestation and the *in* and *ex-situ* conservation of forest genetic resources (Schmit, 2000). There is therefore the greatest need to restore degraded areas to improve their productive capacity, environmental functions, and bio- diversity value (Parrotta, 2002). Persistent physical, chemical, and biological factors are the major barriers that impede forest regeneration, these severely degraded areas need human intervention to initiate recovery. Among the many ecological restoration methods, planting of tree seedlings have been identified as one of the effective measures to reforest degraded lands (Lamb *et al*. 2005).

Annually, about one million hectares (2.47 million acres) in the tropics are planted in tree seedlings, but only a small percentage is indigenous (Bonner, 1992). According to Sacande *et al*. (2004) information about the potential of indigenous species and the availability of their seeds and seedlings are insufficient. Access to seeds and seedlings come with seed handling and storage problems, which limit the use of many high value indigenous trees in tree planting and conservation programmes (Sacande *et al*. 2004). Furthermore, tropical forest seeds which show orthodox seed storage behaviour have storage and dormancy related issues which adversely affect their storability and seed quality. Such seeds quickly lose their viability and vigour before the next planting season due to improper drying methods, storage/packaging materials and poor storage conditions. Longer seed storage durations also facilitate seed deterioration and reduce seed longevity. The three selected important economic indigenous species for this study are; *Peripcopsis elata* (Kokrodua) which is an endangered species (IUCN, 2008), *Sterculia rhinopetala* (Wawabima) and *Guarea cedrata* (Kwabohoro) which have been described as vulnerable, according to the IUCN Red List of Threatened Species (IUCN, 2001). These species therefore require urgent conservation attention.

It is an undeniable fact that, the starting material for reforestation is seed. It is also the most useful material for plant conservation purposes. Very high quality tree seeds are required for Ghana to

reach the target of the Forest Plantation Strategy launched in 2013. This Programme was aimed at planting many hectares of forest each year throughout the country up to the year

2050, with vigourous indigenous and exotic tree species (Ghana Forestry Commission, 2013). From the foregoing, if indigenous species are to be used in afforestation and conservation programmes, it is important to gather relevant scientific knowledge on their seed physiology, identify the appropriate responses of their seeds to desiccation, dormancy breaking methods, storage potential, as well as their seed health improvement techniques. The use of beads, silica gel, calcium chloride, charcoal and rice have been widely used on agricultural seeds with quite an appreciable success (Probert, 2003; Rao *et al.*, 2006; Hay *et al.*, 2012) but scanty research information is reported on tree seeds . Storage life of seeds (longevity) has also been improved by using different packaging materials like jute, nylon, paper bags and airtight containers. There is therefore the need to conduct research on the effects of desiccants, packaging materials and storage periods on seed quality and longevity dynamics of these species. The study primarily is aimed at contributing to scientific information on the effects of seed desiccants and storage packaging materials on seed quality and longevity dynamics of three indigenous forest tree species.

The specific objectives of the study were:

1. To determine the effects of seed desiccants on seed physical quality and chemical characteristics
2. To determine the interactive effects of packaging materials and storage periods on seed quality and proximate composition
3. Evaluate the most effective dormancy breaking methods and moist storage on *Sterculia rhinopetala* and *Guarea cedrata* seeds respectively.
4. To predict the longevity of the species using the Improved seed Viability Equation

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 *PERICOPSIS ELATA*

2.1.1 Origin and Uses

Pericopsis elata is from the Fabaceae or Leguminosaea family, known with the names *afrormosia* (DRC, Congo), *assamela* (Cameroon, Côte d‟Ivoire) and *kokrodua* (Ghana). It is of the Guinean-Congolese forest type. Commercially, the tree is known as African teak (with a dark heartwood which is red in colour). It is an endangered species (IUCN, 2001). Special attention should be paid to the study of the tree‟s autecology and relevant information for sustaining and conservation. Uses of wood derived from *P. elata* are boards for coffins, rails, furniture, joinery, decks and handles of tools. The industrial uses include making floors, furniture, cross frames, beautiful veneer and to build ship (Kukachka, 1960).

2.2 *STERCULIA RHINOPETALA* (K. SCHUM)

Sterculia rhinopetala belongs to the Family Sterculiaceae (Malvaceae). The tree can be found in Ghana, Côte d‟Ivoire, Cameroon and other countries. The tree is known with the trade names: Brown sterculia, Red sterculia, Lotofa and Wawabima in Ghanaian Twi language.

2.2.1 Description and uses

This species thrives in the deciduous and forest zone of Ghana (Irvine, 1961) and is typically a light demanding non-pioneer timber species. It has a slow growth rate and has the ability to withstand shade (Louppe, 2008). The wood of *Sterculia rhinopetala* can be used for construction,

making floors, joinery, indoor trim, panelling, stairs, furniture and cross beams, ship and boat making, handles of tools, toys, poles, veneer and plywood.

2.3 *GUAREA CEDRATA* (A. Chev. Pellegr.)

The tree belongs to the family Meliaceae and can be found in Sierra Leone to Uganda, Gabon and DR Congo. The tree is known commonly as light bosse, pink mahogany, African pink cedar, scented Guarea, and Nigerian pear wood (Louppe, 2008).

2.3.1 Description and uses

It is an evergreen, dioecious, big tree up to 45–55 m tall. Early seedling growth is not fast, not up to 30 cm after a year in the nursery (Louppe, 2008). The wood of *Guarea cedrata* is used for house building, flooring, joinery, interior trim, paneling, window frames, doors, ship building, vehicle bodies, furniture, cabinet work, decorative boxes, crates, veneer and plywood (Louppe, 2008). Seeds of the tree are consumed by certain birds and monkeys in Congo (Irvine 1961).

2.4 CATEGORIES OF SEEDS

2.4.1 Orthodox seeds (Desiccation-tolerant)

Desiccation-tolerant tree species are desiccated to low water content without deleterious effect on viability as compared to desiccation-sensitive seeds (McDonald 2004). Orthodox seeds acquire desiccation tolerance at the stage of seed development and retain viability in the dry condition for predictable storage duration (Pammenter and Berjak, 2000). The FAO/IPGRI Genebank Standards (1994) stipulates drying seeds to low water content (3–7% fresh weight basis, which is species-dependent) and keeping them in airtight containers at low temperature, mostly -18 °C or less. To obtain this, however, seeds need to be desiccated to equilibrium moisture content of 10–

15% RH and a temperature of 10°–25°C (Rao *et al.*, 2006). At low temperatures and moisture contents, it is known that many orthodox species are viable for hundreds of years. Such assumptions are supported by reports of ancient seeds maintaining their viability for many decades (Hay, 2003). There are lots of suggestions that have indicated that the term **“** poikilohydric **”** should replace “ orthodox” to describe seeds that can be maintained in equilibrium with ambient relative humidity for long periods (Berjak *et al.*,1990).

2.4.2 Recalcitrant (Desiccation-sensitive seeds)

These are seeds that do not survive drying to any large degree, and are thus not amenable to long term storage. Recalcitrant seeds are metabolically active after being shed **(**Hay, 2003). Several tree species in the tropics and subtropics produce seeds that are desiccation-sensitive (Berjak and Pammenter 2004). In the seed developmental stages recalcitrant seeds do not undergo a period of maturation drying. Viability of fresh recalcitrant seeds significantly reduces when seed moisture is lost and continues to decline at a particular water content termed the “critical moisture content” (King and Roberts, 1980) or “lowest safe moisture content” (Tompsett, 1984). They may also vary with methods of drying (Pritchard, 1991). Internally, a recalcitrant seed is usually a seedling rather than a seed (Thomsen and Stubsgaard 1998**)**. The large size and nature of the seed coat, most recalcitrant seeds dry slowly (Hay, 2003). Berjak *et al.*,(1990) suggested that, the term **“**homoiohydric **”** should be replaced with “ recalcitrant ” to describe those that cannot tolerate desiccation (Berjak *et al.*,1990). Loss of Membrane integrity and nuclear disintegration are the major causes of seed death in desiccation sensitive seeds due to extreme desiccation (Chin 1995).

2.4.3 Intermediate Seeds

Another group of seeds classified based on their storage characteristics are those whose storage behavior is intermediate between the desiccation tolerant and desiccation sensitive groups (Hong

and Ellis., 1992). Intermediate seeds are tolerant of drying than recalcitrant seeds, but the level of tolerance is much limited as compared to orthodox seeds, and they lose germinability rapidly when temperature reduces. Again, the longevity of dry intermediate seeds is kept low with reduction in storage temperature below about 10°C (Hong and Ellis, 1992**)**.

2.5 SEED DESICCATION

The processes of seed dormancy, longevity, and desiccation tolerance take place during seed maturation. These mechanisms appear linked and possibly share common traits. The induction of desiccation is, physiologically, the most visible trait since it is indicated by a reduction in water content to approximately 7% in dry mature seeds. This feature enables the mature seed to be stored in dry conditions and resume metabolic activity when it comes into contact with water (Farnsworth, 2000).

2.5.1 Seed desiccation tolerance and its scientific basis

The capacity of seeds to cope with high moisture loss to values less than 0.1g of water per gram dry weight and when re-hydrated will not have any detrimental effect accumulating (Hoekstra *et al,*. 2001). Such materials usually do not avoid moisture reduction; instead they manage moisture reduction with the aid of certain protective substances and migrate into a metabolically nonactive condition (Alpert, 2005).The ability of seeds to tolerate drying occurs mostly in seeds of some angiosperms. Research works have revealed a number of biological mechanisms that influence desiccation tolerance (Berjak and Pammenter, 1997).

2.5.2 Properties of seed desiccants

Desiccant drying in a closed container is often suggested as a low-technology strategy to minimize the moisture content of seed germplasm (Hay *et al.*, 2012; Probert, 2003). Most of the past research

works with regard to desiccant drying involves the use of desiccants to dry seed (Probert, 2003). The method is currently studied by many researchers since it has a lot of advantages compared to traditional and other artificial seed drying methods. Desiccants such as zeolite seed drying beads® (Hay *et al.*, 2012), molecular sieves (Probert, 2003), lithium chloride and calcium chloride (Probert, 2003), quick lime (Hayma, 2003), silica gel (Hay *et al.*, 2012;

Ondier *et al.*, 2011) and charcoal (Probert, 2003) have been used in drying seeds for planting. The desiccant is used in close contact with or mixed with the wet seeds in an airtight bottle at ambient temperature (Hay *et al.*, 2012; Daniel *et al.*, 2009; Hayma, 2003; Probert, 2003). The amount of water absorbed by the desiccants is based on several factors such as the ratio of desiccant to seeds, temperature, and the affinity of the desiccant for water (Hay *et al.*, 2012; Probert, 2003). Nassari *et al.*, (2014), investigated the extent and speed of drying tomato seeds using desiccant beads under ambient conditions to ultra-low moisture content. Seed drying beads are made up of ceramic substances (usually silicate of aluminum) that mainly absorb and hold water molecules very tightly in their microscopic pores. He added that the rapid drying of seeds will not have any deleterious effect on seed germination. The beads will continue to absorb water until all of their pores are filled, up to 20% of their initial weight. Charcoal is used to significantly minimize seed moisture for long term storage if a ratio of 3:1 charcoal to seed is applied (Probert, 2003). Buady (2002) reported that charcoal is a good desiccant as compared to rice in a research comparing the effect of different desiccants and containers types on the storage of tomato seeds. Nyarko (2006) also confirmed that the effective use of charcoal as a desiccant in drying and storing roselle seeds in different containers.

2.5.3 Desiccant drying systems

The principle of desiccant drying is that the desiccant dries the seeds surface adsorption and capillary condensation until the two materials reach equilibrium (Hay *et al.*, 2012). Surface adsorption occurs because the desiccant removes moisture from its surroundings hence minimizing the relative humidity. The moisture diffuses from the seed through osmotic or vapor pressure gradient. The moisture content at which the desiccant and seeds are the same resulting in no gain or loss of moisture is called equilibrium moisture content (Rao *et al.,* 2006). The use of desiccant in drying consists of a constant rate and falling rate drying times. This is because at the early stage of drying it is mostly capillary forces (Probert, 2003) that drive free moisture to the surface of the seed and make it wet (Srikiatden and Roberts 2007). External factors like air humidity and temperatures are mainly involved in the elimination of moisture and this is termed as constant rate drying period. By keeping air humidity and temperature constant the drying rate remains constant. As the seed loses moisture and approaches the equilibrium moisture content, the internal resistance to moisture transport gets higher than the external resistance. The surface starts to become dry and the wet region moves into the seed. This water movement is due to the active roles of capillary flow and diffusion of water vapor (Probert, 2003). As diffusion increases, the moisture content reduces, the drying rate eventually slows down and this is termed as the falling-rate drying period. All seeds are classified as hydrophilic or hygroscopic substances which looses water in the bound water region or sorption region. When a water potential gradient is established between the surface of the seed and its internal tissues then water in the seed begins to diffuse along the gradient. Evaporation of water from the surface of the seed is mainly due to the water potential difference between the seed and the surrounding air (Probert, 2003).

2.6 PACKAGING MATERIALS

The type of packaging at the time of seed storage becomes extremely relevant on the quality indicators, when the packaging can minimize the rate of seed spoilage, and continue to regulate the initial water content of seeds in storage, preventing the speed at which seeds respire (Tonin and Perez, 2006). The content of oxygen may be reduced by the procedure of packaging into vacuum sealed impermeable plastic bags, or by injection of an oxygen-free gas. Schmidt (2007) mentioned that seeds of tropical trees, stored into low oxygen levels, reduce the rate at which their seeds deteriorate and age. Using a sweet corn experiment, Camargo and Carvalho (2008) confirmed the advantages of employing hermetically sealed material to maintain quality of seed, under ambient environments. Seeds which are supposed to be planted in the subsequent season need to be dried and stored in moisture barrier materials to control loss of viability and vigour (Justice and Bass, 1979). A lot of containers are used for storing seeds but their preference depends on the species or variety of seed and the protection the material can give the seed when stored. It is recommended that seeds should be packaged in smaller units to avoid risk of physical gradients, particularly vapour pressure, which arise in large bulks (Agrawal, 1995). McCormack (2004) also reported that most commonly used storage materials are plastic bottles with screw tops, polyethylene bags, and fibre board drums. Rao *et al.,* (2006) opined that other packaging materials are bottles, aluminum containers, laminated aluminum foils. Adebisi *et al.* (2008) included bottles as one of the best materials to store okra seeds.. These farmers usually use locally produced storage containers such as jute sacks, clay pots, polyethylene bags and nylon sacks which are cheap and readily available. (McCormack, 2004). Olakojo *et al.* (2007) reported that cowpea seed in plastic materials stored better than when kept in tin and earthen containers under the same conditions and contended that nylon sacks should not be used to avoid complete seed spoilage.

2.6.1 Properties of seed packaging materials

Anon (1996) listed some properties of good storage material as: the storage material should be convenient to stack to allow free flow of air during storage, it should be able to prevent spoilage during transit or storage, it should not be too porous to absorb much moisture in the storage place. It should not be dirty, it should be strong to avoid bursting (Anon, 1996). A new method to storage is based on the hermetic principle using material termed as cocoons. This system allows safe storage by avoiding insects and other respiratory organisms in the seed to generate increased CO_2 levels through respiration thereby reducing the O_2. The hermitic storage cocoons were produced for small trader farmers with 10 - 1000 tonnes capacity, small scale storage of small portable containers of 60 kg to 2 tonnes capacity and for quality preservation, insect control and limit condensation transport (Jonfia-Essien *et al*., (2010).

2.7 SEED QUALITY

Quality seed is evaluated using both genetically and physically pure seeds, free of processing damage and physiologically pure (Hilhorst, 2007). The key quality indicators of seed are germination capacity, seed quiescence, vigour, seedlings dry weight, and, and the ability to grow into a healthy seedling (Bewley, 1997; Angelovici *et al*., 2010). The key objective seed testing is to determine the value of seed for planting and the method used must be based on the scientific knowledge of seed and on the accumulated experience of seed analysts. The method must be accurate and reproducible. If the seed lots have high genetic purity, high germination percentage, a minimum of inert, noxious weeds and other crop seeds and are disease-free, it known to have high quality. Several techniques are available for testing seed quality (ISTA, 2007). The methods used in seed testing procedures are: sampling, analytical purity, germination capacity, viability, vigour, seed health, moisture content, weight determination and varietal purity (ISTA, 2007).

2.7.1 Components of Seed Quality

The important aspects of seed quality are genetic and physical purity, germination capacity, seed health, moisture content, vigour, size and uniformity. Other components of quality are seed treatment, packaging and labeling (van-Gastel *et al.,* 1996). Shu (2012) reported that some determinants that define seed quality are: nutritional value (amino acid composition, protein content, micronutrients, vitamins, secondary metabolites), consumer preference (flavour, texture, colour, grain size/shape), pre and post-harvest and industrial/technological traits (fibre traits, sucrose content, storage quality, sprouting, oil content, starches, processing, bread-making) (Shu, 2012).

2.7.2 Genetic Purity

Genetic purity refers to when a variety is true-to-type, and it still possesses the original genetic make-up (van-Gastel *et al.,* 1996). Varietal or cultivar purity guarantees that the genetic make-up (agro-ecological performance) of the variety as defined by the breeding methodology and is present when the seed of improved variety or species gets to growers (van-Gastel *et al.,* 1996). Elias *et al.* (2011) stated that genetic purity is best determined through a field experiment in pre and post control test plots in which the degree of off-types in a seed lot is evaluated. Genetic purity determination include, screening for transgene (GMO) contamination (FAO, 2010). Field inspection and rogueing on the field is one of the means used to insure varietal purity in seed certification standards (FAO, 2010).

2.7.3 Physical Purity

Physical purity test is the basic test undertaken in seed testing, as the others tests are made only on the pure seed constituents. It is done to evaluate the amount of seed purity, seeds of other species, weed seed, damaged seed and inert matter (FAO, 2010). van-Gastel *et al.* (1996) defined physical

or analytical purity as the proportion of pure seed in a particular lot and the percentage composition of the undesirable matter. Eskandari (2012) stated that the physical properties of the seeds are distinguished by: minimum damaged seed, minimal weed seed or inert matter, diseased seed and near uniform seed size. These can be eliminated during processing (Eskandari, 2012).

2.7.4 Germination Capacity and Viability

According to ISTA (2007), germination of a seed in a laboratory test is the emergence and development of the seedling to a stage where its essential structures indicates whether or not it is able to develop further into a normal plant under favourable conditions (ISTA, 2007). Percentage germination is obtained by performing a standard germination test. Failure in germination can lead to total crop loss (van-Gastel *et al.,* 1996). FAO (2010) stated that seeds satisfy their biological function if they are germinable. A seed germination test is probably the common method (ISTA, 2007). Seed viability is influenced by varying conditions. Certain plants do not produce seeds that have active embryos or the seed may have no embryo, a phenomenon often called empty seeds (FAO, 2010). Predators and pathogens can damage or kill the seed while it is still in the fruit or after dispersal. Environmental factors such as flooding or heat can kill the seed before or during germination. The age of the seed affects its health and its viability because the seed has a living embryo and over time, cells die and cannot be replaced. Certain seeds can live for a long time before germination, while others can only live for a short duration after dispersal until they die (FAO, 2010). Seed viability can be tested in many easy ways (ISTA, 2007).

2.7.5 Seed Vigour

Seed vigour is an important parameter of seed quality which influence yield by affecting how young plants seedling establish, specially under bad growing microclimates (Ghassemi-Golezani *et al*., 2010). According to Hampton and Coolbear (1999), vigour was a concept that described

several features, which in turn were associated with various aspects of performance of germinating seed or subsequent seedling. A broad definition was adopted by the ISTA congress in 1977 as: "the sum total of those properties of seed which determine the level of activity and performance of the seed or seedling emergence. Seeds with good performance are termed as high vigour seeds and those which perform poorly are called low vigour seeds" (Hampton and Coolbear, 1999). Milosevic *et al.,* (2010) also defined it as: “the physiological characteristics of seeds that control its capacity to germinate rapidly in the soil and to tolerate various, mostly negative environmental factors”. The lack of vigour means that the named seeds can emerge in optimal conditions (Duda *et al,* 2008). Adverse storage conditions can provoke significant variations in seed viability (Tatić *et al.,* 2008), and that storage duration is negatively correlated with seed vigour (Šimić *et al.,* 2007). Several vigour methods have been developed to predict field performance (van-Gastel *et al.,* 1996). These include physical test (seed volume, weight, size), biochemical test (tetrazolium, conductivity, respiration) and physiological test (standard germination, speed of germination, seedling evaluation, cold test, accelerated aging, controlled deterioration) (Milošević and Malešević 2004).

2.7.5.1 Electrical Conductivity test

Electrical Conductivity test is done on the principle that as seed deterioration progresses, the cell membrane structures become less rigid and more permeable to water, allowing the cell contents to escape into solution with the water and increasing its conductivity reading on an Electrical Conductivity meter. The test gives an accurate estimation of membrane permeability (ISTA, 2007). Seed lots having high electrolyte leakage, that is, having high leachate conductivity, are considered being low in vigour, whilst those with low leakage (low conductivity) are considered as having high vigour (ISTA, 2007). The electrical conductivity test has been used to evaluate the seeds vigor in several species because it is simple to use, cheap, fast, replicable and with easy interpreting

results (Vieira and Krzyzanowski, 1999). It detects the seeds deterioration rate during the storage period (Abreu *et al*., 2011; Panobianco *et al*., 2007).

2.7.5.2 Accelerated Aging test

Accelerated ageing test is one of the most used vigor testing methods because it is simple, and easily of standardized (Tekrony, 1995). The ISTA standardized this method for seed testing, however a uniform accelerated aging procedure has not been developed for testing other crops. Studies of Leeks (2006) relating to seeds revealed a high correlation between germination obtained by using vigour test and field germination (Miloševic *et al*., (2010); Woltz and Tekrony, 2001). Among the vigour evaluation tests, the accelerated aging test has been shown to define seed vigour and therefore, predict their storage potential. In this method the storage potential is known, because it delays the germination process and the growth of the embryo (Maia *et al*., 2007). This test can also be used to evaluate the physiological potential of seeds after certain storage periods (Panobianco *et al*., 2007). Accelerated aging denatures DNA and mRNA causing a biochemical deterioration of the seed internal essential structures and thereby reducing the vigor and seedling establishment immediately after germination. However, the process of accelerated aging is the same as those under ambient environments. The only difference is that the rate of seed spoilage is much higher enhancing the possibility to predict storage potential. In accelerated ageing test the seeds are exposed to stress condition of relative humidity 100% and adverse levels of temperature (40-45ºC) for varying duration followed by germination trials (Chhetri, 2009).

2.7.6 Moisture Content

The primary aim of this test is to determine the moisture content of seeds by methods suitable for predictable use (ISTA, 2007). The moisture content of a sample is the loss in weight of dried seeds in conformity with standard rules. It is normally expressed as a percentage (ISTA, 2007). Quality

seeds need to have an acceptable moisture content to enhance its longevity (van-Gastel *et al.*, 1996). Since moisture content influences seed quality during harvesting, processing and storage, it should be maintained at all stages. High moisture content at harvest damages the seed coat, whereas during storage, it initiates fungal growth, insect activity, heating and germination, which result in rapid seed deterioration. Low seed moisture content can also lead to germination problems such as inducing secondary dormancy (van-Gastel *et al.*, 1996)

2.7.7 Seed Health

The health of seed refers primarily to the presence or absence of disease-causing organisms, such as fungi, bacteria and viruses, and animal pests, including nematodes and insects, but physiological conditions such as trace elements deficiency may be involved (ISTA, 2007). Seedborne pathogens are a serious threat to seedling establishment. General tests such as the blotter test and the agar plate test reveal a wide range of fungal and bacterial pathogens (Mathur and Kongsdal, 2001). Seed health is a component of quality just as viability, vigour and purity (vanGastel *et al.*, 1996). Seed can serve as a transport for the dissemination of plant pathogens, which can lead to disease outbreaks. Seed-transmitted pathogens include fungi, bacteria, nematodes and viruses. They can be transmitted as contaminants with seed, either on the seed surface, or through in the endosperm or embryo. The vast majority of plant diseases are caused by fungal pathogens. Healthy seed is a major requirement for a high-yielding species (van-Gastel *et al.*, 1996). Phyto-sanitary seed quality aspects can be evaluated in laboratories in (FAO, 2010).

2.7.8 Seed Storability

Abreu *et al*., (2011) reported that the factors that affect the quality of seeds in storage are; initial quality; the storage environment (with fluctuations in temperature, moisture, oxygen availability; and the container used for storage) together with features inherent to the kind of seed in study.

The peroxidation of lipids may be the most major cause of deterioration and loss of viability of seeds, since it is a factor that leads to reduction on content of lipids in seeds during the storage procedure. Many times, such factor may be activated by the action of oxygen on a given polyunsaturated fatty acid, which is present in the membranes of seeds. Furthermore, in the process of seed deterioration, the increased on peroxidation of lipids results in damages to the cell membrane, and consequent generation of toxic sub-products (Schwember and Bradford, 2010). Enzymatic changes may seem to be also useful in studies on seed deterioration. Thus, the decrease of antioxidant enzymes is linked to increased peroxidation of lipids as well as to accelerated aging, with a positive correlation between antioxidant capacity of the enzyme and the vigour of seeds (Bailly *et al.*, 2002). The content of oil in seeds may vary according to plant genotype and the storage conditions, especially temperature and relative humidity (Koutroubas *et al.*, 2000) and such variations directly influence degradation of the oil during storage. For

Walters *et al.*, (2010), the chemical degradation of seed components during storage occurs through damages caused by oxidant agents, but the speed of such reactions is defined by properties of the seeds, which in their turn are affected by temperature as well as by moisture.

2.7.8.1 Moist storage of recalcitrant seeds

„Moist storage" involves storage of seeds in media with some moisture-retention ability to prevent dehydration, such as perlite, vermiculite, sawdust, coconut dust, damp charcoal and moist sand. This reduces seed deterioration from dehydration stress, and provides sub-optimal conditions for seed germination. Details of this method are: mixing seeds and the media appropriately; adjusting the medium moisture level so that the stored seeds will not dehydrate rapidly and die, or imbibe lots of water and germinate quickly; allowing necessary ventilation; and possibly maintaining a low temperature but without causing chilling injury. Seeds of *Hopea hainanensis* (Song *et al.*,

1984), *Podocarpus milanjianus* and *Prunus africana* (Schaefer, 1991) have been reported to live longer under these conditions. The two main problems of moist storage are fungal proliferation and early germination of seeds. Anti-fungal sprays and other antimicrobial substances can lower microbial infection (Finch-Savage *et al.*, 2003), but application of natural germination inhibitors like abscisic acid (ABA) has largely failed to prevent germination of recalcitrant seeds in storage, because recalcitrant seeds mostly are not sensitive to ABA. Thus „moist storage" of recalcitrant seeds is only beneficial in the short term.

2.7.9 Effect of Ambient Storage on Seed Quality

FAO (1981) reported that farmers in the developing world still store their produce including seed under the ambient environment. Basu (1995) indicated that serious losses of viability have been reported from areas believed to have suitable climate for the production and storage of seed. In tropical areas, such as Brazil, ambient temperatures of storage are observed above 20 °C, and the decrease in germination was more alarming (Dhingra *et al*., 1998). In general, storage for long or short term is improved under ambient humidity if the seed is well packaged (McCormack, 2004).

2.7.10 Storage effects on internal food reserves

The nutrient reserves of seeds affect germination, growth and seedling survival (Khan and Shankar, 2001), leading to a higher seedling competiveness of heavy seeds (Upadhaya *et al.*, 2007). Therefore, more seed reserves increase the possibility of successful seedling establishment (Khan *et al.*, 2004). The chemical composition of seed with high oil content is correlated to specific processes taking place in the seed during storage (Milosevic and Malesevic, 2004). Such changes during aging are significant in terms of seed quality, characteristics feature that, particularly also implies seed longevity (Milosevic and Malesevic, 2004). The chemical composition of oilseeds leads to specific processes to occur during storage. The seeds containing lipids have low longevity

due to their specific chemical composition. For example, soybean seed storage requires special attention due to its oil content, otherwise certain m mechanisms may occur that lead to the loss of germination ability and seed viability (Balesevic-Tubic *et al*., 2007). Fungal growth can cause changes that are harmful to nutritive content during seed storage. Specifically, nutrients are reduced because of changes in carbohydrates, protein, lipids, and vitamins (Bothast, 1978).

2.7.10.1 Carbohydrate

Conditions that promote fungal activity result in carbohydrate breakdown. Sugars are consumed and decomposed into CO_2 and H_2O. At moisture levels of about 15%, seed looses both starch and sugar and the dry weight reduces (Bothast, 1978). Houghton *et al.*, (2006) reported that carbohydrates are the main food reserve in seeds of most plants. The most common is starch, although hemicelluloses, amyloids, and raffinose oligosaccharides are also relevant. The carbohydrates nourish the embryo plant until it emerges from the soil and can starts producing its own food by means of photosynthesis. In a dicot seed, such as a bean, two cotyledons fill the interior part of the seed. The cotyledons (seed leaves) are essentially part of the embryo. They primarily play the role as food storage and supply the developing embryo with food until its own leaves are prominent.

2.7.10.2 Protein

The total protein percentage of seed as calculated from its nitrogen content is mostly assumed to be constant during storage (Bothast, 1978). All seeds contain one or more groups of proteins that are available in high amounts that function to provide a store of amino acids for use during germination and seedling development. These storage proteins are of utmost importance because they determine not only the total protein content of the seed but also its quality for various purposes (Shewry *et al.,* 1993). Despite huge variation in their detailed structures, all seed storage proteins

have a number of similar properties. First, they are produced at high levels in specific cells and at certain stages of plant growth. Their production is regulated by nutrition, and they act as a sink for excess nitrogen. However, most proteins also contain cysteine and methionine, and specific amount of sulfur is needed for their synthesis. Differences in speed of water absorption observed in different species would be mainly related to seed chemical constituents; higher protein content usually corresponds to a faster water uptake by certain oil seeds (Shewry *et al.,* 1993).

2.7.10.3 Lipids

Because most molds have a high lipolytic activity, fats and oils in seed are readily decomposed into free fatty acids and partial glycerides during the fungal rotting of seeds. These changes are highly accelerated when moisture and temperature are conducive for fungal growth (Bothast, 1978). Lipid oxidation is a spontaneous and inevitable process with direct impact on the market value of the fatty bodies and all the substances formulated from them. The hydro peroxides made from the reaction between oxygen and unsaturated fatty acids are the primary products (Silva *et al*., 1999). Even though these substances do not exhibit taste or odour, they are quickly decomposed at room temperature into aldehydes, ketones, alcohols, hydrocarbons, esters, lactones and furans, resulting in unpleasant taste and odor in oils and fats through peroxide index (Eys *et al*., 2006) with harmful effects on the seed quality. One of the methods used to determine the level of oxidation in fats and oils is the peroxide index. The peroxide index (PI) is the extent of oxidation or rancidity in its initial phase (O‟Brien, 2004). These changes over time may cause seed quality reductions for other oil seed crops in storage. Oil seeds are very sensitive to harsh environmental conditions. It is postulated that their oil content easily oxidizes, leading to deterioration of the seed's health in storage (Kausar *et al*., 2009). Vegetable oils contain relatively high amounts of free fatty acids (FFA) if the grains or seeds present get damaged due to activities in the field or improper storage practices. During storage, lipids are hydrolyzed by the lipases in free fatty acids

(FFAs) and glycerol, mainly in high temperatures and moisture contents (Molteberg *et al.*, 1995). Tweneboah (2000) reported that plants manufacture two kinds of oil: the fixed, „non-volatile" and volatile „essential" oil which serve as food reserves in the plant, and stored in the seed.

2.7.11 Seed moisture content

Safe storage of seeds depends primarily on its moisture percentage, temperature and storage duration. Sastry *et al*., (2007) reported that low moisture content reduces respiration and deterioration and thereby enhances the quality of stored seeds. In storage, seeds are affected by air movement within the seed causing moisture condensation and mould decay (Anon, 1996).

The duration of the storage period is of utmost importance as the maximum moisture levels need to be controlled for safe storage (Anon, 1996).

2.9 SEED LONGEVITY

The germination percentage of long-term stored seeds is due to a combination of growth machinery protection, maintenance, and repair. The longevity of a seed lot is the period of time the seeds remain germinable after they are matured physiologically. For seed storage reasons, longevity is used in the same way as storability. To conserve the initial seed quality, seeds must be well-stored between harvesting period and the planting of a subsequent crop. Sun (1997) reported that the seed glass state is needed for long-term storage. The glass state is strongly related to the accumulation of high temperature oligosaccharides including verbacose, sucrose, raffinose and stachyose (Williams and Leopold, 1995). The amount of oligosaccharides within a seed affects the firmness and amount of the glass state. Therefore, the higher the amount of oligosaccharide contents in the seed, the higher the stability of the glass and the longer the seed can store (Bernal-Lugo and Leopold, 1998). Harrington"s rule of thumb (1972) states that under the optimal levels of moisture

and temperatures for stored seed: each 1% reduction in seed moisture or each 5.6^{o}C reduction in temperature doubles the life span of the seed. These rules will not apply to seeds of moisture contents higher than 14% due to increased respiration and fungal proliferations; and will not apply when moisture content is less than 5% because of the collapse of membrane structure due to the reorientation of hydrophilic compounds in the membranes; and below 0^{o}C, this rule may not hold due to the fact that biochemical reactions related to deterioration are hugely absent. Consequently, seed longevity is a quantitative trait that is characteristic of the species and the storage environment can only help conserve it (Delouche, 1968).

2.9.1 Deterioration-related damages

The result of respiration at all moisture contents is the peroxidation of membrane unsaturated lipids. Lipid peroxidation can be non-enzymatic (autoperoxidation) or enzymatic (by lipooxygenase) and both mechanisms lead to aging in seed (Nagel and Börner, 2010). Autoperoxidation is started by oxygen around unsaturated or polyunsaturated fatty acids such as linoleic and linolenic acids found mainly in seed membranes and storage oils (Copeland and McDonald, 2001). In intact seeds, autoperoxidation normally begins in the mitochondria polar lipids of the embryonic axes (Priestley *et al.*, 1980). This results in the formation of free radicals that are transferred to other seed membranes. Lipid peroxidation occurs in all cells, but in fully imbibed cells, water acts as a buffer between the free radicals generated by autoxidation and the target macromolecules, thereby reducing damage. The choice of the lipid peroxidation reaction is dictated by the moisture content of the seed. Thus, as seed water content is reduced, autoperoxidation prevails and is accelerated by high temperatures and high oxygen amounts (Trawatha *et al.*, 1995). Autoperoxidation is the main cause of seed deterioration at moisture contents below 6%, while above 14% moisture content, lipid peroxidation is stimulated by the

activity of hydrolytic oxidative enzymes such as lipoxygenase that become more active with increasing water content (Krishnan, 2000). According to Nagel and Börner (2010) the chemical composition of the seed affects its sorption properties; the available potential sites for free radical attack and the presence and activity of protective compounds within the seed. The consequence of peroxidation of polyunsaturated fatty acids of the seed membranes is the destabilization of the membranes, which leads to uncontrolled leakage of solutes (Priestley *et al.,* 1980). Other residues of the seed aging process that can result to seed deterioration are the Amodori and Maillard products. The Amodori and Maillard are formed as a result of sugar hydrolysis and lipid peroxidation coupled with non-enzymatic protein modification (Murthy and Sun, 2000). Amadori reactions lead to the chemical modification of proteins by reducing sugars to form fructosyl derivatives, or glycosylated proteins within the seed at storage (Wettlaufer and Leopold, 1991). This process gradually reduces the ability to limit free radical damage and prevents the repair of damage during seed germination (Murthy *et al.*, 2003; Murthy and Sun, 2000). Maillard products are formed through subsequent complex interactions between glycosylated Amadori products to form polymeric brown-coloured products. Maillard products were observed in naturally aged soybean seeds and were associated with the loss of seed viability under long term storage conditions (Sun and Leopold, 1995).

2.10 CLASSES OF SEED DORMANCY

Nikolaeva, (2004), classified dormancy based on both morphological and physiological features of the seed in study. Baskin and Baskin (2004), however have grouped seeds into five types namely: physiological, morphological, morpho-physiological, physical and combinational.

2.10.1 Physiological dormancy

This is the most common type in seeds of gymnosperms and most angiosperm species. It is the most known dormancy form in temperate seed banks and the most common dormancy class „in the field". PD is also the major form of dormancy in most seed model species „in the laboratory", including *A. thaliana*, *Helianthus annuus*, *Lactuca sativa*, *Lycopersicon esculentum*, *Nicotiana* spp., *Avena fatua* , and many cereals. PD can be divided into three levels: deep, intermediate and non-deep (Baskin and Baskin, 2004). For PD deep, embryos removed from these seeds either do not grow or will produce abnormal seedlings; GA treatment does not break their dormancy, and several months of cold stratification are required before germination can take place (Baskin and Baskin, 2004; Baskin *et al*., 2005). For PD non-deep, many seeds exhibit non-deep PD (Baskin and Baskin, 2004). Embryos removed from such seeds form normal young plants; GA treatment can release this dormancy and, depending on species, dormancy can also be controlled by scarification, post-ripening at the time of drying, and water stratification.

2.10.2 Morphological dormancy (MD)

This type is dominant in seeds which have their embryo not matured (based on size), but differentiate into cotyledons and radicle-hypocotyl. Such embryos not quiescent on physiological basis, but simply need time to grow and germinate like in the case of *Apium graveolens* (Jacobsen and Pressman, 1979).

2.10.3 Morpho-physiological dormancy

This type occurs in embryos which are not well developed with a physiological aspect to their dormancy (Baskin and Baskin, 2004). They need a dormancy releasing application, like a mixture of warm and/or cold water which sometimes can be replaced by GA treatment. For example: *Trollius* Ranunculaceae (Hepher and Roberts, 1985)

2.10.4 Physical dormancy

This type occurs as a result of impermeable sections of palisade cells in the seed that regulate water penetration. Mechanical or chemical water treatment can control this type of dormancy.

Examples: *Melilotus* and *Trigonella* (Baskin and Baskin, 1998).

2.10.5 Combinational dormancy

This type of dormancy is where seeds with water-impermeable seed coat together with physiological type (Baskin and Baskin, 2004) especially occurring in *Geranium* and *Trifolium.*

2.11 DORMANCY-BREAKING ACTIVITIES

Hartmann *et al*. (1997), reported that germination of seeds with hard testa may be induced by any means that soften or scarify the seed testa. Several techniques have been researched on using methods like growth hormones, ethrel, hydro-priming, acetone and potassium nitrate which were known to break seed dormancy in certain oil seeds (Maiti *et al*., 2006).

2.11.1 Hydro-priming

Osmo-priming is a method employed primarily to fasten seed emergence as a result of a presowing hydration (McCue *et al*., 2000). KH_2PO_4 and KNO_3 solvents are normally used for seed osmo-conditioning. Mng"omba *et al*. (2007) indicated that when GA3 was applied to *X*. *caffra* seeds in storage, it improved germination significantly. However improved emergence was obtained when seeds were soaked with water.

2.11.2 Seed scarification

Certain seeds have a very hard seed coat that inhibits water absorption like *Pterocarpus angolensis.* The hard testa was raptured using fire to enhance imbibition (Banda *et al*., 2006), though this method may kill the seeds. Ethylene has been known to play a key role in seed germination (

Hilhorst *et al*. 2006). Warm water, H_2SO_4, filing and sand paper have been used to soften the seed testa. However, optimal amounts depend on species (Hilhorst *et al*. 2006).

2.11.3 Use of plant growth hormones

Plant growth regulators hormones are widely used to release dormancy in certain species. Keegan *et al.* (1989) showed that *S. rautanenii* seeds germinated when treated with ethrel. Two major roles of GA in germination are; increasing growth potential of the embryo leading to emergence and also overcoming the mechanical restraint by the materials covering the seed and making weak cells that surround the radicle. (Kucera *et al*., 2005).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Location of Seed Collection

Bobri Forest Reserve was the selected location for the identification of the species which began in August 2015 and seed collection activities also took place in December, 2015. This Forest Reserve is in the south-east sub-type of moist semi-deciduous (MSSE) forest in Ghana, making an area of about 5,445 ha. The forest was demarcated in 1936 and reserved in its unexploited state in 1939 (Alder, 1993). It lies between latitudes 6º39‟ and 6º44‟N and longitudes 1º15‟ and 1º23‟W. The annual rainfall is close to 1,500 mm. The Bobiri forest reserve is home to a wide range of undisturbed tropical forest trees species and serves as a main area for tree seed collection both for tree seed conservation and reforestation programmes by the National Tree Seed Center (NTSC) of Ghana. The three selected trees are among the dominant species in this forest.

3.2 Laboratory Experiments

The laboratory analyses were carried out at the Department of Horticulture, KNUST. These were seed desiccation, seed storage, determination of seed proximate composition (moisture, protein oil, crude fibre and ash), germination test and 1000 seed weight. Department of Crop and Soil Sciences (Seed conductivity test), CSIR-Crops Research Institute (Seed health test) and Forestry Research Institute of Ghana, National Tree seed Center (Accelerated Ageing test)

3.2.1 Seed Desiccation Experiment

A 1:1 seed to desiccant ratio was used. 100g each of the seeds of the three species were weighed using an electronic scale. 100g each of the desiccants were weighed and placed in airtight bottle containers. The desiccants were put in gauze and held above the seeds in the container to prevent the desiccants from having direct contact with the seeds. The treatments were laid in a simple completely randomized design for each of the species and were replicated three times.

3.2.1.1 Seed Desiccants for the experiment

3.2.1.2 Zeolite beads

The beads were obtained from Crop Research Institute, Fumesua, Kumasi after they were heated to remove possible moisture absorption from its ambient environment.

3.2.1.3 Charcoal

This was obtained from Charcoal producers at Ejisu-Juabeng district of Ashanti Region. The wood used for the production of the charcoal was mahogany. The charcoal was well dried by heating thoroughly in the oven for two hours before it was used for the experiment.

3.2.1.4 Rice

Jasmine rice variety obtained from the Department of Horticulture was well toasted in the oven for two hours to remove any seed moisture before it was used for the experiment.

The control treatment was when seeds were allowed to dry under ambient conditions without the use of any desiccant.

3.2.1.5 Biochar

The feedstock which was used in the production of biochar was sawdust. It was collected from the Wood Village at Sokoban in Kumasi, Ashanti Region of Ghana. Sawdust of mahogany was collected

and charred with the Biochar Reactor – a furnace for charring at Chirepatre, in Kumasi (Ashanti Region).

3.2.1.6 Parameters taken in this experiment

Time taken (days) for seeds to be completely dried. Carbohydrate, protein, oil content, germination percentage, moisture content and 1000 seed weight.

3.2.2 Seed Storage Experiment

A 3 X 6 factorial completely randomized design in three replications was used for this experiment. The factors were; three storage periods (no storage, three months and six months storage) six different packaging materials [jute (0.8mm thickness), nylon (0.1mm), paper (0.2 mm), ziplock bag (45 m), airtight bottle and no packaging] were used for each of the species after the seeds were equilibrated. Germination percentage, moisture content, vigour, vigour index, 1000 seed weight and seed proximate compositions (proteins, carbohydrates and oil content) were recorded.

3.2.3 Moist storage of *G. cedrata* seeds

Saw dust, rice husk and river sand were oven-dried at 103 ± 2^{o}C for 17 h and cooled down before use. 50 g dry saw dust was placed into jars and mixed with 90g and 105g distilled water. Then 100 seeds were then put into each jar, shaken up, and stored at 15^{o}C with the top covered loosely. The moisture content of the media was set at 25% for each of the moist media and that of the dried media was kept at 0%. Three jars were sampled for germination evaluation after storage for 1-4 weeks. In order to limit fungal infection and growth, the seeds were cleaned and surface air-dried before storage using sodium hypochlorite.

3.2.4 Accelerated Ageing test

Accelerated Ageing test was carried out at the end of sixth month when stored seeds were subjected to very high temperature and humidity regimes to artificially age the seeds and to predict the vigour

and longevity of the stored seeds. This test was performed for each lot in individual plastic containers with wire mesh suspended inside ("gerbox" with dimensions (11 x 11 x 3 cm), where 100g each of *P. elata* and *S. rhinopetala* seeds were distributed. Within each compartment, 40 mL of distilled water was added, to give 100% relative humidity (Jianhua and McDonald, 1996) and the boxes were closed with the seeds inside and taken to a germination chamber in the laboratory for 96 hours with an aging temperature of 47ºC). After that, the seeds were sterilized for five minutes in 2% sodium hypochlorite solution, they were then rinsed in distilled water prior to the standard germination test (Gordin *et al.,* 2012), recording the germination percentage at seven days after sowing for *P. elata* and 10 days for *S. rhinopetala.*.

3.2.5 Dormancy breaking experiment of *S. rhinopetala* seeds

30 seeds of *S. rhinopetala* were used for each of the dormancy breaking methods and were replicated three times in a simple Completely Randomized Design (Brasil, 2009): 1) control where seeds were not treated; 2) sandpaper scarification of the seed coat; 3) Dipping in both concentrated and dilute H_2SO_4 98% for 1 minute and rinsing in distilled water; 5) gibberellic acid (GA_3 0.5%). Cold water treatment (water not heated for 48 hours in a 20 mL water) and hot water treatment (seeds are put in 100◦C 20 mL hot water for 30 minutes). Germination test was conducted for all these treatments and the days to emergence and the germination percentages were evaluated.

3.3 Data collection

3.3.1 1000 Seed Weight

One thousand seed weight was determined by counting out at random 8 replicates of 100 seeds from the pure seed sample. Each replicate was then weighed with an electronic balance and the weight recorded. The mean weight of the 8 replicates was calculated, and multiplied by 10 (ISTA, 2007).

3.3.2. Measurement of Temperature and Relative Humidity of Storage Room

The ambient storage room temperature and relative humidity readings were taken at specified times of 9:00 am, 12:00 pm and 6.00 pm. Acurite manufactured indoor digital humidity and temperature monitor (00325) was used in taking the readings.

3.3.3 Moisture Content

The low constant temperature oven method (AOAC, 2007) was used to determine the moisture content of the seeds. Empty crucible was thoroughly washed, cleaned and dried for one hour at 130 °C and placed in a desiccator to cool. The crucible and its cover were weighed before and after filling. About 5 g milled each of *P. elata, S. rhinopetala* and *G. cedrata* seed from each sample was weighed and transferred into a previously weighed empty glass crucible and put in an oven with a temperature of 105 °C and dried for 5 h. At the end of the time, the container was covered and removed from the oven and made to cool in a desiccator to room temperature. After cooling, the crucible with its cover and content was reweighed and figures recorded. Loss in weight was calculated as percentage moisture content on wet basis (AOAC, 2007). Calculation of moisture content:

$$\% \text{ Moisture (wet basis)} = \frac{(\text{weight of wet sample} - \text{weight of dry sample})}{\text{Weight of wet sample}} \times 100$$

3.3.4 Crude Fat Content

The sample used for the moisture content determination was transferred into a paper thimble, labeled and placed in a thimble holder for the crude fat determination. 150 mL of petroleum ether was poured into a pre-weighed 500 mL round bottom flask and assembled on a semicontinuous soxhlet extractor and fluxed again for 16 h. The hexane was recovered after removing the paper thimble from the thimble holder and the flask holding the fat heated for 30 min in an oven at 103

°C to get rid of the residual hexane. The flask with the fat was re-weighed after being cooled in a desiccator (AOAC, 2007). The increase in weight was calculated as percentage crude fat as shown below.

Calculation of fat content:

% Fat = (weight of fat) x 100 weight of sample

3.3.5 Protein Content

The protein content was determined using the Kjeldahl method in three steps which were digestion, neutralization and distillation and titration.

3.3.5.1 Digestion method:

About 2 g of the prepared sample was weighed into a digestion flask and mixed with 25 mL of concentrated dilute sulphuric acid, selenium catalyst and some anti-bumping agents. The content of the flask was digested by heating in a fume chamber till the colour of the solution was clear.

3.3.5.2 Neutralization and Distillation process:

After the digestion has ended, the flask was made to cool and transferred into a 100 mL volumetric flask and the volume made up to the 100 mL mark using distilled water. The distillation device was flushed out with distilled water and 10 mL of digested sample transferred into the distillation instrument. The solution was neutralized with 18 mL sodium hydroxide

NaOH and boiled under distillation water in a steam generator. Circulation was done for about 10 min. A conical flask was filled with 25 mL of 2% boric acid and 3 drops of mixed indicator (methylene blue and methylene red) added. The conical flask and its content were put under the condenser in such a way that the tip of the condenser was completely dipped into solution for 10 the min and the end of condenser was washed with distilled water.

3.3.5.3 Titration process:

The nitrogen content was determined by titrating the ammonium borate formed in the conical flask with 0.1M dilute HCl solution. Titre values of the replicate samples were recorded and percentage nitrogen calculated as in the formula below. A blank sample was prepared simultaneously as the sample was being evaluated.

Calculation of crude protein content (AOAC, 2007)

$$\%\text{Nitrogen} = (St - Sb) \times \frac{NA \times 100 \times 0.1 \times 0.014 \times 100}{\text{Sample weight} \times 10}$$

St= Titre of sample

Sb= Titre of blank

NA = Normality of acid

%Protein = % N x F

N= Nitrogen; F= Factor (6.25)

3.3.6 Seed Health

The blotter method was used to determine the presence or absence of seed borne fungi (Mathur and Kongsdal, 2001). A prepared sample of 400 seeds was randomly taken to run this test. The petri dishes were properly washed and cleaned. Each dish was labeled with the necessary seed information for easy identification. Three filter papers were used for each dish. The filter papers were wetted in distilled water and raised till the last drop fell before taking it to the dish. Ten (10) seeds were counted and gently placed on each petri dish. After this all the petri dishes of a sample were put in a tray for incubation. The petri dishes were then incubated for 7 days at 22 ºC under alternating periods of 12 h darkness and 12 h ultraviolet light. Each seed was examined using a stereomicroscope. The seeds were evaluated on the basis of fungal vegetative growth, fruiting bodies, and the characteristic symptoms on the seedlings. Growth nature of the fruiting body was used for the identification of the fungi (Mathur and Kongsdal, 2001). The results were expressed as percentage by number of seeds infected (Mathur and Kongsdal, 2001). Data on the number of

seeds that were infected by fungal species were transformed by using Square root transformation. Results were made on the transformed data.

3.3.7Germination Percentage

Germination test was conducted in fine sand (1 litre of sand: 160mls of water). Plastic trays (30 x 25cm) were used for the test. Germination test was carried out to determine the germination percentage of the three species used for this study. 400 seeds from the pure seed fraction of a purity test were used to conduct the germination test. The seeds were arranged in four replications of 100 each on a counting board and planted in a level layer of moist sand in a perforated container and covered. First count was done on day seven (7). On day ten, each replicate was examined and evaluated separately. Seedlings were counted and grouped into normal, abnormal, freshly ungerminated and dead seeds. The percentage germination was based on the normal germination as specified by the ISTA, (2007).

$$\text{Germination \%} = \frac{\text{Number of germinated seeds X 100}}{\text{Number of total seeds planted}}$$

3.3.8 Seed Vigour

Electrical Conductivity test was used in eveluating the vigour of the seeds. Four replicates of 50 seeds of each sample were drawn at random and tested for electrical conductivity. Seeds were placed in Erlenmeyer flasks containing 75 ml deionized water equilibrated to 25 °C, then maintained at 25 °C for 24 h. After 24 h of soaking, the flasks were shaken for 10-15 sec and seeds were taken out of water with a sterilized forceps (ISTA, 2007). An electrical conductivity meter was inserted into the steep water until a stable reading was obtained and recorded. The average of the two control flasks (sterilized distilled water) served as the control. Conductivity was calculated using the formula below (ISTA, 2007).

Conductivity ($\mu S\ cm^{-1}g^{-1}$) = (Conductivity reading - control reading).

3.3.9 Vigour index determination

Using a metre rule the shoot and root length of 10 germinated seeds after two weeks after germination were measured in three replicates and the average computed. Vigour index was calculated as follows by Abdul-Baki and Alderson (1973)

Vigour Index = (Shoot length + Root length) X Germination Percentage

3.3.10 Using the seed viability equation to predict longevity of stored seeds

Explaining the terms used in the viability equation and viability modelling

$$v = K_i - \frac{p}{10^{K_E - C_W \log m - C_H t - C_Q t^2}}$$

(Ellis and Roberts, 1980)

v = final viability (expressed as %, NEDs or probits) after p days storage.

P = storage time (days) **m** = % moisture content (fresh weight basis) **t** = temperature (°C)

$\mathbf{K_i}$ = initial viability of the seed lot at p = 0 days (seed lot constant)

$\mathbf{C_H}$ and $\mathbf{C_Q}$ = species-specific temperature constants

$\mathbf{K_E}$ and $\mathbf{C_W}$ = species-specific moisture content constants.

C_H and C_Q are species-specific temperature constants. The effect of temperature on seed longevity is similar for all species, at least between −30°C and +90°C, and if individual constants are unknown the "universal" C_H, and C_Q constants of 0.0329 and 0.000478 may be used. Storage temperature after six months storage =28.78°C and 47°C during the aging period.

For both *P. elata* and *S. rhinopetala* the average storage temperature (28.78°C), the initial moisture content, initial germination percentage and the germination percentage after 6 months of storage was used for the longevity calculation. After seeds were aged the aged temperature (47°C) and their corresponding moisture content, germination percentages were used to compute the longevity of the seeds.

3.3.4.11 Data Analysis

Data collected from the laboratory experiments were subjected to analysis of variance using Statistix Student Version 9.0. Tukey's HSD (Honest Significant Difference) was used for mean separation at probability level 0.01 for the laboratory experiments.

CHAPTER FOUR

4.0 RESULTS

4.1 Seed initial quality characteristics

There were significant differences between the treatments for seed moisture content, vigour, vigour index, and thousand seed weight and germination percentage (Table 4.1). *Guarea cedrata* had the significantly highest moisture content (27%) and thousand seed weight (1089.7g). On the other hand, *Pericopsis elata* recorded significantly the highest vigour index (2689.7) but the least moisture content (7.5%) and least thousand seed weight (254.67g). There were also significant differences ($p \leq 0.01$) between the treatments for germination percentage such that *P. elata* recorded significantly the highest germination (96%) percentage yet similar to that of *S. rhinopetala* (95%). There were however no significant differences ($p \leq 0.01$) between the treatments for seed vigour (Table 4.1).

Table 4.1 Initial seed quality characteristics of *G. cedrata*, *S. rhinopetala* and *P. elata* after seed collection.

Species	Moisture Content (%)	Vigour ($\mu S\ cm^{-1}g^{-1}$)	Vigour Index	1000 seed weight (g)	Germination (%)
P. elata	7.5	23.0	2689.7	254.7	96.3
S. rhinopetala	10	22.5	2376.7	779.7	95.4
G. cedrata	27	25.4	2251.7	1089.7	90.7
HSD (0.01)	3.7	4.36	27.96	5.59	3.66

4.2 Seed initial proximate composition

There were significant differences between the *P. elata, S. rhinopetala* and *G. cedrata*. *P. elata* recorded the highest seed oil (31.25%) and protein (37.41%) contents but the least carbohydrate (1.93%) content. The least oil (23%) and protein (9.1%) contents were recorded by *Guarea cedrata*. Conversely, the significantly highest carbohydrate (19.43%) content was recorded by *Guarea cedrata*.

Table 4.2: The initial proximate composition of the three species after seed collection.

Species	Oil %	Protein %	Carbohydrate %
P. elata	31.3	37.4	1.9
S. rhinopetala	23.0	19.2	17.4
G. cedrata	13.5	9.1	19.4
HSD (1%)	10.85	3.23	3.81

4.3 Number of days taken for seeds to attain dryness

There were significant differences between the species for the number of days taken for each of the seed species to dry to a moisture content of 3.5% (Table 4.3). For *P. elata*, it took 2 days for the beads to attain dryness, significantly less in time than the other desiccants. There were however no significant differences in the number of days taken by charcoal and biochar. The longest time for drying was recorded under the control treatment which was not different from the rice

treatment. The rice desiccant treatment took 6.5 times more days than the beads desiccant (Table 4.3).

For *S. rhinopetala* it took 3 days for the beads to attain the moisture content which was significantly less in time than the other desiccant treatments. There were however no significant differences in the number of days used in the drying by charcoal and biochar. The longest time for the attainment of dryness was experienced under no desiccant treatment which was not different from the rice treatment. The rice desiccant treatment took 6.4 times more days than the beads desiccant (Table 4.3).

For *G. cedrata* it took 12.3 days for the beads to effect the drying which was significantly less than the other desiccant treatments. There were however no significant differences in the number of days taken in the drying between charcoal and biochar. The control treatment took the longest time to attain dry seeds but not different from the rice treatment. The rice desiccant treatment took 6.7 times more days than the beads desiccant (Table 4.3).

Table 4.3 Number of days taken for seeds to attain dryness

P. elata

Desiccant	Number of days to drying
Beads	2.0
Charcoal	6.0
Biochar	6.3
Rice	13.0
No desiccant	13.5
HSD (0.01)	**3.55**

S. rhinopetala

Desiccant	Number of days to drying
Beads	3.3
Charcoal	9.8
Biochar	9.8
Rice	21.1
No dessicant	21.9
HSD (0.01)	**3.55**

G. cedrata

Desiccant	Number of days to drying
Beads	12.3
Charcoal	36.8
Biochar	38.6
Rice	79.6
No dessicant	82.7
HSD (0.01)	**3.55**

4.4 Effects of desiccants on proximate composition, vigour and 1000 seed weight of the seed species

For all the three seed species, there were no significant differences between the constituents of the proximate composition. For *P. elata,* carbohydrate content ranged from 1.21% to 1.25%; oil content ranged from 31.52% to 31.58% and protein content ranged from 38.06% to 38.08% . There were also no significant differences between the treatments for seed vigour which ranged from 24.56 μS $cm^{-1}g^{-1}$ to 24.65 μS $cm^{-1}g^{-1}$.

For *S. rhinopetala,* carbohydrate content ranged from 16.53% to 16.95%; oil content ranged from 23.32% to 23.68% and protein content ranged from 20.55% to 20.57%. There were also no significant differences between the treatments for seed vigour which ranged bewteen 26.08 μS $cm^{-1}g^{-1}$ and 26.14 μS $cm^{-1}g^{-1}$.

For *G. cedrata,* carbohydrate content ranged from 18.07% to 18.28%; oil content ranged from 6.52% to 6.58% and protein content ranged from 11.06% to 11.08 %. There were also no significant differences between the treatments for seed vigour such that it ranged from 27.23 μS $cm^{-1}g^{-1}$ to 32.43 μS $cm^{-1}g^{-1}$.

Contrarily for all three seed species, there were significant differences in the 1000 seed weight (Table 4.4). For *P. elata,* the heaviest seeds were attained in the biochar and rice desiccants and theo desiccation control. The lightest seeds were recorded by the bead desiccants but was different from the charcoal desiccant.

For *S. rhinopetala*, the heaviest seeds were recorded by the biochar and rice desiccants as well as the no dessication control. The lightest seeds were attained by the beads desiccant which was not different from the charcoal desiccant.

Table 4.4 Effects of desiccants on the 1000 seed weight of the three species

Species	Desiccants	1000 seed Weight %
	Beads	254.33
	Charcoal	257.00
P. elata	Biochar	258.00
	Rice	258.80
	No desiccant	258.90
	HSD (0.01)	**3.57**
	Beads	781.00
S. rhinopetala	Charcoal	781.43
	Biochar	781.70
	Rice	782.00
	No desiccant	768.10
	HSD (0.01)	**0.74**

	Beads	1099.00
	Charcoal	1098.90
G. cedrata	Biochar	1098.10
	Rice	1097.10
	No desiccant	1094.40
	HSD (0.01)	**3.58**

4. 5 Effects of desiccants on germination (viability) of *G. cedrata* after desiccation.

There were no significant differences among the desiccant treatments for the germination percentage of *G. cedrata* seeds. Germination percentages ranged betwen 8.32% and 12.33%.

4.6 Ambient conditions of storage

Relative humidity ranged from 63.2% to 77.82% whereas temperature was between 27.9ºC and 28.2 ºC. The minimum relative humidity was recorded in January, 2016 and the maximum in June, 2016. The minimum temperature was recorded in January, 2016 and the maximum in March, 2016 (Table 4.35).

Table 4.5 showing the relative humidity and temperature for the storage experiment

Month	**Relative Humidity**	**Temperature**	**Maximum Relative Humidity**	**Minimum Temperature**

January	63.2	28.2	75.25	22.6
February	64.5	29.1	80.72	23.21
March	75.45	29.45	82.91	24.34
April	72.3	29.79	79.25	24.18
May	76.46	28.21	84.92	24.16
June	77.82	27.9	86.23	23.85

4.7 PROXIMATE, PHYSICAL AND HEALTH COMPOSITION OF *P. ELATA* SEEDS

4.7.1 Effects of Packaging material and storage periods on carbohydrate content of *P. elata* seeds

Significant packaging material x storage period interactions were observed in the percent carbohydrate of *P. elata* seeds (Table 4.6). Seeds in airtight bottle under ambient conditions produced significantly the highest percent carbohydrate, but was not different from seeds stored in airtight bottles and ziplock for three and six month of storage periods. The jute packaged seeds stored for six months contained the least carbohydrate content yet but was not different from those stored in nylon bags, paper bags and unpackged. Among the storage periods, seeds which were not stored produced significantly the highest carbohydrate percentage, 2 times greater than the least obtained from seeds stored for six months (Table 4.6). Among the packaging materials, seeds stored in airtight bottles recorded significantly the highest carbohydrate content which were similar to those stored in ziplock bags. The least was recorded in the control.

Table 4.6 Effects of storage and packaging materials on protein and carbohydrate *of P. elata*

Packaging Materials	Carbohydrate % Storage Periods			
	0 Month	**3 months**	**6 Months**	**Mean**
Jute	1.21	0.70	0.3	0.74
Nylon	1.22	0.80	0.4	0.81
Paper	1.23	0.75	0.45	0.81
Ziplock	1.23	1.20	1.00	1.17
Airtight bottle	1.25	1.24	1.10	1.20
No Packaging	1.20	0.60	0.35	0.72
Means	1.22	0.90	0.60	
Tukeys HSD (0.01):	Pack. Material= 0.3 Storage Periods=0.2 Pack Mat. x Storage periods=0.7			

4.7.2 Effects of packaging materials and storage periods on the oil content of *P. elata* seeds

Significant packaging material x storage period interactions were observed in the percent oil of *P. elata* seeds (Table 4.7). Seeds in any of the packaging materials but not stored contained significantly the highest percent oil content. The unpackaged seeds stored for six months had the least oil content. Among the storage periods, seeds not stored produced significantly the highest oil percentage (31.13%), 1.2 times greater than that obtained from seeds stored for six months (26.14%) (Table 4.7). Among the packaging materials, seeds stored in airtight bottles recorded significantly the highest oil content but was not different from values obtained from the ziplock bags. The least oil content was recorded by the unpackaged seeds which were also similar to those in jute, nylon and paper bags.

Table 4.7 Effects of storage periods and packaging materials on oil content of *P. elata*

	Oil %			
Storage periods **Packaging materials**	**0 Month**	**3 months**	**6 Months**	**Mean**
Jute	31.08	26.83	24.83	27.58
Nylon	31.23	27.83	25.33	28.13
Paper	31.33	28.33	25.88	28.50
Ziplock	30.83	29.83	28.33	29.66
Airtight bottle	31.27	30.63	28.83	30.24
No Packaging	31.03	25.83	23.67	26.84
Means	31.13	28.22	26.14	

HSD (1%): Pack. Material= 1.2 Storage Periods=0.7 Pack Mat. x Storage periods=2.6

4.7.3 Effects of storage periods on the protein content of *P. elata* seeds

There were significant differences between the storage periods for the protein content of *P. elata* seeds (Table 4.8). Significantly highest protein was produced by seeds which were not stored (37.81%), while the least was produced by seeds stored for 6 months (36.36%).

Table 4.8 Effects of storage periods on the protein content of *P. elata* seeds

Storage Periods	protein%
No storage	37.81
3 Months storage	37.32
6 months storage	36.36
HSD (1%)	0.8

4.7.4 Effects of packaging materials and storage periods on the germination percentage of

***P. elata* seeds**

There were significant packaging materials x storage period interactions for the germination percentage of *P. elata* seeds (Table 4.9). Seeds in airtight bottle without storage recorded the highest germination (96.33%) although not significantly different from seeds stored in airtight bottle (93.33%) and ziplock bags (92.40%) for six months. The least germination was recorded by the conrtol stored for six months (69.67%) which was similar to seeds stored in jute bag (71.33%), nylon (72.33%) and paper bags (73.30%) for the same period. Among the packaging materials, seeds stored in airtight bottles produced significantly the highest germination percentage (92.40%) which was similar to seeds stored in ziplock bottles (91.33%), but were 1.1 times greater than the least germination recorded by the control (80.44%). Among the storage periods, seeds with no storage recorded a significantly highest germination percentage (94.18%),
1.3 times more than the least germination percentage produced by seeds stored for 6 months (73.89%).

Table 4.9. Effects of packaging materials and storage periods on the Germination percentage of *P. elata* seeds

	Germination %			
	Storage periods			
Packaging materials	**0 Month**	**3 months**	**6 Months**	**Mean**
Jute	93.31	81.33	71.33	82.54
Nylon	93.33	83.32	72.32	82.67
Paper	94.32	84.33	73.30	84.23
Ziplock	95.10	93.40	92.40	91.33
Airtight bottle	96.33	95.43	93.33	92.40

No Packaging	92.33	80.31	69.67	80.44
Means	94.18	83.67	73.89	

HSD (1%): Pack. Material=2.5 Storage Periods= 1.5 Pack Mat. x Storage periods= 5.3

4.7.5 Effects of packaging materials and storage periods on moisture content of *P. elata* seeds

There were significant packaging materials x storage period interactions on the moisture content of *P. elata* seeds (Table 4.10). Seeds in airtight bottle without storage had significantly the lowest moisture content (3.33%), yet similar to all the other treatment combinations except seeds in jute package stored for six months and unpackaged seeds stored for three and six months. Unpackaged seeds stored for six months had the highest moisture content which was similar to the seeds in jute package stored for six months. Among the packaging materials, unpackaged seeds had significantly the highest moisture (5.78%), similar to moisture content from seeds stored in jute (4.94%), nylon (4.84%) and paper bags (4.77%). Among the storage periods, seeds stored for six months produced the highest moisture (5.03%), which was 1.4 times more than the least moisture produced by seeds without storage (3.52%)

Table 4.10 Effects of packaging materials and storage periods on the moisture content of *P. elata* seeds

	Moisture content %			
Packaging materials	**Storage periods**			**0 month 3 months 6 Months Mean**
Jute	3.53	5.03	6.03	4.94
Nylon	3.63	4.63	5.33	4.84
Paper	3.73	3.83	5.8	4.77
Ziplock	3.38	3.34	3.45	3.35

Airtight bottle	3.33	3.33	3.38	3.32
No Packaging	3.83	6.33	7.17	5.78
Means	3.52	4.42	5.03	

HSD (0.01): Pack. Material=1.2 Storage Periods= 0.7 Pack Mat. x Storage periods= 2.6

4.7.6 Effects of packaging materials and storage periods on the vigour of *P. elata* seeds

There were significant packaging materials x storage period interactions on the vigour of *P. elata* seeds (Table 4.11). Unpackaged seeds stored for six months recorded significantly the highest vigour (34.90μScm^{-1g-1}), which was not different from seeds packaged in jute bags (34.80 μScm^{1g-1}) , nylon (34.85 μScm^{-1g-1}) and paper bags (34.10 μScm^{-1g-1}). The least vigour was produced by seeds in airtight bottles without storage (22.85 μScm-1g-1) similar to seeds packaged in airtight bottle (23.85 μScm-1g-1) and ziplock bags (23.85 μScm-1g-1) and stored for six months.

Among the packaging materials, unpackaged seeds produced the highest vigour (28.78 μScm-1g1), which was 1.1 times more than the least vigourous recorded by seeds in airtight bottles (24.72 μScm-1g-1). Across the storage periods, seeds stored for 3 months had significantly the highest vigour (30.10 μScm-1g-1), which was 1.2 times greater than the vigour produced by seeds which were not stored (24.17 μScm-1g-1).

Table 4.11 Effects of packaging materials and storage periods on the vigour of *P. elata* seeds

	Vigour ($\mu S\ cm^{-1}g^{-1}$)			
Storage periods			**Packaging materials**	**0**
Month 3 months 6 Months Mean				
Jute	24.85	28.35	34.80	28.69
Nylon	24.45	27.85	34.85	27.72
Paper	24.65	26.81	34.10	27.12
Ziplock	22.90	22.94	23. 85	25.18
Airtight bottle	22.85	22.95	23. 85	24.72
No Packaging	24.84	29.83	34.90	28.78
Means	24.17	27.367	30.10	

HSD (1%): Pack. Material=0.5 Storage Periods= 0.3 Pack Mat. x Storage periods= 1.1

4.7.7 Effects of packaging materials and storage periods on the vigour index of *P. elata*

seeds

There were significant packaging materials x storage periods interactions ($p \leq 0.01$) on the vigour index of *P. elata*, (Table 4.12). Seeds in airtight bottle with no storage recorded significantly the highest vigour index (2668.30) which was similar to seeds stored for six months in airtight bottle (2665.34) and ziplock bags (2664.35). The least vigour index was recorded by seeds unpackaged and stored for six months which similar those stored in jute, nylon and paper bags and stored for the same period. Across the packaging materials seeds packaged in airtight bottles recorded significantly the highest vigour index (2181.73), which was 1.1 times more than seeds not packaged. Among the storage periods, seeds which were not stored recorded significantly the highest vigour index (2627.70), which was 1.8 times more than the least recorded by seeds stored for six months (1473.62).

Table 4.12 Effects of packaging materials and storage periods on the vigour index of *P. elata* seeds

Vigour Index

	Storage periods			
Packaging materials	**0 months**	**3 months**	**6 Months**	**Mean**
Jute	2604.41	2106.20	1422.32	2042.72
Nylon	2576.20	2158.10	1421.43	2041.30
Paper	2632.14	2184.30	1420.53	2041.31
Ziplock	2665.30	2662.32	2664.35	2180.70
Airtight bottle	2668.35	2663.43	2665.34	2181.73
No Packaging	2604.40	2080.55	1419.72	2042.85
Means	2627.72	2067.12	1473.62	

HSD (1%): Pack. Material=2.5 Storage Periods= 1.5 Pack Mat. x Storage periods=5.3

4.7.8 Effects of packaging materials and storage periods on 1000 seed weight of *P. elata* seeds

There were significant packaging materials x storage periods interactions for the thousand seed weight of *P. elata* (Table 4.13). Seeds in airtight bottles without storage recorded the highest thousand seed weight which was not different from seeds stored in airtight bottles and ziplock bags for six months and 3 months. The unpackaged seeds stored for six months and three months recorded significantly lowest thousand seed weight which were similar to seeds stored for six months and 3 months in jute bags, nylon bags and paper bags. Across the packaging materials, seeds packaged in airtight bottle and ziplock bags recorded significantly highest thousand seed weight (248.62 g) and (248.34g) respectively. Seeds not packaged recorded the least thousand seed weight yet similar to those stored in jute, nylon and paper. Among the storage periods, seeds which were not stored recorded significantly the highest thousand seed weight (259.33 g).

The least was produced by seeds stored for six months (247.61 g).

4.13 Effects of packaging materials and storage periods on the thousand seed weight of *P. elata* seeds

1000 Seed Weight (g)

Storage periods 8	**Packaging materials**	**0 months**	**3 months**	**6 Months**	**Mean**
Jute	249.33	232.33	235.31	238.67	
Nylon	248.31	233.33	237.33	237.67	
Paper	249.33	237.33	239.32	238.22	
Ziplock	250.30	248.33	247.33	248.34	
Airtight bottle	251.33	249.33	243.13	248.62	
No Packaging	247.32	235.33	237.10	239.22	
Means	259.33	254.33	247.61		

HSD (1%): Pack. Material=5.7 Storage Periods= 4.1 Pack Mat. x Storage periods=6.1

4.7.9 Effect of packaging material and storage periods on the number of seeds infected with *Aspergillus niger* on *P. elata* seeds

There were significant packaging material x storage periods interactions on the number of seeds infected with *Aspergillus niger* (Table 4.14). The highest number of seeds infected with *A. niger*

was found in unpackaged seeds stored for six months. The least number of infected seeds were obtained from seeds in airtight bottles and ziplock and without storage. Among the packaging materials, unpackaged seeds recorded the highest number of seeds infected with *A. niger* whereas the least was recorded by seeds in airtight bottle and ziplock material. Among the storage periods, seeds stored for three and six months recorded the highest number of seeds infected with *A. niger* while the least was recorded by seeds without storage.

Table 4.14 Effect of packaging material and storage periods on number of seeds infected with *Aspergillus niger*

	Number of seeds infected with *Aspergillus niger*			
Storage periods **Packaging materials**	**0 months**	**3 months**	**6 Months**	**Mean**
Jute	10.00	14.00	20.00	14.67
Nylon	11.00	14.00	16.33	13.78
Paper	12.00	15.00	15.00	14.00
Ziplock	9.00	11.00	12.00	11.68
Airtight bottle	9.00	12.00	12.01	11.33
No Packaging	11.00	17.00	21.00	16.33
Means	10.34	14.18	16.22	

HSD (0.01): Pack. Material=1.6 Storage Periods= 0.9 Pack Mat. x Storage periods=3.3

Table 4.7.10 Effect of packaging material and storage periods on number of seeds infected with *Fusarium moniliforme* on *P. elata* seeds

There were significant packaging material x storage periods interactions on the number of seeds infected with *F. moniliforme* (Table 4.15). The highest number of seeds infected with *F. moniliforme* was found in unpackaged seeds stored for six months. The least number of infected seeds were obtained from seeds in airtight bottles and ziplock and without storage. Among the packaging materials, unpackaged seeds recorded the highest number of seeds infected with *F. moniliforme*. whereas the least was recorded by seeds in airtight bottle and ziplock material. Among the storage periods, seeds stored for three and six months recorded the highest number of seeds infected with *F. moniliforme* while the least was recorded by seeds which were not stored.

Table 4.15 Effect of packaging material and storage periods number of seeds infected with *Fusarium moniliforme*

	Number of seeds infected with *Fusarium moniliforme*			
Storage periods				
Packaging materials	**0 months**	**3 months**	**6 Months**	**Mean**
Jute	2.54	3.23	3.53	3.10
Nylon	2.73	3.23	3.38	3.12
Paper	2.91	3.38	3.23	3.18
Ziplock	2.33	2.37	3.38	2.37
Airtight bottle	2.33	2.35	2.35	2.36
No Packaging	2.73	3.53	3.80	3.35
Means	2.60	3.23	3.32	

HSD (0.01): Pack. Material=0.04 Storage Periods= 0.02 Pack Mat. x Storage periods=0.08

Table 4.7.11 Effect of packaging material and storage periods on number of seeds infected with *Penicillium* spp. on *P. elata* seeds

There were significant packaging material x storage periods interactions on the number of seeds infected with *Penicillium* spp. (Table 4.16). The highest number of seeds infected with *Penicillium* spp was found in unpackaged seeds stored for six months. The least number of infected seeds were obtained from seeds in airtight bottle and ziplock and without storage.

Among the packaging materials, unpackaged seeds recorded the highest number of seeds infected with *Penicillium* spp. whereas the least was recorded by seeds in airtight bottle and ziplock material. Among the storage periods, seeds stored for three and six months recorded the highest number of seeds infected with *Penicillium* spp while the least was recorded by seeds which were not stored.

Table 4.16 Effect of packaging material and storage periods on number of seeds infected with *Penicillium* spp. on *P. elata* seeds

	Percent *Penicillium spp* (%)			
Storage periods **Packaging materials**	**0 months**	**3 months**	**6 Months**	**Mean**
Jute	6.00	10.00	16.00	10.67
Nylon	7.00	10.00	12.33	9.78
Paper	8.00	11.00	11.00	10.00
Ziplock	5.00	9.00	9.00	7.67
Airtight bottle	5.00	8.00	8.00	7.00

No Packaging	7.00	13.00	17.00	12.33
Means	6.33	10.167	12.22	

HSD (0.01): Pack. Material=2.03 Storage Periods= 1.22 Pack Mat. x Storage periods=4.22

4.7.12 Estimate of seed longevity after 6 months storage and accelerated aging of *P. elata* seeds

P. elata seeds stored in airtight bottle after six months had a predicted longevity of 243 years and those not packaged had 5 years. After the six months when the seeds were aged, the longevity greatly dropped to 164 days for the airtight bottle and 20 days for the no packaging (Table 4.17).

Table 4.17 Estimate of seed longevity after 6 months storage and accelerated aging of *P. elata* seeds

Packaging	After 6 months of Storage Longevity (years)	After Accelerated Aging materials Longevity (Days)
Jute	11	31
Nylon	21	49
Paper	38	64

Ziplock	200	155
Airthight bottle	243	164
No packaging	5	20

4.8 PROXIMATE, PHYSICAL AND HEALTH COMPOSITION OF *S. RHINOPETALA* SEEDS

4.8.1 Effects of packaging materials and storage periods on carbohydrate content of *S. rhinopetala* seeds

Significant packaging material x storage period interactions were observed in the percent carbohydrate of *S. rhinopetala* seeds (Table 4.18). Seeds in airtight bottle but not stored produced significantly the highest percent carbohydrate yet not different from seeds stored in airtight bottles and ziplock for six month. The unpackaged seeds stored for six months produced the least carbohydrate which were also not different from those stored in jute bags, nylon bags and paper bags. Among the packaging materials, seeds stored in airtight bottles recorded significantly the highest carbohydrate content (16.87%) although similar to those stored in ziplock bags (16.15%). The least was recorded by seeds stored without any packaging material (13.30%) which were also similar to those in jute (13.30%), nylon (14.13%) and paper bags (14.80%). Among the storage periods, seeds not stored produced significantly the highest

carbohydrate content (16.39%), 1.3 times greater than the least obtained from seeds stored for six months (12.73%).

Table 4.18 Effects of packaging materials and storage periods on carbohydrate content of *S. rhinopetala* seeds

	Carbohydrate %			
Packaging Materials	**Storage Periods**			
	0 Month	3 months	6 Months	Mean
Jute	16.50	13.00	11.50	13.30
Nylon	16.40	14.00	12.00	14.13
Paper	16.00	15.40	13.30	14.80
Ziplock	16.60	16.10	14.90	16.15
Airtight bottle	16.80	16.60	15.60	16.87
No Packaging	16.20	12.90	10.50	13.30
Means	16.39	14.70	12.73	

Tukey HSD (0.01): Pack. Material= 1.7 Storage Periods= 1.0 Pack Mat. x Storage periods= 3.5

4.8.2 Effects of packaging materials and storage periods on oil content of *S. rhinopetala*

seeds

Significant packaging material x storage period interactions were observed in the percent oil content of *S. rhinopetala* seeds (Table 4.19). Seeds in any of the packaging materials but not stored contained significantly the highest percent oil content. The unpackaged seeds stored for six months had the least oil content. Among the packaging materials, seeds stored in airtight bottles recorded significantly the highest oil content (22.03%) although similar to those stored in ziplock bags (21.83 %). The least was recorded by seeds stored in the unpackaged material (18.90%) which was also similar to those in jute (19.53%), nylon (19.10%) and paper bags (20.30%). Among the storage periods, seeds which were not stored produced significantly the highest oil content (23.11%), 1.3 times greater than the least obtained from seeds stored for six months (18.17%) (Table 4.18).

Table 4.19 Effects of packaging materials and storage periods on oil content of *S. rhinopetala* seeds

Oil content (%)	**Packaging Materials**			
Storage Periods				
	0 Month	**3 months**	**6 Months**	**Means**
Jute	23.08	18.83	16.83	19.53
Nylon	23.23	19.83	17.32	19.10
Paper	23.33	20.33	17.82	20.30
Ziplock	23.13	22.63	21.73	21.83
Airtight bottle	22.83	21.70	21.83	22.03
No Packaging	23.03	17.83	15.83	18.90
Means	23.11	20.22	18.17	

Tukey HSD (1%): Pack. Material= 1.3 Storage Periods= 0.7 Pack Mat. x Storage periods= 2.7

4.8.3 Effects of storage periods on the protein content of *S. rhinopetala* seeds

There were significant differences between the storage periods for the protein content of *S. rhinopetala* seeds (Table 4.20). The significantly highest protein was produced by seeds which were not stored (19.91%), whiles the least was produced by seeds stored for 6 months (17.94%).

Table 4.20 Effects of storage periods on the protein content of *S. rhinopetala* seeds

Storage Periods	Protein %
No storage	19.91
3 Months storage	17.99
6 months storage	17.94
HSD (1%)	**0.79**

4.8.4 Effects of packaging materials and storage periods on the germination percentage of *S. rhinopetala* seeds

There were significant packaging materials x storage period interactions on the germination percentage of *S. rhinopetala* (Table 4.21). Seeds in airtight bottles without storage recorded the highest germination (95.33%) which was not significantly different from seeds stored in airtight bottle (92.33%) and ziplock bags (91.40%) for six months. The least was recorded by unpackaged

seeds stored for six months (69.32%) which was similar to seeds stored in jute bag (70.10%), nylon (71.30%) and paper bags (72.20%). Among the packaging materials, seeds stored in airtight bottle produced significantly the highest germination percentage (92.60%) which was similar to seeds stored in ziplock bottles (91.60%). The least germination was recorded by unpackaged seeds (78.20%). Among the storage periods, seeds without storage recorded the significantly highest germination percentage (91.12%), which was 1.2 times more than the least germination percentage produced by seeds stored for 4 months (73.40%).

Table 4.21 Effects of packaging materials and storage periods on the germination percentage of *S. rhinopetala* seeds

	Germination %			
Storage periods			**Packaging materials**	**0**
Month	**3 months**	**6 Months**	**Mean**	
Jute	88.33	77.32	70.10	78.67
Nylon	89.31	78.32	71.30	79.80
Paper	90.33	79.00	72.20	80.70
Ziplock	93.30	92.20	91.40	92.60
Airtight bottle	95.33	93.32	92.33	91.60
No Packaging	90.33	75.40	69.32	78.20
Means	91.12	80.50	73.40	

HSD (0.01): Pack. Material=2.62 Storage Periods= 1.58 Pack Mat. x Storage periods= 5.45

4.8.5 Effects of packaging materials and storage periods on the moisture content of *S.*

***rhinopetala* seeds**

There were significant packaging materials x storage period interactions on the moisture content of *S. rhinopetala* seeds (Table 4.22). Seeds in airtight bottle without storage recorded significantly lowest moisture, which was similar to seeds stored in airtight bottle and ziplock bags for six months. However, seeds which were not packaged but stored for six months recorded the highest moisture content although similar to those stored in jute, nylon and paper bags for the same period. Among the packaging materials, unpackaged seeds had significantly the highest moisture (6.35%), while the least moisture percentages were produced by seeds packaged in airtight bottles (3.57%) and ziplock bags (3.62%). Among the storage periods seeds stored for six months produced the highest moisture (5.73%), which was 1.5 times more than the least moisture produced by seeds without storage (3.71%).

Table 4.22 Effects of packaging materials and storage periods on the Moisture content of *S. rhinopetala* seeds

	Moisture content %			
Storage periods			**Packaging materials**	**0 Month**
	3 months	6 Months	Mean	
Jute	3.83	6.10	7.00	5.61
Nylon	3.80	6.51	6.10	5.47
Paper	3.70	5.80	5.80	5.51
Ziplock	3.51	3.62	3.65	3.62
Airtight bottle	3.50	3.60	3.60	3.57
No Packaging	3.90	7.00	8.20	6.37
Means	3.71	5.30	5.73	

HSD (0.01): Pack. Material=0.8 Storage Periods= 0.5 Pack Mat. x Storage periods= 1.7

4.8.6 Effects of packaging materials and storage periods on the vigour of *S. rhinopetala* seeds

There were significant packaging materials x storage period interactions for the vigour of *S. rhinopetala* seeds (Table 4.23). Unpackaged seeds stored for six months recorded significantly the highest solutes leakage (32.01μScm^{-1g-1}), which was not different from seeds packaged in jute bags (31.20 μScm^{-g-1}), nylon (31.15 μScm^{-1g-1}) and paper bags (31.08 μScm^{-1g-1}). The least solutes leakage was produced by seeds which were not stored (22 μScm^{-1g-1}) yet similar to seeds packaged in airtight bottle (24.02 μScm^{-1g-1}) and ziplock bags (24.50 μScm^{-1g-1}) stored for six months. Among the packaging materials, unpackaged seeds produced the highest solutes leakage (27.67 $\mu Scm^{-1}g^{-1}$), which was 1.2 times more than the least solutes leakage recorded by seeds in airtight bottles (23.01$\mu Scm^{-1}g^{-1}$). Across the storage periods, seeds stored for 6 months had significantly highest solutes leakage (28.42$\mu Scm^{-1}g^{-1}$), whereas the least solutes leakage was produced by seeds which were not stored (23.02 $\mu Scm^{-1}g^{-1}$).

Table 4.23 Effects of packaging materials and storage periods on the vigour of *S. rhinopetala* seeds

	Vigour ($\mu S\ cm^{-1}g^{-1}$)			
Storage periods	**Packaging materials**			**0**
Month 3 months 6 Months Mean				
Jute	23.20	27.90	31.20	27.38
Nylon	23.60	27.00	31.15	26.87
Paper	23.80	25.00	31.08	25.93
Ziplock	22.50	23.20	24.50	23.40

Airtight bottle	22.00	23.00	24.02	23.01
No Packaging	23.00	28.00	32.01	27.67
Means	23.02	25.68	28.42	

HSD (0.01): Pack. Material=1.7 Storage Periods= 1.0 Pack Mat. x Storage periods= 3.5

4.8.7 Effects of packaging materials and storage periods on the vigour index of *S. rhinopetala* seeds

There were significant packaging materials x storage periods interactions on the vigour index of *S. rhinopetala,* (Table 4.24). Seeds with no storage recorded significantly high vigour index (2229.30) similar to seeds stored for six months in airtight bottle (2321.32) and ziplock bags (2321.30). Seeds unpackaged and stored for six months recorded the least vigour index. Across the packaging materials seeds packaged in airtight bottles recorded significantly highest vigour index (2324.85), which was 1.4 times more than seeds not packaged. Among the storage periods, seeds which were not stored recorded significantly highest vigour index (2271.20), which were 1.7 times more than the least produced by seeds stored for six months (1320.35).

Table 4.24 Effects of packaging materials and storage periods on the vigour index of *S. rhinopetala* seeds

	Vigour Index			
Storage periods				
Packaging materials	**0 months**	**3 months**	**6 Months**	**Mean**
Jute	2200.31	1232.30	1229.34	1718.31

Nylon	2225.01	1230.30	1225.30	1740.04
Paper	2250.30	1238.30	1248.32	1761.72
Ziplock	2325.34	2319.30	2321.30	2324.70
Airtight	2329.30	2326.30	2321.32	2324.85
No Packaging	2221.32	1234.30	1224.32	1708.30
Means	2271.20	1764.00	1320.35	

HSD (1%): Pack. Material=2.52 Storage Periods= 1.58 Pack Mat. x Storage periods=5.45

4.8.8 Effects of packaging materials and storage periods on the 1000 seed weight of *S. rhinopetala* seeds

There were significant packaging material x storage periods interactions on the 1000 seed weight of *S. rhinopetala* (Table 4.25). Seeds in airtight bottle but not stored recorded the highest 1000 seed weight which was not different from seeds stored in airtight bottles and ziplock bags for six months and 3 months, respectively. The unpackaged seeds stored for 6 months and 3 months recorded significantly the lowest 1000 seed weight. Across the packaging materials, seeds packaged in airtight bottle and ziplock bags recorded significantly the highest 1000 seed weight (766.33g) and (765.33g) respectively. Among the storage periods, seeds which were not stored recorded significantly the highest 1000 seed weight (771.11) whiles the least was produced by seeds stored for six months.

Table 4.25 Effects of packaging materials and storage periods on the 1000 seed weight of *S. rhinopetala seeds*

1000 Seed Weight (g)

Storage periods	Packaging materials	0 months	3 months	6 Months	Mean
Jute		763.05	769.01	745.23	747.67
Nylon		744.01	771.50	748.11	748.67
Paper		775.43	772.10	759.00	760.67
Ziplock		775.20	769.20	764.12	765.33
Airtight bottle		776.61	768.61	766.00	766.33
No Packaging		765.21	765.24	750.41	742.67
Means		771.11	738.50	722.67	

HSD (1%): Pack. Material=5.5 Storage Periods= 5.1 Pack Mat. x Storage periods=10.8

4.8.9 Effects of dormancy breaking techniques on number of days to emergence and germination capacity of *S. rhinopetala* seeds

There were significant differences between the treatments for the number of days taken for seeds to emerge and the germination percentage of *S. rhinopetala* (Table 4.26). GA_3 recorded a significantly least number of days to emergence (7.30 days) with a corresponding highest germination percentage (96.33%). These were not different from seeds treated with cold water for 24 hours (7.40 days to emergence and germination percentage of 95.30%), dilute H_2SO_4 (8.8 days and 92% germination) and sand paper scarification (12.7 days and 85.% germination). The highest number of days to emergence (17.33 days) and the corresponding germination percentage (10.40%) were recorded by the seeds which were not treated (control).

Table 4.26 Effects of dormancy breaking techniques on number of days to emergence germination capacity of *S. rhinopetala* seeds

Dormancy-breaking Method	Days to Emergence	Germination%
GA_3	7.30	96.33
Cold water 48 hours	7.40	95.30
Dilute H_2SO4	8.83	92.00
Sand paper	12.7	85.30
Hot water treatment	13.33	70.33
Conc. H_2SO4	15.20	30.40
No treatment	17.33	10.40
HSD (0.01)	5.38	11.20

4.8.10 Effects of packaging materials and storage periods interaction on number of seeds infected with *Aspergillus flavus* on *S. rhinopetala*

There were significant packaging material x storage periods interactions on the number of seeds infected with *Aspergillus flavus* (Table 4.27). The highest number of seeds infected with *A. flavus* was found in unpackaged seeds stored for six months. The least number of infected seeds were obtained from seeds in airtight bottles and ziplock bags and without storage. Among the packaging materials, unpackaged seeds recorded the highest number of seeds infected with *A. flavus* whereas the least was recorded by seeds in airtight bottles and ziplock bags material. Among the storage periods, seeds stored for three and six months recorded the highest number of seeds infected with *A. flavus* while the least was recorded by seeds without storage.

Table 4.27 Effects of packaging materials and storage periods interaction on percent incidence of *Aspergillus flavus*

	Number of seeds infected with *Aspergillus flavus*			
Storage periods				
Packaging materials	**0 months**	**3 months**	**6 Months**	**Mean**
Jute	15.00	19.00	21.33	18.44
Nylon	16.00	19.00	20.00	18.33
Paper	17.00	19.00	20.00	18.67
Ziplock	14.00	15.00	15.50	15.44
Airtight bottle	13.00	14.90	15.30	15.33
No Packaging	16.00	21.00	23.00	20.11

Means	15.22	18.89	19.56

HSD (0.01): Pack. Material=0.8 Storage Periods= 0.5 Pack Mat. x Storage periods=1.8

4.8.11 Effects of packaging materials and storage periods interaction on number of seeds infected with *Botridiplodia theobrome*

There were significant packaging materials x storage periods interactions on the number of seeds infected with *Botridiplodia theobrome* (Table 4.28). The highest number of seeds infected with *B. theobrome* was found in unpackaged seeds stored for six months. The least number of infected seeds were obtained from seeds in airtight bottle and ziplock and without storage. The unpackaged seeds recorded the highest number of seeds infected with *B. theobrome* whereas the least was recorded by seeds in airtight bottles and ziplock material. Seeds stored for three and six months recorded the highest number of seeds infected with *B. theobrome* while the least was recorded by seeds without storage.

Table 4.28 Effects of packaging materials and storage periods interaction on number of seeds infected with *Botridiplodia theobrome*

Packaging materials	**Storage periods**			
	0 months	**3 months**	**6 Months**	**Mean**
Jute	9.00	13.00	15.00	12.33
Nylon	10.00	13.00	15.00	12.33

Paper	11.00	13.00	14.00	14.67
Ziplock	8.00	9.53	9.20	10.67
Airtight bottle	8.00	9.40	9.31	9.87
No Packaging	10.00	15.00	17.00	16.00
Means	9.33	13.00	13.67	

HSD (0.01): Pack. Material=1.7 Storage Periods= 1.0 Pack Mat. x Storage periods=3.5

4.8.12 Effects of packaging materials and storage periods interaction on number of seeds infected with *Aspergillus niger*

There were significant packaging materials x storage periods interactions on the number of seeds infected with *A. niger* (Table 4.29). The highest number of seeds infected with *A. niger* was found in unpackaged seeds stored for six months. The least number of infected seeds were obtained from seeds in airtight bottles and ziplock bags and without storage. Unpackaged seeds recorded the highest number of seeds infected with *A. niger* whereas the least was recorded by seeds in airtight bottles and ziplock material. Seeds stored for three and six months recorded the highest number of seeds infected with *A. niger* while the least was recorded by seeds without storage.

Table 4.29 Effects of packaging materials and storage periods interaction on number of seed infected with *Aspergillus niger*

Number of seeds infected with *Aspergillus niger*

Storage periods				Packaging materials	0 months	3 months	6 Months	Mean
Jute	10.00	14.00	16.00	13.33
Nylon	11.00	14.00	15.00	13.33
Paper	12.00	14.00	15.00	13.67
Ziplock	9.00	10.40	11.10	11.30
Airtight bottle	9.00	10.00	11.00	11.20
No Packaging	11.00	16.00	18.00	15.00
Means	10.44	14.00	14.67	

HSD (0.01): Pack. Material=1.7 Storage Periods= 1.0 Pack Mat. x Storage periods=3.5

4.8.13 Estimate of seed longevity after 6 months storage and accelerated aging of *S. rhinopetala* seeds

Seeds of *S. rhinopetala* stored in airtight bottles after six months had a predicted longevity of 177 years and those not packaged had 99 years. After seeds were aged after the six months period the longevity greatly dropped to 79 days for the airtight bottle and 64 days for the no packaging (Table 4.30)

Table 4.30 Estimate of seed longevity after 6 months storage and accelerated aging of *S. rhinopetala* seeds

Packaging	After 6 months of Storage Longevity (years)	After Accelerated Aging materials Longevity (Days)
Jute	88	65
Nylon	94	65
Paper	114	66
Ziplock	144	76
Airtight bottle	177	79
No packaging	99	64

4.9 PHYSICAL AND HEALTH COMPOSITION OF *G. CEDRATA* SEEDS

4.9.1 Effects of storage periods on moisture content, germination percentage and vigour of *G. cedrata*

There were significant differences in storage periods for moisture content, germination and vigour of *G. cedrata* seeds (Table 4.31). Seeds stored for six months had the highest moisture content (6.72%) and the least was recorded by the no storage treatment (3.5%). No storage registered the highest germination (3.58%) and the least germination (1.21%) was recorded after six months of storage. Seed electrical conductivity was highest after six and three months of storage (45.33 $\mu Scm^{-1}g^{-1}$) while no storage recorded the least solutes leakage (38.67 $\mu Scm^{-1}g^{-1}$).

Table 4.31 Effects of storage periods on moisture content, germination percentage and vigour of *G. cedrata*

Storage Periods	Moisture Content %	Germination %	Vigour $\mu Scm^{-1}g^{-1}$
No storage	3.5	3.58	38.67
3 Months	6.33	2.12	45.33
6 months	6.72	1.21	45.33
HSD (0.01)	0.79	0.46	2.61

4.9.2 Effects of moist storage on the longevity of *G. cedrata* ceeds.

There were significant differences between moist storage methods for the number of days taken for seeds to deteriorate and their percent viability (Table 4.32). Seeds stored in moist sawdust significantly took the highest number of days to deteriorate (24.67%) with the highest viability (96%) and seeds stored in dry topsoil took the least number days to deteriorate (3.96 days) with the lowest viability (7.67%)

Table 4.32: Effects of moist storage on the longevity of *G. cedrata* seeds.

Media for Moist storage	Days taken for seeds to deteriorate	Viability (%)
Moist sawdust	24.67	96.00
Moist rice	12.67	79.67
Moist topsoil	12.00	49.00
Dry saw dust	6.00	11.00
Dry rice husk	5.50	8.00
Dry topsoil	3.96	7.67
HSD (0.01)	6.40	54.18

4.10 Relationship between seed damage by *Menechamus* spp and seed viability

There was a significant negative relationship between seed damage by *Menechamus* spp and viability (Eqn 1). Seed damage by *Menechamus* spp explained 93% of the variation in seed

viability.

$Y_{viability}$= 82.0208-0.76083$X_{(damaged\ seeds)}$; P = 0.000; R^2= 0.94; n = 18…….Eqn 1.

CHAPTER FIVE

5.0 DISCUSSON

5.1 Initial seed quality and proximate composition of the three species

The differences observed in the initial seed quality could be attributed to the high genetic variations that existed between the species. Seeds of *P. elata* and *S. rhinopetala* were shed with relatively lower moisture contents of 7.5% and 10%, respectively, which is characteristic of orthodox seeds. According to Berjak and Pammenter (2004), viability of orthodox seeds can be maintained even when the moisture content is reduced and can also be dried further to enhance their longevity. The results of the present study showed that *P. elata* and *S. rhinopetala* seeds could remain viable for a long period of time when moisture was reduced. *G. cedrata* seeds, however were shed at very high moisture content (27%) and the seeds were metabolically active and also recorded high germination which is characteristic of recalcitrant seeds. Hay (2003) reported that recalcitrant seeds are metabolically active and would have high germination capacity when planted immediately after seed collection. The results of the present study clearly confirm that *G. cedrata* had an initial high seed moisture and a high initial germination probably showing recalcitrant seed storage behaviour. The initial vigour index was highest (2689.7) whilst the initial vigour (in terms of solute leakage) were low and within the recommended leakage levels as reported by Milosevic, (2010) that seeds with leakage below 25 μS $cm^{-1}g^{-1}$ were of high vigour whilst those with vigour more than 35 μS $cm^{-1}g^{-1}$ were of low vigour.

5.2 Effects of desiccants on seed quality and proximate composition of the three species.

The beads were significantly able to dry the seeds at a faster rate as compared to charcoal, biochar, rice and the control. This could be attributed to the presence of aluminum silicates that fill the micropores which have high affinity to hold water in these micro molecular pores for a longer duration. The results of the current study confirms the findings of Nassari *et al.* (2014) who investigated the drying ability of beads on the quality of tomato seeds and reported that the beads were significantly effective to reduce/absorb seed moisture at the fastest rate. Hay *et al.* (2012), also reported on the advantages of using the beads as a desiccant including their greater affinity for water, especially at low humidity; more rapid drying; and no hysteresis effect, which lowered the amount of water that could be adsorbed after regeneration. Buady (2002), reported that charcoal was a good drying agent and was found to keep stored seeds viable quite better as compared to dried rice used as a desiccant. Moreover, Nyarko (2006), indicated that rice was a poor desiccant as compared to charcoal just as was found in the present study. Additionally, for *P. elata* and *S. rhinopetala*, the desiccants did not have any deleterious effect on the vigour (solutes leakage), vigour index, germination percentage, seed protein, oil content and carbohydrate. This could be due to the fact that the two species are orthodox seeds and that desiccation to a lower moisture content rather improved viability thereby confirming Harrington's principle that for every 1% reduction in seed moisture there was a doubling of the viability of the seed (Harrington, 1972). McDonald (2004) also reported that desiccationsensitive seeds cannot be dried to lower moisture content without deleterious effect on viability as compared to desiccation-insensitive seeds.

The deleterious effects of desiccation on *G. cedrata* seeds which was evident in the significantly reduced germination percentage, confirmed their high sensitivity to drying. According to Pritchard

(1991), seeds that are desiccation-sensitive lose their viability considerably after dehydration. Hoekstra *et al*. (2001) also indicated that desiccation resulted in reduced cellular volumes and caused the compaction of cytoplasmic components. This compaction increased molecular interactions leading to protein denaturation and membrane fusion. Furthermore, Chin (1995) opined that death of recalcitrant seeds was due to a reduction in moisture and was basically due to the loss of membrane integrity and nuclear disintegration. The results of the present study for *G. cedrata* confirm these findings.

5.3 Effects of packaging materials and storage periods on the seed quality of the three species.

Seeds stored in airtight and ziplock bags for three and six months periods had the highest seed quality but those which were not packaged but stored for 3 months and six months had reduced seed quality. The other porous materials like jute, nylon and paper also lost their seed quality considerably in storage. Again an increase in storage periods under relatively high temperature and humidity also caused a reduction in vigour and viability of the stored *P. elata* and *S. rhinopetala*. These observations could be due to the rapid exchange of gases between the seeds and their ambient environment under high relative humidity for the storage experiment, resulting in reabsorption of moisture by the seeds due to their hygroscopic nature and thereby enhancing metabolic activities and oxidation processes. These metabolic activities and oxidation processes eventually depleted the essential food reserves in the seed leading to the gradual loss of vigour and viability. According to Tonin and Perez (2006), the type of packaging at the time of seed storage becomes extremely relevant on the quality indicators, when the packaging can minimize the rate of seed spoilage, and continue to regulate the initial water content of seeds in storage, preventing the speed at which seeds respire. Furthermore, in the process of seed deterioration, the increase on peroxidation of lipids results in damages to the cell membrane, and consequent generation of toxic

by-products (Schwember and Bradford, 2010). There was therefore the need to use moisture proof containers like the airtight bottle and the ziplock material which served as barrier against gaseous exchange and maintained the quality of the seeds within the period of storage. The results of the current study also support the findings of Schmidt (2007) that seeds of tropical trees, stored into a low oxygen levels, reduce the rate at which their seeds deteriorated and aged. Sastry *et al*. (2007) reported that seeds with low moisture content reduced respiration and deterioration and thereby enhanced the quality of the stored seeds. In storage, seeds were affected by air movement within the seed causing moisture condensation and mould decay (Anon, 1996). The duration of the storage period was of utmost importance as the maximum moisture levels needed to be controlled for safe storage (Anon, 1996).

5.4 Effects of packaging materials and storage periods on chemical properties of the three species.

Seeds stored in airtight bottles and ziplock bags for three months and six months periods had the highest carbohydrate, oil and protein contents but those which were not packaged but stored for 3 months and six months had reduced amount of these nutrients. The longer the storage periods, the higher the loss of carbohydrate, protein and oil. Such observations could be attributed to the hydrolysis of available carbohydrates into sugars, peroxidation of seed oil and possible denaturation of storage proteins as storage duration increased. According to Bemal-Lugo and Leopold (1992), carbohydrate hydrolysis into sucrose reduced vigour and germinability of stored seeds over time. Houghton (2006) reported that carbohydrates were the main food reserve in seeds of most plants and the most common was starch, although hemicelluloses, amyloids, and raffinose oligosaccharides were also relevant. Lipid peroxidation could also be non-enzymatic (autoperoxidation) or enzymatic (lipooxygenase) and both mechanisms led to aging in seed (Nagel

and Börner 2010). Murthy and Sun (2000) also indicated that lipid peroxidation could give rise to secondary products that could denature proteins and DNA in a non-enzymatic manner by means of the Maillard and Amadori reactions. Furthermore, Mutters (2003) also reported that chemical constituents greatly change in storage especially within 3-4 months at very high temperatures. The results of this study agree with previous findings that the consequence of peroxidation of polyunsaturated fatty acids of seed membranes was the destabilization of the membranes, which led to uncontrolled leakage of solutes (Priestley *et al.,* 1980). Seed vigour significantly decreased after increasing the storage periods which eventually caused seed deterioration (Delouche and Baskin, 1973).

5.5 Effects of packaging materials and storage periods on seed health characteristics of the species.

Seeds stored in airtight bottles and ziplock bags for three and six months periods had the least number of seeds infected with pathogens but those which were not packaged but stored for 3 months and six months had the highest number of seeds infected. The longer the storage periods, the higher the number of seeds infected. Agarwal and Sinclair (1997) considered many of these fungi as "storage fungi" that could be involved in the deterioration of seeds during storage. However, Agarwal (1995) indicated that seed-borne microflora living in association with seed did not cause disease automatically. According to Agrios (2005), for a disease to occur, the three major components of disease cycle (host, pathogen and environment) effectively had to interact. When any of the three components was held at zero, there could be no disease. As one component varied, it affected the level of disease severity within a particular host (Agrios, 2005).

Though these pathogens were identified in *P. elata* and *S. rhinopetala*, the number of seeds infected did not exceed economic injury level to cause disease conditions to reduce in seed quality

particularly before three months storage. However, after six months of storage, these pathogenic fungi species significantly contributed to reducing the quality and longevity of the seed.

5.6 Effects of moist storage methods on *G. cedrata* seeds

The moist storage of *G. cedrata* seeds using moist saw dust showed significant improvement in longevity of the seeds. This is probably due to the reduced dehydration offered by the moisture in the moist saw dust. This created a microclimate for the seed by providing adequate ventilation to delay deterioration caused by temperature build up and lipid peroxidation, reducing microbial build up, and maintaining optimum temperature. According to Wen (2008), moist storage reduces deterioration caused by dehydration stress thereby providing sub-optimal conditions for seed germination. He added that the period of seed longevity in moist storage would only be appropriate for short term storage or would be used for seedling storage as suggested by Krishnapilly (2000).

5.7 Relationship on damaged seeds and viability of *G. cedrata* seeds

Menchamus spp weevil is known to have a life cycle of about 4 weeks feeding on the seeds of *G. cedrata* and it causes serious damage within a short period of time. The results of the present study showed that the severity of damage could be due to the nature of its mouthpart called snout (rostrum) used for boring holes and laying eggs in seeds eventually causing complete destruction of the seeds as reported by (Wagner *et al*. 1991).

5.8 Effects of dormancy breaking techniques on *S. rhinopetala* seeds and its relationship with seed longevity.

Seeds of *S. rhinopetala* exhibit hard seed coat dormancy probably due to its thickened cuticular seed testa consisting of palisade cells which prevent water permeability into the seed and reducing levels of water imbibition. Baskin (2003), reported that physical dormancy occurs as a result of

water impermeable layers of suberin-impregnated palisade cells in the seed coat that regulate water movement and that mechanical or chemical water treatment can break such dormancy.

Results from the current study showed that dormancy was broken when the seed coats were hydrolyzed to enhance easy movement of water into the most essential parts of the seed for germination to take place. Hartmann *et al*. (1997), reported that germination of seeds with hard testa may be induced by any means that soften or scarify the seed testa. Maiti *et al*. 2006 revealed that several techniques have been researched on using methods like growth hormones, ethrel, hydro-priming, acetone and potassium nitrate which were known to break seed dormancy in certain oil seeds. GA3 is a growth hormone which releases dormancy in seeds and improve germination. Mng‟omba *et al*. (2007) indicated that when GA3 was applied to *X. caffra* seeds in storage, it improved germination significantly. Moreover, two major roles of GA in germination are; increasing growth potential of the embryo leading to emergence and also overcoming the mechanical restraint by the materials covering the seed and making weak cells that surround the radicle. (Kucera *et al*., 2005).

Sand paper scarification could also reduce the thickened cuticular nature of the seed coat allowing water permeability and thereby inducing germination as shown by the results of the present study. Previous studies had established the relationship of seed dormancy to seed longevity and germination (Nguyen, 2012). Tran and Cavanagh (1984) indicated that seed dormancy is biologically beneficial for long term seed survival and can be important for wild plants. This suggests the possible improvement in the longevity of stored *S. rhinopetala* seeds.

5.9 Predicting seed longevity after 6 months storage and accelerated aging

P. elata and *S. rhinopetala* can be stored for longer periods in airtight containers and that the accelerated aging conditions fastened the deterioration of the seeds, hence reducing their viability drastically. This suggests that under harsh conditions when seeds are aged, seed moisture increases under high temperature and relative humidity which promotes protein degradation, sugar hydrolysis, rise in metabolic and oxidation reactions. Murthy *et al.* (2002) reported that temperature and moisture content are the most important factors affecting the rate of seed deterioration and used to determine the relationship between advances in seed ageing and several possible primary biochemical deterioration processes, including lipid peroxidation and sugar hydrolysis.

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 CONCLUSIONS

Results obtained from this study hav shown that among the four desiccants used in drying *P. elata, S. rhinopetala* and *G. cedrata,* beads had the fastest drying time without any deleterious effect on the physical and chemical properties of the seeds. *G. cedrata* seeds lost viability considerably after desiccation and therefore could not be stored. To improve their longevity however, moist storage with moist saw dust extended the life span of the seeds. Airtight bottles and ziplock bags used for the storage of *P. elata* and *S. rhinopetala* seeds improved seed viability with less fungal damage to seeds stored for three and six months as compared to jute, paper, nylon and no packaging materials. Airtight bottles and ziplock bags which are non-porous materials maintained seed moisture, vigour, vigour index, protein, carbohydrate and oil contents. Seed deterioration was minimized in these packaging materials as compared to the others (jute, nylon paper and the control) but temperature and relative humidity were high and fluctuated under ambient storage conditions which ultimately contributed to reducing seed quality and longevity. After the sixth month storage, longevity (years) of *P. elata* was 243 years when stored in airtight bottles and that of *S. rhinopetala* stored in airtight bottles was 177 days. When seeds were aged artificially, their longevity hugely reduced to 164 days and 79 days for *P. elata* and *S. rhinopetala* respectively when stored in airtight bottles. Seeds were infected with saprophytic fungi which did not cause disease infestation but resulted in the gradual loss of seed viability and longevity. *S. rhinopetala* showed hard seed coat dormancy and therefore dormancy was broken with GA_3, dilute H_2SO_4, cold water treatment for 48 hours and sand paper scarification.

6.2 RECOMMENDATIONS

- Further research should be conducted on the molecular characterization of seed storage proteins and their effect on quality of the two orthodox species.

- Future experiments on *G. cedrata* should focus on cryopreservation for short-term storage in gene banks
- Varying desiccant:seed ratio should be studied on *P. elata and S. rhinopetala*
- Further research should be conducted on seed treatment methods to manage *Menechamus spp.* on *G. cedrata* seeds after collection.
- The Improved seed viability equations should be used to predict the longevity of other important endangered indigenous forest tree species.

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APPENDICES

Appendix 1. Analysis of Variance Table for Carbohydrate

Source	DF	SS	MS	F	P
Species	2	552.127	276.064	254.83	0.0000
Error	6	6.500	1.083		
Total	8	558.627			

Grand Mean 12.953 CV 8.04

Appendix 2. Analysis of Variance Table for Oil

Source	DF	SS	MS	F	P
Species	2	471.561	235.780	26.86	0.0010
Error	6	52.667	8.778		
Total	8	524.227			

Grand Mean 22.594 CV 13.11

Appendix 3. Analysis of Variance Table for Germination

Source	DF	SS	MS	F	P
Species	2	62.0000	31.0000	31.00	0.0007
Error	6	6.0000	1.0000		
Total	8	68.0000			

Grand Mean 93.667 CV 1.07

Appendix 4. Analysis of Variance Table for Moisture content

Source	DF	SS	MS	F	P
Species	2	675.500	337.750	337.75	0.0000
Error	6	6.000	1.000		
Total	8	681.500			

Grand Mean 14.833 CV 6.74

Appendix 5. Analysis of Variance Table for Protein

Source	DF	SS	MS	F	P
Species	2	1235.09	617.543	793.98	0.0000
Error	6	4.67	0.778		
Total	8	1239.75			

Grand Mean 21.896 CV 4.03

Appendix 6. Analysis of Variance Table for 1000 seed weight

Source	DF	SS	MS	F	P
Species	2	1068950	534475	229061	0.0000
Error	6	14	2		
Total	8	1068964			

Grand Mean 708.00 CV 0.22

Appendix 7: Analysis of Variance Table for Vgour Index

Source	DF	SS	MS	F	P
Species	2	305438	152719	2618.04	0.0000
Error	6	350	58		
Total	8	305788			

Grand Mean 2439.3 CV 0.31

Appendix 8: Analysis of Variance Table for Vigour

Source	DF	SS	MS	F	P
Species	2	13.3021	6.65104	5.71	0.0512
Error	5	5.8267	1.16533		
Total	7	19.1287			

Grand Mean 23.788 CV 4.54

Appendix 9: Analysis of Variance for the Desiccant Experiment

Analysis of Variance Table for Carbohydrate

Source	DF	SS	MS	F	P
Desicants	4	0.15	0.039	0.00	1.0000
Species	2	1877.61	938.807	26.57	0.0000
Desicants*Species	8	0.38	0.048	0.00	1.0000
Error	30	1059.99	35.333		
Total	44	2938.14			

Grand Mean 10.284 CV 57.80

Appendix 10: Analysis of Variance Table for Days to complete desiccation

Source	DF	SS	MS	F	P
Desicants	4	7480.0	1870.00	1870.00	0.0000
Species	2	15567.6	7783.82	7783.82	0.0000
Desicants*Species	8	4610.1	576.27	576.27	0.0000
Error	30	30.0	1.00		

Total 44 27687.8

Grand Mean 23.854 CV 4.19

Appendix 11: Analysis of Variance Table for oil

Source	DF	SS	MS	F	P
Desicants	4	0.02412	6.030E-03	0.00	1.0000
Species	2	4872.40	2436.20	384.66	0.0000
Desicants*Species	8	6.740E-29	8.425E-30	0.00	1.0000
Error	30	190.000	6.33333		
Total	44	5062.42			

Grand Mean 20.489 CV 12.28

Appendix 12: Analysis of Variance Table for Protein

Source	DF	SS	MS	F	P
Desicants	4	3.600	9.000E-04	0.00	1.0000
Species	2	5628.30	2814.15	1206.06	0.0000
Desicants*Species	8	1.324	1.655	0.00	1.0000
Error	30	70.0000	2.33333		
Total	44	5698.30			

Grand Mean 23.237 CV 6.57

Appendix 13: Analysis of Variance Table for Vigour

Source	DF	SS	MS	F	P
Desicants	4	18.561	4.6403	5.95	0.0012
Species	2	187.664	93.8322	120.37	0.0000
Desicants*Species	8	38.194	4.7742	6.12	0.0001
Error	30	23.387	0.7796		
Total	44	267.806			

Grand Mean 26.738 CV 3.30

Appendix 14: Analysis of Variance Table for 1000 seed weight

Source	DF	SS	MS	F	P
Desicants	4	70	18	25.42	0.0000
Species	2	5401816	2700908	3899289	0.0000
Desicants*Species	8	17	2	3.02	0.0129
Error	30	21	1		
Total	44	5401924			

Grand Mean 712.19 CV 0.12

ANALYSIS OF VARIANCE FOR THE STORAGE EXPERIMENT OF *P. ELATA*

Appendix 15: Analysis of Variance Table for Carbohydrate

Source	DF	SS	MS	F	P
PM	5	2.14008	0.42802	10.70	0.0000
SP	2	3.50023	1.75012	43.75	0.0000
PM*SP	10	1.00657	0.10066	2.52	0.0207
Error	36	1.44000	0.04000		
Total	53	8.08688			

Grand Mean 0.9061 CV 22.07

Appendix 16: Analysis of Variance Table for oil

Source	DF	SS	MS	F	P
PM	5	73.077	14.615	25.97	0.0000
SP	2	226.348	113.174	201.10	0.0000
PM*SP	10	37.962	3.796	6.75	0.0000
Error	36	20.260	0.563		
Total	53	357.646			

Grand Mean 28.495 CV 2.63

Appendix 17: Analysis of Variance Table for Germination percentage

Source	DF	SS	MS	F	P
PM	5	358.31	71.66	32.25	0.0000
SP	2	3702.26	1851.13	833.01	0.0000
PM*SP	10	101.96	10.20	4.59	0.0003

Error 36 80.00 2.22
Total 53 4242.54

Grand Mean 83.907 CV 1.78

Appendix 18: Analysis of Variance Table for Moisture content

Source	DF	SS	MS	F	P
PM	5	37.8582	7.5716	13.30	0.0000
SP	2	20.7203	10.3602	18.20	0.0000
PM*SP	10	16.0398	1.6040	2.82	0.0109
Error	36	20.4967	0.5694		
Total	53	95.1151			

Grand Mean 4.3256 CV 17.44

Appendix 19: Analysis of Variance Table for Protein

Source	DF	SS	MS	F	P
PM	5	4.9729	0.99458	1.52	0.2089
SP	2	19.6041	9.80207	14.95	0.0000
PM*SP	10	0.4971	0.04971	0.08	0.9999
Error	36	23.6011	0.65559		
Total	53	48.6752			

Grand Mean 37.160 CV 2.18

Appendix 20: Analysis of Variance Table for 1000 seed weight

Source	DF	SS	MS	F	P
PM	5	556.31	111.263	7.14	0.0001
SP	2	1245.59	622.796	39.94	0.0000
PM*SP	10	4.63	0.463	0.03	1.0000
Error	36	561.33	15.593		
Total	53	2367.87			

Grand Mean 253.76 CV 1.56

Appendix 21: Analysis of Variance Table for Vigour

Source	DF	SS	MS	F	P
PM	5	177.568	35.514	383.93	0.0000
SP	2	317.493	158.747	1716.18	0.0000
PM*SP	10	62.827	6.283	67.92	0.0000
Error	36	3.330	0.093		
Total	53	561.218			

Grand Mean 27.211 CV 1.12

Appendix 22: Analysis of Variance Table for Vigour index

Source	DF	SS	MS	F	P
PM	5	184183	36837	16576.5	0.0000
SP	2	1.215	6075154	2733819	0.0000
PM*SP	10	30951.0	3095	1392.79	0.0000
Error	36	80.0000	2		
Total	53	1.237			

Grand Mean 2089.4 CV 0.07

ANALYSIS OF VARIANCE FOR THE SEED HEALTH OF *P. ELATA*

Appendix 23: Analysis of Variance Table for *Aspergillus niger*

Source	DF	SS	MS	F	P
PM	5	173.648	34.730	24.68	0.0000
SP	2	321.593	160.796	114.25	0.0000
PM*SP	10	93.296	9.330	6.63	0.0000
Error	36	50.667	1.407		
Total	53	639.204			

Grand Mean 13.574 CV 8.74

Appendix 24: Analysis of Variance Table for *Fusarium moniliforme*

Source	DF	SS	MS	F	P
PM	5	94.000	18.8000	18.80	0.0000
SP	2	196.000	98.0000	98.00	0.0000
PM*SP	10	28.000	2.8000	2.80	0.0113
Error	36	36.000	1.0000		

Total 53 354.000

Grand Mean 9.0000 CV 11.11

Appendix 25: Analysis of Variance Table for *Penicillium*

Source	DF	SS	MS	F	P
PM	5	173.648	34.730	24.68	0.0000
SP	2	321.593	160.796	114.25	0.0000
PM*SP	10	93.296	9.330	6.63	0.0000
Error	36	50.667	1.407		
Total	53	639.204			

Grand Mean 9.5741 CV 12.39

Appendix 26: Analysis of Variance Table for *Rhizopus*

Source	DF	SS	MS	F	P
PM	5	94.000	18.8000	18.80	0.0000
SP	2	196.000	98.0000	98.00	0.0000
PM*SP	10	28.000	2.8000	2.80	0.0113
Error	36	36.000	1.0000		
Total	53	354.000			

Grand Mean 13.000 CV 7.69

ANALYSIS OF VARIANCE FOR THE STORAGE EXPERIMENT OF *S. RHINOPETALA*

Appendix 27: Analysis of Variance Table for Carbohydrate

Source	DF	SS	MS	F	P
PM	5	66.549	13.3098	13.31	0.0000
SP	2	120.677	60.3387	60.34	0.0000
PM*SP	10	30.128	3.0127	3.01	0.0072
Error	36	36.000	1.0000		
Total	53	253.354			

Grand Mean 14.608 CV 6.85

Appendix 28: Analysis of Variance Table for oil

Source	DF	SS	MS	F	P
PM	5	68.972	13.794	23.65	0.0000
SP	2	221.906	110.953	190.20	0.0000
PM*SP	10	39.599	3.960	6.79	0.0000
Error	36	21.000	0.583		
Total	53	351.477			

Grand Mean 20.497 CV 3.73

Appendix 29: Analysis of Variance Table for Germination percentage

Source	DF	SS	MS	F	P
PM	5	739.33	147.87	63.37	0.0000
SP	2	2800.33	1400.17	600.07	0.0000
PM*SP	10	73.67	7.37	3.16	0.0054
Error	36	84.00	2.33		
Total	53	3697.33			

Grand Mean 81.778 CV 1.87

Appendix 30: Analysis of Variance Table for Moisture content

Source	DF	SS	MS	F	P
PM	5	57.566	11.5133	46.05	0.0000
SP	2	40.758	20.3790	81.52	0.0000
PM*SP	10	24.098	2.4098	9.64	0.0000
Error	36	9.000	0.2500		
Total	53	131.422			

Grand Mean 4.9106 CV 10.18

Appendix 31: Analysis of Variance Table for Protein

Source	DF	SS	MS	F	P
PM	5	4.35600	0.87120	1.49	0.2162
SP	2	21.0000	10.5000	18.00	0.0000
PM*SP	10	2.999E-30	2.999E-31	0.00	1.0000
Error	36	21.0000	0.58333		

Total 53 46.3560

Grand Mean 18.820 CV 4.06

Appendix 32: Analysis of Variance Table for 1000 seed weight

Source	DF	SS	MS	F	P
PM	5	202.83	40.567	1.62	0.1790
SP	2	1454.33	727.167	29.09	0.0000
PM*SP	10	975.67	97.567	3.90	0.0012
Error	36	900.00	25.000		
Total	53	3532.83			

Grand Mean 765.72 CV 0.65

Appendix 33: Analysis of Variance Table for Vigour

Source	DF	SS	MS	F	P
PM	5	185.768	37.154	37.15	0.0000
SP	2	262.453	131.227	131.23	0.0000
PM*SP	10	70.267	7.027	7.03	0.0000
Error	36	36.000	1.000		
Total	53	554.488			

Grand Mean 25.706 CV 3.89

Appendix 34: Analysis of Variance Table for Vigour index

Source	DF	SS	MS	F	P
PM	5	325818	65164	27927.2	0.0000
SP	2	8148853	4074426	1746183	0.0000
PM*SP	10	23209	2321	994.67	0.0000
Error	36	84	2		
Total	53	8497963			

Grand Mean 1785.2 CV 0.09

ANALYSIS OF VARIANCE FOR THE SEED HEALTH OF *S.RHINOPETALA*

Appendix 35: Analysis of Variance Table for *Aspergillus flavus*

Source	DF	SS	MS	F	P
PM	5	132.000	26.4000	21.60	0.0000
SP	2	196.000	98.0000	80.18	0.0000
PM*SP	10	35.333	3.5333	2.89	0.0093
Error	36	44.000	1.2222		
Total	53	407.333			

Grand Mean 17.889 CV 6.18

Appendix 36: Analysis of Variance Table for *Botridiplodia theobrome*

Source	DF	SS	MS	F	P
PM	5	94.000	18.8000	18.80	0.0000
SP	2	196.000	98.0000	98.00	0.0000
PM*SP	10	28.000	2.8000	2.80	0.0113
Error	36	36.000	1.0000		
Total	53	354.000			

Grand Mean 12.000 CV 8.33

Appendix 37: Analysis of Variance Table for *Penicillium*

Source	DF	SS	MS	F	P
PM	5	94.000	18.8000	18.80	0.0000
SP	2	196.000	98.0000	98.00	0.0000
PM*SP	10	28.000	2.8000	2.80	0.0113
Error	36	36.000	1.0000		
Total	53	354.000			

Grand Mean 10.000 CV 10.00

Appendix 38: Analysis of Variance Table for *Aspergillus niger*

Source	DF	SS	MS	F	P
PM	5	94.000	18.8000	18.80	0.0000
SP	2	196.000	98.0000	98.00	0.0000
PM*SP	10	28.000	2.8000	2.80	0.0113
Error	36	36.000	1.0000		

Total 53 354.000

Grand Mean 13.000 CV 7.69

ANALYSIS OF VARIANCE FOR THE DORMANCY-BREAKING ACTIVITIES OF *S. RHINOPETALA*

Appendix 39: Analysis of Variance Table for number of days to emergence

Source	**DF**	**SS**	**MS**	**F**	**P**
Methods	6	289.376	48.2294	18.92	0.0000
Error	17	43.333	2.5490		
Total	23	332.710			

Grand Mean 11.971 CV 13.34

Appendix 40: Analysis of Variance Table for Germination

Source	**DF**	**SS**	**MS**	**F**	**P**
Methods	6	21576.0	3596.00	326.33	0.0000
Error	17	187.3	11.02		
Total	23	21763.3			

Grand Mean 68.833 CV 4.82

Analysis of Variance for *G. cedrata*

Appendix 41: Analysis of Variance Table **for Germination**

Source	**DF**	**SS**	**MS**	**F**	**P**
Desiccant	4	30.0000	7.50000	3.21	0.0611
Error	10	23.3333	2.33333		
Total	14	53.3333			

Grand Mean 10.333 CV 14.78

Appendix 42: Analysis of variance table for days to deterioration

Source	DF	SS	MS	F	P
Treat	5	885.100	177.020	53.59	0.0000
Error	12	39.640	3.303		
Total	17	924.740			

Grand Mean 10.800 CV 16.83

Appendix 43: Analysis of variance table for Viability

Source	DF	SS	MS	F	P
Treat	5	23038.4	4607.69	19.49	0.0000
Error	12	2837.3	236.44		
Total	17	25875.8			

Grand Mean 41.889 CV 6.7

Appendix 44: Analysis of Variance for *G. cedrata* insect damage regression analysis Source

DF	SS	MS	F	P
Regression	1	3366.68	3366.68	236.97	0.0000
Residual	16	227.32	14.21		
Total	17	3594.00			
Lack of Fit	15	226.818	15.1212	30.24	0.1419
Pure Error	1	0.500	0.5000		

Cases Included 18 Missing Cases