BOTSWANA UNIVERSITY OF AGRICULTURE AND NATURAL RESOURCES



IDENTIFICATION OF MONGONGO (*SCHINZIOPHYTON RAUTANENII*) AND MORAMA (*TYLOSEMA ESCULENTUM*) INSECT PESTS AND THEIR NATURAL ENEMIES IN BOTSWANA.

A dissertation submitted to the Department of Crop Science and Production in partial fulfillment of the requirements of the award of the Master of Science Degree in Crop Science (Crop Protection)

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APPROVAL

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DECLARATION

I hereby declare that this submission is my original work and has not been presented for another Degree in this or any other university.

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AUTHOR'S NAME AND SIGNATURE

DATE

DEDICATION

This Thesis is dedicated to my loving fiancée Ms. Kelebonye Ramolekwa who always encouraged and stood by me during the journey of my studies and my supervisors who has been like a parent to me. To all my colleagues who participated in my academic life and to God who is the leader of my life.

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ABSTRACT

The study was conducted at the Botswana University of Agriculture and Natural Resources (BUAN) Sebele, Gaborone. Sampling sites for this study was done in Kweneng district and Okavango district, and the experiments were conducted in the entomology lab during 2016/2017 season. Farmer's knowledge on insect pests of morama and mongongo were investigated by interviewing 150 users of morama in Kweneng district and 150 users of mongongo in Okavango. Most of the farmers gathered mongongo and morama in the forest and stored them in woven polypropylene bags. They considered insect, wild and domesticated animals as a major constraint of mongongo and morama production. Beetles and caterpillars were identified as the major insect pests of the fruits and seeds in storage. None of the farmers use pesticides to control these pests. Majority of the respondents do not use any pest control. Eleven different insect species were identified to be hosted by mongongo tree in the forest and two insect species in storage (Lasioderma serricorne and Plodia interpuctella). Six insect species were hosted by morama in the forest and two insect species in storage (Plodia interpuctella and Tribolium confosum). There was no significant difference (P>0.05) between storage methods (jute bag, woven polypropylene bag and bottle container) on managing the infestation insect population. Natural enemies (B. *hebetor*) has significantly effect (P<0.05) on the population of storage pest (*P.interpuctella*).

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ACRONYMS

ANOVA	Analysis of Variance
ARC	Agricultural Research Council
BGCSE	Botswana General Certificate of Secondary Education
BUAN	Botswana University of Agriculture and Natural Resources
Ca	Calcium
Cm ³	Cubic centimetre
CO ₂	Carbon dioxide
COX 1	Cyclooxygenase-1
DHL	Dalsey Hillblom Lynn
DNA	Deoxyribonucleic acid
FAO	Food and Agricultural Organisation
Fe	Iron
FST	Food Science Technology
h	Hour
Κ	Potassium
Kj	Kilojoles
Km	Kilometer
LSD	Least Significant Difference
Mg	Magnesium
mg	Milligrams
ml	Milliters
mm	Millmeter
O ₂	Oxygen

O'LEVEL	Odinary Level
Р	Phosphorus
PICS	Purdue Improved Crop Storage
PP	Polypropylene
PROC GLM	General Linear Model Procedures
PROC MIXED	Mixed Model Procedure
RV's	Retroviruses
SAS	Statistical Software
SASSCAL	Southern African Science Service Centre for Climate Change and Adaptive Land Management
spp	Species
SPSS	Statistical Package for the Social Sciences
USA	United States of America
Zn	Zinc

CHAPTER ONE

INTRODUCTION

1.0 General introduction

The demand for food is increasing in the world due to increase in human population and this call for increase in food production to ensure that the high demand for food is met. Provision of food is going to continue to be a challenge facing mankind, especially in both under developed and developing parts of the world such as Africa (FAO, 2015). In arid conditions such as Botswana, food security is still a challenge and is worsened in these areas by the erratic and unreliable rainfall and other weather phenomena (Temoso *et al.*, 2015). This situation can be helped by the use of locally adapted plants referred to as indigenous plants. Plants have been a valuable resource of food and medicine in the history of mankind (Museler, 2005). An indigenous plant is the one that occurs in a particular region, ecosystem, and habitat without direct or indirect human actions (Richards *et al.*, 1998). These plants provide food in the form of fruits, materials for a variety of utensils and medicines (Turner, 2005). In Botswana, rural communities rely on indigenous plants 40 percent of the food intake of Batswana.

Botswana is one of the countries that are well endowed with natural resources and the communities in Botswana especially those living in marginal regions have derived their livelihood from these indigenous plant resources as food, medicine, fuel, fodder, fertilizer and timber (Mogotsi *et al.*, 2005). As agricultural production intensified over the years, these indigenous resources have experienced high levels of exploitation due to land degradation, deforestation, over-grazing and bush encroachment (Letsholo, 2017). There is also decline in their usage because of lack of promotion in terms of their usability, potentials and overreliance on the exotic species. However, there is lack of robust research on how to improve or recreate the situation such as through process of domestication and cultivation of these indigenous plant species (Mogotsi *et al.*, 2005). There is a need to intensify research on indigenous plants to enable their domestication since they are adapted to the local conditions. Lack of knowledge and skills in research and development in the new and emerging crops has left the agriculture sector highly dependent on exotic and imported materials that require high inputs for increased production. Shifting from domesticated crops to that of a wide spectrum of indigenous plant species that can meet the environmental and economic challenges of this century will increase food security in the country.

Plant domestication is regarded as an evolutionary process whereby a population of plants becomes accustomed to human provision and control (Pourkheirandish and Komatsuda, 2007). Domestication is generally considered to be the end-point of a continuum that starts with exploitation of wild plants, continues through cultivation of plants selected from the wild but not yet genetically different from wild plants and ends with the adaptation to the agro ecology through conscious or unconscious human morphological selection, and hence genetic differences distinguishing the domesticated species from its wild progenitor (Zohary and Hopf, 1993). According to local communities in Okavango and Kgalagadi regions, the collection of plants from the wild for cultivation on farm (fields or home gardens) is a common practice continually being carried out under diverse agro ecosystems. Many varieties, landraces and cultivars of plants have been developed through this process to meet human demand for food, fibre, medicine and building materials (Sweeney and Mc Couch, 2007). As the public becomes more concerned about the reduction in the number of indigenous plants through climate change and over utilization, the interest in the preservation and restoration of native plant communities increases as well. This can

be accomplished through domestication of these native plants. Under domestication, these plants are likely to do well because they are adapted to local conditions, therefore, they will tend to resist damage from drought, common diseases, and herbivores if planted in that same local region. Some of the indigenous plants that are commonly used which can assist to battle desertification, climate change, food insecurity and improving the livelihoods of the vulnerable population of Botswana include morama (*Tylosema esculentum*) and mongongo (*Schinziophyton rautanenii*). Therefore, the improved production, domestication and utilization of these plants can offer unique opportunities to address and support poverty eradication efforts in Botswana. It can play a pivotal role in establishing sustainable livelihoods of small scale farmers and their families, providing food security and income. The domestication of indigenous plants, as they are adapted to the local conditions, can be one way of solving the food security problems. Food production can be increased through extension of agriculture. That is, bringing new plants under cultivation through domesticating plants that are well adapted to local conditions.

The problem of increasing food production in the rain-fed, often semi-arid, agricultural systems is a multi-factorial interaction between many biotic and abiotic factors. There is also decline in their usage because of lack of promotion in terms of their usability and potentials and overreliance on the exotic species. Worldwide, many plant communities are becoming increasingly dominated by non-native/exotic plant species (Mack *et al.*, 2000). Several studies have noted that these exotic species are having adverse effects on native plant communities and altering ecosystem-level processes (Alvarez and Cushman, 2002). However, important among these is the failure of resource-poor farmers to prevent losses due to pre-and postharvest pests. Therefore, bringing the indigenous plants under domestication has to be preceded by studying these factors that can hinder their production. This study is aimed at looking at insects as a factor that can affect the production of morama and mongongo. This is because pests can limit food crop harvests. Studies indicate that losses due to pests overall in Africa are in the region of 30 % (Oerke and Dehne, 2004). While crop pests are a problem in all cropping systems globally, their impact is much greater in poorer communities especially where poverty, limited knowledge and poor agricultural systems are pronounced. Much of subsistence farming for these people is conducted without access to effective crop protection knowledge or resources (Njuki *et al.*, 2004).

1.1 Morama (*Tylosema esculentum*)

The morama bean was described in a National Academy of Science USA, (1979) report as a legume crop of considerable potential in arid land agriculture. It is native to the Kalahari Desert and neighbouring sandy semi-arid regions of Southern Africa, in particular Namibia, Botswana and South Africa (Hartley *et al.*, 2002). The bean has been an important food source for the people of the Kalahari Desert for centuries (Bower *et al.*, 1988) and a staple food for the Basarwa people of Kgalagadi, Botswana (Amarteifio and Moholo, 1998). It occurs naturally in the arid, dry parts of Southern Africa. The plant has high nutrient value in the seeds and tubers, rich in protein, oil and starch.

1.1.1 Origin and History of morama

Tylosema esculentum is indigenous to Southern Africa and has been used by the San people for as long as history can tell. It occurs in South Africa (Western, North West and Northern Cape); Botswana (Kgalagadi); Eastern and Northeastern of Namibia (Castro *et al.*, 2005). The plant grows in sandy soils which have limited water- holding capacity and is frequently exposed to very high light intensity, extreme temperature and prolong drought (Mitchell *et al.*, 2005).

1.1.2 Taxonomy of morama bean

Morama bean (*Tylosema esculentum* Burchell A. Schreiber) also known as gemsbok bean, marama bean, moramaboontjie, elandboontjie, braaiboonjie, marumana, tsi, tsin, gami, and ombanui is a wild perennial legume belonging to the family Fabaceae, subfamily Caesalpinioideae, and is thus related to *Cercis* and *Bauhinia* (Wunderlin *et al.*, 1981). Before its establishment as a separate genus, *Tylosema* was included in *Bauhinia* (Castro *et al.*, 2005). There are four other known species within the genus, namely, *T. fassoglense, T. argenteum, T. humifusum and T. angolense* (Castro *et al.*, 2005).

1.1.3 Biological Descriptions



Figure 1: Morama plant

Tylosema esculentum is a perennial species, producing a prostrate vine with numerous prostrate stems of up to three meters in length which spread from an enormous woody tuber below the ground (Powell, 1987). The plant has a big tuber which weighs 1kg (Van der Maesen, 2006) and up to 10kg (Mmonatau, 2005). The leaves are deeply two-lobed, hairless, and firm in texture. Attractive bright yellow flowers are born along the stems, each with erect petals and stamens. In its natural stands, morama bean takes between 18-24 months to reach reproduction maturity. It

takes between 8-21 days to germinate on wet soils and then the plant grows vegetative for the next 5-6 months.

Morama is a potential crop for arid areas where few conventional crops can survive and there is increasing interest in the cultivation of morama bean (Nepolo *et al.*, 2009). It grows at altitudes of between 1 000 and 1 500 m with 300 to 700 mm rainfall, and at a minimum temperature above 15 °C and a maximum of approximately 33 °C (Müseler and Schönfeldt, 2006). It is dormant in winter and regrows from the tuber in spring. The plant grows in well-drained, fine, generally calcareous sands, but also in regions of harder calcareous conglomerates, at pH 6 to 8, with very little organic matter, nitrate or phosphate (Lawlor, 2004).

1.1.4 Traditional use

Traditionally, morama beans are gathered by hand from the wild, roasted in hot sand and the cotyledons eaten as a snack (Amarteifio and Moholo, 1998). The beans may also be ground and made into porridge after roasting or may be boiled and eaten as other beans before they are fully ripe (Holse *et al.*, 2010). Mogotsi and Ulian (2013) stated that Morama beans are also boiled with maize meal or ground and pounded to a powder, for making a cocoa-like beverage. In some areas small tubers and young stems are also roasted and eaten, having a pleasant flavour. The seeds and tubers of this plant are not only used as a food source for both the natives of the areas in which it grows but they also have major health benefits. The morama bean plant has been shown recently to portray anti-bacterial and anti-retroviral properties and since Retroviruses (RV's) are a major source of diarrhoea in infants, the plant is traditionally used as a treatment against diarrhoea (Chingwaru *et al.*, 2011).

1.1.5 Economic Importance of Morama bean

Research has been done on morama including research directed towards domestication, despite its potential as a food and cash crop (Amarteifio and Moholo, 1998). Some farmers use morama beans as a food supplement for fattening pigs (Elfant *et al.*, 1985). The seeds produces oil with pleasant odour and can be used in food and cosmetic industries (Amarteifio and Moholo, 1998). Koenen, (2001) stated that phenolic compounds in the leaves are used to treat wounds and arthritis by the Kalahari people. Morama beans are a potential source of protein and oil. The protein content is about 30% to 35% and oil content varies from 35% to 42% (Bower *et al.*, 1988) being similar in composition to groundnuts and soybeans (Ntare, 2007). Several products are under development from morama bean including morama flour, morama butter, morama cookies, morama yoghurt, morama milk and snack roasted nuts (Van Wyk, 2011).

1.1.6 Composition and nutritional value

The composition and nutritional value of the seed compete with that of common cultivated leguminous plants (for example, pigeon peas and cow peas) as it is rich in oil and protein (Mogotsi and Ulian, 2013). The seeds are known to contain 30-39% protein and 34-43% fat (Holse *et al.,* 2010). Amaerteifio and Moholo, (1998) analysed morama seed collected from Botswana and found it to contain 3.7% ash; 33.5% crude fat; 34.1% crude protein; 4.4% crude fibre; 7760 mg/kg K; 3970 mg/kg P and 1520 mg/kg Ca. The bean also contains significant amounts of vitamins (A, B3, B6, folic acid, B12 and E) and minerals (iodine, iron and zinc) (Müseler and Schönfeldt, 2006). It is also reported to be a potential source of phytonutrients, which have been shown to contribute to health (Jackson *et al.,* 2010).

1.2 Mongongo (Schinziophyton rautanenii)



Figure 2: Mongongo tree, fruits, leaf and nuts.

The mongongo tree is the second plant under research study; it is a member of the family *Euphorbiaceae* and of the monotypic genus *Schinziophyton*. The mongongo is distributed widely throughout southern Africa. There are several distinct belts of distribution, the largest of which reaches from northern Namibia into northern Botswana, south-western Zambia and western Zimbabwe (Curtis and Mannheimer, 2005). In Botswana the tree is located in northern part of the country (Okavango region), (Curtis and Mannheimer, 2005). Mongongo is a large, spreading tree, which reaches 15-20 metres tall. It is found on wooded hills and amongst sand dunes, and is associated with the Kalahari sand soil-types. The leaves are a distinctive hand-shape (Fig 2), and the pale yellow wood is similar in characteristics to balsa, being both lightweight and strong (Palgrave, 1981). *Schinziophyton rautanenii* has multiple uses, including oil production, source of edible fruits and kernels, source of timber and wood as well as different plant parts used as herbal medicines (Chidumayo, 2016).

1.2.1 Growth requirements

S. rautanenii is found on wooded hills and amongst sand dunes, and is associated with alluvial soils near rivers, but most commonly with stabilised dunes and raised sandy plains of the deep Kalahari sands (Palmer and Pitman, 1972). Its core area is mainly in more upland areas, generally above 1200 metres, although it is found down to 200 metres. The rainfall varies over the various regions of its range and can be from 400mm to 1000mm (Keegan, 1982). Biesele *et al.* (1979) reported that the species tolerates drought and cannot tolerate areas subject to flooding. The maximum daily temperatures required often exceed 30°C in the area where the species occurs (Keegan, 1982). Although it tolerates light winter frost (Peters, 1987) temperatures below 7°C kill young plants (Anon, 1999).

1.2.2 Life cycle

A large portion of the nuts remain dormant for at least a year (Peters, 1987). The breaking of the dormancy has been tested in several studies using ethylene treatment and has had the highest degree of success in germinating seed artificially, with a germination rate of 80% or more within 6 days (Keegan *et al.*, 1989). Biesele *et al.* (1979) tested mechanical scarification, grinding off the tip of the testa until the endocarp became visible and the seed took few weeks before the radicle started to emerge. The seedlings are very quickly developing deep roots (Ronne and Joker, 2006). *Schinziophyton rautanenii* requires between 15-25 years reaching maturity, before it will bear fruit (Peters, 1987) but with irrigation they may start as early as after 4 years (Ronne and Joker, 2006). The flowers are produced in early summer (Lee, 1973), in October to November (Palgrave, 1983). Unlike many other species, the Mongongo fruit ripens after falling to the ground in April or May (Arnold *et al.*, 1985). Chimbelu (1988) reports some success with the planting of truncheons by

the Luchazi people of Zambia. Some estimates indicate yields of 200–1000 kg/ha of mongongo nuts in northern Namibia, and about 300 kg/ha in Angola (Graz, 2007).

1.2.3 Traditional uses

Von Koenen, (2001) reported that the fruit and nuts of the mongongo tree have been described as a "staple diet" in some areas by the San of northern Botswana and Namibia. Some San remove the flesh from the fresh fruit, dry it in the sun and store it for use later in the year. Both Bantu and San are reported to use the fruit of mongongo, with the modern preference being to boil the fruit and remove the tough and indigestible outer skin, and make a sweet maroon porridge. The nuts are chopped open, white kernels are peeled, pounded and boiled with water to extract the bright yellow cooking oil (Heath and Heath, 2009). Graz, (2002) stated that nuts are either eaten straight, or pounded as ingredients in other dishes. The oil from the nuts has also been traditionally used as a human body rub in the dry winter months, to clean and moisten the skin, while the hard, outer nutshells are popular as divining bones. The wood, being both strong and light, makes excellent fishing floats, toys, insulating material and drawing boards. More recently, it has been used to make dart-boards and packing cases.

In other part of the country where mongongo do not grow in abundance in the central part of Botswana such as Dagwi and Changate village, Bakalaka do not use mongongo as an edible plant to make food, but they use it to make woodcraft such as tables, stool, traditional drums, joko and wooden spoon.

1.2.4 Commercial Potential of S. rautanenii

Research by Bennett, (2006) showed that about 200,000 people are currently employed in gathering *S. rautanenii* fruits and trading in this species, and its products have the potential of generating close to US\$20 million in Southern Africa. *Schinziophyton rautanenii* and its products are traded in informal markets in Malawi, Mozambique, Zambia and Zimbabwe (Cunningham, 1993) while oil extracted from the species is exploited commercially in Zimbabwe (Chivandi *et al.*, 2008). In Zambia, the wood and timber obtained from the species is used in producing curios and other crafts that are marketed in tourist resorts (Chidumayo, 2016). Seed oil extracted from *S. rautanenii* has over the years been used in producing lubricants, soaps and personal cosmetic care products, as well as health products used in the topical treatment of various ailments and conditions such as hair dandruff, muscle spasms, varicose veins and wounds (Zimba *et al.*, 2005). Therefore, commercialization of the oil derived from the species may result in several economic activities that positively impact local communities and may contribute to household economy and their livelihoods.

1.2.5 Nutritional composition

The nutritional composition of the fruit pulp per 100 g edible portion is: water 13.4 g, energy 1307 kJ (312 kcal), protein 6.6 g, fat 0.6 g, carbohydrate 70.2 g, fibre 3.5 g, Ca 89.6 mg, Mg 195 mg, P 46.0 mg, Fe 0.7 mg, Zn 1.4 mg, thiamine 0.28 mg, riboflavin 0.11 mg, niacin 0.12 mg, ascorbic acid 8.5 mg (Graz, 2007). According to Wikipedia, (2009), mongongo shelled nuts, per 100 grams consist 57 g fat (44% polyunsaturated, 17% saturated and 18% monounsaturated), 24 g protein, 193 mg calcium, 4 mg zinc, 2.8 mg copper and 565 mg vitamin E (almost entirely as y-tocopherol).

In spite of aforementioned numerous benefits of these crops, their production in Botswana is not done. There is a ready local market for these crops which is supposed to be an incentive for increased production. However, insect pests such as foliar and also in storage are a major constraint for the morama and mongongo under cultivation. Due to lack of documentary on insect pests of morama and mongongo, research is needed to determine farmers knowledge on insect pests of morama and mongongo. This study was conducted in two districts, which are Okavango and Kweneng district, where mongongo and the morama bean plants grow in natural abundance and the residents of those areas highly utilise those plants for food, medicine etc. Questionnaire was used to determine their knowledge of insects of morama, mongongo and the damage they cause. The people were asked to indicate how they manage morama insect pests, and effectiveness of the management options. The information gathered during the survey was used as background information of possible pests of morama and mongongo.

1.3. Justification of study

Botswana is undergoing economic diversification drive and one of the areas being looked at is agriculture. One way of improving or increasing agricultural output is through domestication of indigenous high value plant species and their commercialization. Research on indigenous plant species like morama and mongongo is ideal for battling desertification, food insecurity and improving the livelihoods of vulnerable population of Botswana especially the rural communities. Improved production and utilization of these plant species offers unique opportunities to address and support poverty eradication efforts in Botswana and the region, by playing an essential role in the sustainable livelihoods of small scale farmers and their families, providing food security and

income to the most vulnerable group, the women and children. If these plants are domesticated, they can help reduce hunger and poverty.

Although indigenous crops generally have the ability to grow in a wide range of environmental conditions, biotic stress in the form of diseases and pests is considered as a major constraint limiting their vegetative and reproductive growth and yield. Morama bean and mongongo has been demonstrated to have vast agronomic potential and because of their good adaptation to climatic conditions of some parts of Botswana, they might do well if domesticated. However, studies on the morama bean and mongongo have been mostly on the chemical, compositional and nutritional properties, with limited investigation on insects of mongongo and morama which can assist in the domestication process. Currently the potential pests of morama and mongongo are poorly understood and this is very crucial to cropping efforts. Without this knowledge, domestication of these plants cannot be achieved.

Despite the domestication potential of these plants as arid agricultural crops and as traditional source of food in Botswana, little is known about their insect pest and natural enemies. Therefore this study will bring background knowledge on which insect pests affect morama and mongongo and suitable methods to control them. These will enhance their domestication process and their utilization.

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1.4. Objectives

- 1.4.1 To assess farmers knowledge on insect pests of morama and mongongo.
- 1.4.2 To determine diversity and abundance of insect pests and natural enemies of wild morama and mongongo plants in the Kgalagadi and Okavango regions.
- 1.4.3 To determine diversity and abundance of storage insect pests and natural enemies of morama and mongongo nuts in storage.
- 1.4.4 Effect of natural enemies and storage methods on population of storage pest of the morama and mongongo nuts.

1.5. Hypotheses

1.5.1 Null hypothesis: insect pests do not attack indigenous plant species (mongongo and morama).

Alternative hypothesis: insect pests attack indigenous plant species (mongongo and morama).

1.5.2 Null hypothesis: natural enemies and storage methods do not supress potential population of insect pests of morama and mongongo.

Alternative hypothesis: natural enemies and storage methods supress potential population of insect pests of morama and mongongo.

CHAPTER TWO

LITERATURE REVIEW

2.1. Potential effects of insects on Mongongo

Though mongongo has many uses and potential benefits, the potential effects of insects on it cannot be ignored. There is no clear-cut documentation on insect pests associated with S. rautenenii. Literature that is available just mentions that it can be attacked by insects and diseases and not specify which ones. This plant produces a sturdy and thin exocarp which is pale green when the fruit falls from the tree (Biesele et al., 1979). When the fruit dries the surface appears slightly wrinkled. It has been reported that the exocarp is often eaten by insects (Peters, 1987). Parker, (1978) reported that various fungi and insects attack the seed and wood of the tree. In addition, Botelle, (1999) reported that yield is affected by consumption from mammals, birds, insects, and by fungal attacks. Fallen fruits are susceptible to attack by moth larvae which eventually eat all the fleshy parts. The wood is not durable and susceptible to termite attack (Graz, 2007). Insects and animals destroy the fruit where they fall (Graz, 2000). The author further said that there are also no insects known to attack the nut in storage, either in the bush or at the home. These studies did not specify the type of insects that are associated with this plant hence more studies are needed to identify the insects related to mongongo which is valuable information for domestication processes.

Even though no insects are clearly and specifically documented for mongongo, some of the plants in the same family (Euphorbiaceae) as mongongo which include *Jatropha curcas* Linn are known to be attacked by specific insect pests. *Jatropha curcas* has been found to be attacked by insect pest like *Pachycoris klugii* (Heteroptera: Scutelleridae) in a study done in Nicaragua (Peredo, 2002). This insect was found to be feeding on flower and fruit, causing malformation of the seeds, which can reduce the quality of the oil. *Leptoglossus zonatus* (Dallas, 1852) (Heteroptera: Coreidae) has also been observed in Nicaragua on *Jatropha curcas* plants. This is polyphagous, and also infests sorghum, maize, and tomato (Grimm and Maes, 1997). In Senegal, *Oedaleus senegalensis* (Krauss, 1877) (Orthoptera: Acrididae) was observed to cause damage to seedling leaves of *J. curcas* (Grimm and Maes, 1997). The larvae and adults of *Calidea panaethiopica* (Kirkaldy, 1909) (Scutelleridae) have also been observed in Senegal on *J. carcus* by Terren *et al.* (2012). Other reports of insect pests of *J. curcas* were made in Brazil (Foidl *et al.*, 1996), in India (Shanker and Dhyani, (2006) and in West Africa (Terren *et al.*, 2012). The revelation from the *Jatropha* findings which is a member of mongongo family shows that it is possible to have insects affecting mongongo.

Domestication of plants can bring with it problems such as diseases and insects. Tchoundjeu *et al.* (2006) reported that the strategy of practicing domestication of agroforestry species in West and Central Africa was done based mostly on the individuals with desirable characteristics. Some of the species enrolled in the program including *Ricinodendron heudotii* (Euphorbiaceae family) had problems with insects and other pests. Four types of caterpillar were found to defoliate the plant and these were *Lobobunaea phaedusa* (Drury), *Imbrasia petiveri* (Guerin-meneville), *I. obscura* (Butler) and probably *I. melanops* (Bouvier) and *I. epimethea* (Drury) (Latham, 2003). In Cameroon a psyllid, *Dichlidoplebia xuani* was found to be attacking the buds of *R. heudotii* seedlings in the nursery (Messi and Tamesse, 1999). The other plant in the same subfamily with mongongo, which has been domesticated in America and Africa, is Cassava *Manihot esculenta*. Bellotti, (2002) reported that important pest of *M. esculenta* include the mite, *Mononychellus*

tanajoa, the mealybug, *Phenacoccus herrenii*, the hornworm *Erinnyis ello*, the stemborer, *Chilomima clarkei*, the fruit fly, *Anastrepha manihoti*, the thrips, *Scirtothrips manihoti*, and the whiteflies, *Bemisia tabaci*, *Aleurotrachelus socialis*, and *Aleurothrixus aepim*. The report further indicated that extended attacks (3-6 months) by mites, mealybugs, thrips, and whiteflies can cause up to a 79% loss in root yields (Bellotti *et al.*, 1999). The revelations of the studies of related plant species to mongongo indicate that this plant has potential to be affected by insects hence the need of this study. Therefore, this study aims to investigate insects associated with *Schinziophyton rautanenii* in Botswana and to identify those that can be pests of this plant.

2.2 Potential pests of morama

Morama is now being considered as a new alternative crop where it occurs. It has been demonstrated to have vast agronomic potential because of its fruits yield ability and good adaptation to climatic conditions of some parts of Botswana which are characterised by low rainfall and nutritionally poor soils (Hartley *et al.*, 2002). This makes it a potential domestication crop for semi-arid and arid agriculture. Due to its potential under these harsh conditions, there are interests in domesticating it through cultivation. In a field experiment carried out in Texas (USA) the morama bean plant successfully took 4 years to develop and reach maturity and produce edible seeds (Powell, 1987). Successful cultivation was also done in Australia, Kenya, Israel, Namibia and South Africa (Van der Maesen, 2006). Other cultivation trials were also carried out in Botswana at Botswana College of Agriculture (Ramolemana *et al.*, 2002). Even with this research work done on morama plant, there is lack of reports of insect pests of morama bean.

Majority of research works focused on exotic species with an economic or commercial value susceptible to insect pests and diseases and neglecting indigenous plant species that are an integral

component of the environment such as morama. Despite all the good known about morama plant, there is lack of information about its associated insects both beneficial and pests. Knowledge about these insects will allow a better understanding of the yield potential of morama, its functioning ability in the absence and presence of insects and even come up with proper techniques to solve the intricate problems that might come from these insects. Nepolo *et al*, (2009) reported that the natural population of morama bean is under pressure from animals and human being through exploitation of the seeds. Insect pests have been observed on the wild stand of morama causing seed damage, however, the identity of the insects is yet to be ascertained (Jackson *et al.*, 2010). Some of the insects observed on morama bean flowers were reported to be the pollinators. Jackson (2010) reported that a bee (on its own) has been observed and implicated to be the pollinator for morama and the taxonomic identity of the bee was still being investigated. Mbewe, (1992) observed that ants were found on the flowers and suggested that they may be the main pollinators, while Beattie *et al.* (1984) reported that ants are nectar robbers of morama rather than pollinators.

Due to the limited documentation about insect pests on morama, I believe that morama, as it is common with other legume crops, may become infested with insect from seedling to storages some of them being pests. Even though there is no documentation on the insect pests of morama, there are studies done on the plants of the same subfamily Caesapinioideae as morama. *Senna siamea* in the same subfamily is reported to be liable to browsing damage, susceptible to attack by scale insects, and sapwood is susceptible to *Lyctus* beetles. In Vietnam, the butterfly *Captosilia crocale* was reported as a serious pest. Its larvae were found feeding on the foliage of *S. siamea* (Orwa *et al.*, 2009).

Before its establishment as a separate genus, *Tylosema* was included in *Bauhinia* (Castro *et al.*, 2005). Orwa *et al.* (2009) has reported *Buahinia purpurea* to be attacked by unidentified species

of borers, mites and larvae of several insects. Trees of *Buahinia variegata* host larvae of several insects. Mature nymphs of the *Psylla simlae* (Hemiptera) feed on sap of leaves and young twigs. Leaves and flowers infested by nymphs shrivel and fall (Orwa *et al.*, 2009). Malaysian Locust *Valanga nigricornis* probably the most important of Brunei's grasshopper pests, this species feeds on a variety of plants including ornamental shrubs and ground cover plants may also be attacked and *Bauhinia* seem to be particular favourites. *Caryedon gonagra* (Groundnut bruchid-Chrysomelidae) has been found feeding in several species of *Bauhinia* and *Cassia* in Thailand (Eungwijarnpanya and Hedlin, 1984). *Caryedon serratus* attacks the seeds of *Bauhinia spp*. The adults lay eggs on the pods and then the larvae bore into the seed often completely destroying the seed and hence preventing germination (El Atta and Abdel Nour, 1995).

The groundnut seed-beetle *C. serratus* is an African species (Decelle, 1981) which was first described in 1790 from specimens collected in Senegal. Its hosts belong to the family Caesalpiniaceae. The commonest plants in Africa are *Piliostigma tbonningii* and *P. reticdatum*, *Bauhinia rufiscens* and *Tamaridus indica*. *Bauhinia lunarioides* (Bconjesta) young trees are infested by sucking insects such as aphids, thrips, whiteflies and psyllids. During dry months in dusty conditions spider mites attack the young trees of *Bauhinia*. Doucette, (1962) reported that in South Africa, Lewis spider mite *Eotetranychus lewisi* (McGregor) recorded from the *Bauhinia sp*. False Codling Moth (*Thaumatotibia leucotreta*) larvae chew holes into the fruit which become filled with frass on the host plant *Bauhinia galpinii* (Venette *et al.*, 2003). The giant whitefly, *Aleurodicus dugesii*, was reported to directly cause damage to *Bauhinia galpinii*. Both nymphal and adult whiteflies feed by inserting their needlelike mouthparts into the vascular tissue or phloem of the leaves and suck out the plant sap (Dreistadt *et al.*, 2001).
2.3 Potential storage pests and their control

There is no information about the storage pests of morama and mongongo. However there is literature on insect pests affecting other products in storage. Insect pests that are associated with stored products such as cereals, legumes, oilseeds, dried fruits, nuts, and many other value-added whole or processed food products cause substantial economic and quality losses to the products (Pimentel, 1991). These insects can also be potential pests of stored morama and mongongo. The most economically important families of insects that infest stored products are in the order Coleoptera and Lepidoptera. About 600 species of beetles and 70 species of moths are associated with stored products in various part of the world (Arbogast, 1991). Ghimire, (2008) stated that stored-product moths are among the most destructive insects of stored grain and processed food throughout the world. Larvae of these moth species do their damage by directly consuming various stored products and also by subsequent silken webbing of their food into contaminated masses.

2.3.1 Confused flour Beetle Tribolium confusam

2.3.1.1 Taxonomy and history

The genus *Tribolium* is a member of the Order Coleoptera, Family Tenebrionidae and Subfamily Ulominae. The confused flour beetle originated in India, but now found all over the tropical, subtropical and warm temperate regions of the world. It is one of the common pests of stored products.

2.3.1.2 Biology

The female lays eggs in damaged wheat grain, grain dust and flour throughout their adult life. The number of eggs laid depends on temperature; 2 eggs per day at 25°C and 11 eggs per day 32.5°C

(Howse, 1962). Within 5 to 12 days the eggs hatch. The larvae undergo 6-7 moulting and become full grown in 22-25 days at 30°C. The pupae take 4-5 days and the adult can live for about six months (Howe, 1956).

2.3.1.3 Damage

Confused flour beetle have chewing mouth parts and can penetrate deeply into the stored commodity. The larvae and adults feed on a wide range of stored grain products such as flour, cereals, beans, nuts and seed (Via, 1999). Larvae and adult feed in the internal part of the seed contaminate products with faeces and promote moulds by increasing moisture content of grain through their respiration.

2.3.1.4 Control

According to Koehler, (2003) the first step in managing an infestation is to find the source of infestation. Clean the store between harvests, removing and burning infested residues, immersing grain sacks in boiling water and removing wood from stores. Confused flour beetle can feed and survive on smallest bits of grains. Remove the adult insects and larvae from the grain by sieving and discard them in a sealed bag or container. Adding ash and clay to the grain can reduce insect numbers by causing the insects to die from desiccation. Placing infested material in a freezer for four to five days kills adults but these beetles may survive freezer times shorter than this (Arbogast *et al.*, 2000).

2.3.2 Indian meal moth *Plodia interpunctella*

The Indian meal moth, *Plodia interpuctella* (Lepidoptera, family Pyralidae), is a moth in the subfamily Phycitinae. It is a worldwide pest of stored and processed durable food commodities

(Rees, 2004). This insect can complete its life cycle in 27 to 52 days depending on factors such as temperature, food and others (Mohandass *et al.*, 2006). It becomes reproductively active within 24 hours of eclosion from the pupae. Mating occurs when "calling" or pheromone releasing female induce male to copulate in a sequence of events (Rees, 2004). Mated females lay 100- 150 eggs on average during their lifetime, depending on the quality of food. The mating and laying of eggs occurs about three days after adult emergence. The eggs can be laid singly or clusters and are oviposited directly on food source. The eggs hatch in 7-8 days at 20°C and 3-4 days at 30°C. Upon hatching the larvae can complete their development in 6-8 weeks at a temperature ranging from 18-35°C. The larvae go through five instars and pupate. The pupal stage can last from 15-20 days at 20°C and 7-12 days at 30°C.

2.3.2.1 Damage

Plodia interpunctella is an external feeder. The larvae continuously spin a silken web both inside and on top of the food surface and feed within the web. The webbing contains larva excreta (frass) and exuvia (cast skin) and give unpleasant odor to the commodities. Silken webbing causes a mat covering the surface of the commodity and reduces the product quality (Phillips *et al.*, 2000).

2.3.2.2 Control

Grieshop, (2005) reported that population suppression has been observed in the laboratory using egg and larvae parasitoids. The larvae parasitoid, *Bracon hebetor* and the egg parasitoid *Trichogramma pretiosum* have demonstrated Indian meal moth population suppression. *T. pretiosum* offered 37.3 percent suppression rate while *B. hebetor* provided a 66.1 percent suppression rate. Fields, (1992) stated that low temperature storage and heat treatment of storage facilities have potential to control Indian meal moth. Johnson *et al.* (1997) reported that at 10°C a

stress is imposed on adult moths causing an increase in adult mortality and surviving adults exhibit decreased egg production. Chandhry, (1997) reported that fumigation is an important management option that can control all stages of Indian meal moth. Some *P. interpunctella* strains have developed low-levels of resistance to phosphine. Cox, (2004) reported that pheromone traps have used for mass trapping, attracting and killing, mating disruption, as repellents, and as specific behavioral stimulants or deterrents for *P. interpunctella* but did not receive widespread commercial use.

2.3.3 Parasitoid wasp Bracon hebetor

Bracon hebetor Say (Hymenoptera: Braconidae) is a gregarious, idiobiont ectoparasitoid that attacks larvae of several species of Lepidoptera, mainly pyralid moths infesting stored products. It is an important potential biological control agent of stored product moths (Brower *et al.*, 1996). *B. hebetor* females first paralyze their host larva by stinging and then laying variable numbers of eggs singly on or near the surface of paralyzed hosts (Antolin *et al.*, 1995). The paralyzed host larvae are then used as food sources for developing wasps and also for the adult females. Normally the female *B. hebetor* paralyzes a number of larvae and returns afterwards to oviposit on some of them. The paralysis is ultimately fatal, though paralyzed larvae may continue to live for nearly a month if not parasitized and consumed by wasp larvae (Richards and Thomson, 1932).

2.3.3.1 Life cycle of B. hebetor

The *B. hebetor* females prefer to attack and oviposit on last instar (fifth) larvae, although younger instars will also be stung and used (Benson, 1973b). *B. hebetor* females continually produce eggs throughout their lifetime (synovigenic) and reproductive females engage in host-feeding which is essential for the maturation of additional eggs (Benson, 1973a). Egg development time varies from

12 h at temperatures of 27-34°C, to eight days when at 4-14°C. There are four larval instars with total larval developmental time 36 h to five days, depending upon rearing temperatures (Benson, 1973a). The last instar larvae spin small white cocoons before pupation, either on or near the host remains. The pupal period lasts from three to four days. The overall development time from oviposition to adult emergence is 10-13 d at 27°C (Strand and Godfray, 1989). *B. hebetor* is able to live and be active in all stages between the temperatures of 14.5-40°C (Noor-ul-Ane *et al.*, 2018).

2.3.3.2 Bracon hebetor as biological control agent

B. hebetor is primarily known as a parasitoid of pyralid moths in the sub-family Phycitinae that are associated with durable stored food products, and include the Indianmeal moth, Plodia interpunctella (Hübner), Mediterranean flour moth, Ephestia kuehniella (Zeller), tobacco moth E. elutella (Hübner), driedfruit moth, Vitula edmansae (Packard), Moodna sp, and almond moth, E. cautella (Walker) (Brower et al., 1996). According to Krombein et al. (1979) B. hebetor also attacks several other non-phycitine pyralid moths, such as the rice moth, Corcyra cephalonica (Stainton) (sub-family: Galleriinae), the greater wax moth, Galleria mellonella (Linnaeus) (subfamily: Galleriinae), grass moth Laetilia coccidivora (Comstock) (sub-family: Crambidae), and some species outside Pyralidae such as potato tuberworm, Phthorimaea operculella (Zeller) (family: Gelechiidae), Angoumois grain moth, Sitotroga cerealella (Olivier) (Gelechiidae) in the Nearctic region. Van Alpen and Jervis, (1996) stated that some gregarious parasitoids can optimise their reproductive potential by regulating the number of eggs on a host (clutch size) and progeny sex ratio. Bracon hebetor Say attacking larvae of Plodia interpunctella also showed similar oviposition behaviour related to host size (Taylor, 1988b) depositing more eggs on larger hosts than smaller ones.

2.4 Host suitability for parasitism

Host quality strongly influences the main components of parasitoid fitness (Godfray, 1994). Factors used to assess host suitability for parasitoids include; The numbers of hosts parasitized, numbers of eggs laid each day on each host (mean daily fecundity), development time, parasitoid survival to adulthood and Progeny sex ratio (Ghimire, 2008). King (1994) stated that female parasitoids typically allocate more male progeny, which are unfertilized eggs, to lower quality hosts, while reserving more female offspring, from fertilized eggs, for high quality hosts in order to increase her reproductive fitness.

2.5 Storage methods

Botswana is a semi-arid area, with a period of drought spell and therefore storage methods have helped in times of drought spells. Seeds are well stored for the next season of ploughing using different methods of storage. Brooker *et al.* (1992) stated that during storage the grains can lose value due to physical factors, such as temperature and humidity; chemical factors, such as oxygen supply; and biological factors, such as bacteria, fungal, insects and rodents. Therefore storage preserves the qualitative and quantitative aspects of the grains by providing unfavorable conditions for the development of insects, rodents and microorganisms (Bailey, 1974). It is therefore necessary to ensure that after harvest, the seeds are stored appropriately in different storage types that will protect from damage caused by pests.

2.5.1 Jute bag

Tortora, (1987) described a jute bag as a soft, fine and lustrous natural cellulose fibre obtained from the stem of the corchorous plant grown in India, Bangladesh, and Thailand. Advocacy, (2012)

pointed out that jute exhibits many positive properties which include reinforcement materials for composites due to its low density (1.5g/cm3), the material therefore yields considerably light weight composites. Jute fibres are 100% biodegradable and recyclable, and therefore environmental friendly. Furthermore jute has adequate aeration for the seeds to dry. Navarra and Calderon, (1973) explained that moisture content plays an important role, the lower the water present in the grain mass, the higher the mortality, due to the desiccation effect on insects by low O_2 and high CO_2 . Therefore jute hygroscopic nature permits any excess moisture in stored seeds out of the packaging, thus protecting them from damage by mildew. Chikoore, (2013) stated that in Zimbabwe the challenge they usually face is that of damage by pests especially weevils to nyemba (beans) stored in jute bags.

2.5.2. Woven polypropylene bag (maize meal bag)

Normal polypropylene bags are available in the market and easily used by farmers to store their produce. The design and composition of polypropylene bags make them highly resistant to pests that can infest stored foodstuffs and create a great deal of preventable loss. Hell, (2010) find out that mortality rate of *C. quadricollis* and *Tribolium* spp in polypropylene bags also increased with storage time.

2.6 Identification of insects

2.6.1 Morphological identification

Identification of an organism is a key to its classification and taxonomy. A detailed description of the morphology of the larva, the destructive stage, and the pupa is very important for identification

process. Timm *et al.* (2008) stated that morphological features, identifying the younger larval instars and eggs is challenging, since diagnostic characters are difficult to observe. Jalali *et al.* (2015) stated that identifying insects morphologically generally depends on adult stage and male genitalia. Adults of the species of Lepidoptera may be easily distinguished based on wing patterns and have been described elsewhere (Komai, 1999). Pacheco da Silva *et al.* (2014) stated that insects like mealybugs are morphologically very similar and are therefore difficult to tell apart, particularly for non-specialists. Therefore the methods for distinguishing between mealybug species are based on observations of the morphological characteristics of adult female specimens under the microscope. Timm *et al.* (2008) used the morphology of final instar larvae and pupae of *E. acerbella* to develop keys for distinguishing between *Epichoristodes acerbella, Cydia pomonella, Grapholita molesta* and *Thaumatotibia leucotreta* final instar larvae and pupae.

2.6.2 Molecular identification

Morphological data may also not be suitable for identifying damaged or incomplete specimens as diagnostic characters may be obscured or missing. In these cases, the use of molecular information is generally suitable, since analyses can be conducted with small tissue samples on any life stage. Molecular characterization and DNA barcoding is a taxonomic method that uses a short genetic marker in an insect DNA to identify a species, including an unknown species. The DNA barcode method of identification includes identifying insect species from any developing stage and part. Novotny *et al*, (2002) describe DNA barcoding, as a tool of DNA-based taxonomy which is in current use to identify known and unknown species based pattern of nucleotide arrangement in a

fragment of DNA of a particular species. Wilson (2012) further explained that DNA barcoding is the use of a short standardized DNA sequence (in insects, a 658 bp fragment of the mitochondrial cytochrome *c* oxidase (COX *I*) gene) to identify and assign unknown specimens to species besides facilitating the discovery of new species. The main advantage of DNA barcoding is the rapid acquisition of molecular data (Monaghan *et al.*, 2005). Mitochondria are energy-producing organelles, found in nearly every cell in nearly every plant and animal species. The mitochondrial genome in particular has turned out to be exceedingly useful in tracing evolutionary history, as it is present in all eukaryotic organisms, evolves rapidly as compared to nuclear DNA. Nuclear and mitochondrial genomes exhibit different patterns of inheritance (Behura, 2006).

CHAPTER THREE

MATERIALS AND METHODS, RESULTS AND DISCUSSION

3.1 Experiment 1: Farmer's knowledge on pest of morama in Kweneng district and mongongo in Okavango district

3.1.1 Materials and methods

3.1.1.1 Study site



Figure 3: Map of Botswana showing the locations of the study

The study about knowledge of pests was conducted in two districts, which are Okavango (mongongo) and Kweneng district (morama). These are places where mongongo and the morama bean plants grow in natural abundance and the residents of these areas highly utilise those plants for food, medicine and others. Three villages were selected in Kweneng district for morama study

and three villages in Okavango for mongongo study to administer a questionnaire. In Kweneng, survey was done at Malwelwe village (24 km northeast of Letlhakeng), Maboane village (40km south east of Letlhakeng) and Mantshwabisi village (30 km east of Letlhakeng). For mongongo, the survey was conducted in Okavango region at Shakawe/Shaikarawe, Nxamasera and Ngarange villages (Fig 3).

3.1.1.2 Survey method

People were interviewed using a semi-structured face to face interview (Questionnaire). Before beginning the survey, the questionnaire was first pre-tested in Malwelwe (Kweneng District) and then reviewed-based on pre-test findings. It was reviewed to enable clarification and streamlining of the interview questions for a smooth interviewing process. A total of 50 respondents, aged 20 years and above, were randomly picked as they passed by the village Kgotla and interviewed in each village of this study. Respondents were chosen regardless of their education or background. During the interviews, people were asked about their age, level of education and years of their stay in the village. They were also asked about their experiences with morama. Open-ended and closed questions were used to determine their knowledge of insects of morama/mongongo and the damage they cause. The people were also asked to indicate how they manage morama insect pests, and the effectiveness of the management options. The questionnaire had fifteen main questions written in Setswana (local language) and English. This was administered using simple multiple-choice format questions, simple-dichotomy statements (YES/NO) and frequency-determination (i.e. never, once and many).

3.1.1.3 Data Analysis

Responses to the questions were coded before analysis. The open-ended questions were critically analysed to identify recurrent themes which could be quantified to determine people's knowledge of the insects. Qualitative and quantitative data was analysed using SPSS. Statistics such as frequency distributions and percentages was used to analyse and report responses from the people.

3.1.2 RESULTS

3.1.2.1 A survey on pests of morama in Kweneng District

ATTRIBUTES	NUMBER OF RESPONSES			TOTAL
	MALWELWE	MABOANE	MANTSHWABISI	(AVERAGE)
GENDER				
Male	13 (26%)	22 (44%)	16 (32%)	51 (34%)
Female	37 (74%)	28 (56%)	34 (68%)	99 (66%)
MARITAL STATUS				
Yes	15 (30%)	3 (6%)	14 (28%)	32 (21.3%)
No	35 (70%)	47 (94%)	36 (72%)	118 (78.7%)
AGE				
20-29	16 (32%)	24 (48%)	9 (18%)	49 (32.7%)
30-39	21 (42%)	13 (26%)	12 (24%)	46 (30.7%)
40-49	5 (10%)	3 (6%)	4 (8%)	12 (8%)
50-59	2 (4%)	1 (2%)	11 (22%)	14 (9.3%)
60>	6 (12%)	9 (18%)	14 (28%)	29 (19.3%)
EDUCATIONAL				
LEVEL				
Primary	15 (30%)	19 (38%)	27 (54%)	61 (40.7%)
Junior Secondary	18 (36%)	22 (44%)	19 (38%)	59 (39.3%)
Senior Secondary	15 (30%)	7 (14%)	4 (8%)	26 (17.3%)
Tertiary	2 (4%)	2 (4%)	0 (0%)	4 (2.7%)
EMPLOYMENT				
STATUS				
Employed	7 (14%)	8 (16%)	10 (20%)	25 (16.7%)
Unemployed	32 (64%)	33 (66%)	30 (60%)	95 (63.3%)
Farmer	11 (22%)	9 (18%)	12 (24%)	32 (21.3%)

Table 1: Socio-economic characteristics of the respondents in Kweneng

Table 1 shows the socio-economic status of the respondents of all three villages. A total of 150 respondents were interviewed from all the three villages. The results reveal that 34% of all respondents were male and 66% female. In this study the majority of respondents were aged 20-29 with 32.7%, and the lowest aged group was age 40-49 with 8%. Overall of the respondents of all villages, 78.7% were not married and 21.3% were married.

The study revealed that from all the three villages of the study an overall of 40.7% ended upto primary school level in their education. Most of these were from the old education system when primary school education was regarded as basic education. 39.3% of the respondents acquired their education to junior certificate level, that is, the old form 2 and current form 3. 17.3% of the respondents were those who have BGCSE/O-LEVEL. 2.7% of the respondents had a tertiary background.



Figure 4: Methods of storage for morama from three villages

Figure 4 shows the methods of storage of morama in three villages used by the respondents. The respondents indicated that they store their morama in bags than in other storage methods. Seventy-

eight percent of the respondents in Malwelwe stated that they store morama in the bags, compared to 70% in Maboane and 64% Mantshwabisi. Only 22 people in all three villages indicated that there do not store morama in any container. They indicated that they just leave it in the open-space to dry and keep it away from wet or moist conditions. 10% of the respondents in Mantshwabisi said that they use open-space while only 6% said so in both Malwelwe and Maboane.



Figure 5: The shelf-life of the morama fruits in storage

Figure 5 shows the responses of the participants in three villages of study on the shelf-life of morama (freshness) after harvest, while in storage being free from pest and diseases. The respondents in all villages indicated that morama take years keeping its good quality in storage (still fresh) and also being free from the pest and diseases. 92% of the respondents in all three villages indicated that morama take years while fresh in the storage. Eight percent of the respondents indicated that morama doesn't last longer than a period of a year in the storage while fresh and free from insect pests (Fig. 5).

	FREQUENCY %			TOTAL	
	MALWELWE	MABOANE	MANTSHWABISI	(average)	
PLANT PART					
Leaves	10 (20%)	3 (6%)	8 (16%)	21 (14.0%)	
Fruits	32 (64%)	38 (76%)	36 (72%)	106 (70.7%)	
Roots	1 (2%)	1 (2%)	2 (4%)	4 (2.7%)	
Seeds	2 (4%)	5 (10%)	0 (0%)	7 (4.7%)	
Flowers	5 (10%)	3 (6%)	4 (8%)	12 (8.0%)	
STAGE OF PLANT					
Early stage	9 (18%)	13 (26%)	4 (8%)	26 (17.3%)	
Flowering	6 (12%)	5 (10%)	10 (20%)	21 (14.0%)	
Fruiting	30 (60%)	28 (56%)	32 (64%)	90 (60.0%)	
Storage	5 (10%)	4 (8%)	4 (8%)	13 (8.7%)	

Table 2: Parts and growth-stage of morama plant attacked by insects in three villages

Table 2 shows the response of people in the three villages of the study on the parts of morama plant that are mostly attacked by insect pests. It also shows the growth-stages of the morama plant that are prone to insect attack. The table shows the total number of the respondents and their percentages in the brackets. The overall total of the respondents for the three villages of the study were indicated in the last column with the average of the three villages in the brakects. The respondents indicated that morama has more field insect pests that attack the fruits during fruiting stage and few storage pests. It has also been revealed by 14% of the respondents that the leaves are susceptible to insects attack and this has been seen to occur at the early stages when the leaves are fresh (Table 2). 8.7% the respondents indicated that fruits and seeds are attacked by insect pests in storage.



Figure 6: Percentage of respondents using different control methods for morama pests

Figure 6 depicts the percentage of respondents using different control methods for morama pests. Many respondents (77.33%) stated that they do not use any pest control method in storage, while the average of 22.67% of the people stated that they used cultural methods to control storage pests. None of the respondents stated that they use chemicals to control pests as they believed that these indigenous plants are resistant to pest attack. In Maboane many respondents (96%) indicated that they do not use any control method because morama can natural stay for some years without being infested by insect pest. 48% of the respondents in Malwelwe use cultural methods such as ash and solar radiation (exposing the seeds to the sun) to control a white larvae in the storage.

Table 3 illustrates the insect species and animals that have been found in three villages feeding on morama. The respondents said that the invertebrate pests are the most important constraint in morama production. The other constraint mentioned but perceived as less important was damage caused by wild animals. Due to the low education level and lack of knowledge of names of insects by the respondents, they were forced to describe the insects instead of stating the names. The most

commonly cited insect pests of morama were caterpillar no 1, caterpillar no 2 and beetle no1 (Table 3). Caterpillar no 2 were considered the most serious pest in all three villages feeding on the pods and leaves. The other pests were considered less important in morama production (Table 3). Wild animals and domestic animals were cited to browse on morama leaves in the field. Duiker was the most important browsers of morama while other animals were less important browsers of morama (Table 3).

	FREQUENCY %			TOTAL
	MALWELWE	MABOANE	MANTSHWABISI	(Average)
INSECTS				
Caterpillar no 1	10 (20%)	15 (30%)	13 (26%)	38 (25.3%)
Caterpillar no 2	13 (26%)	18 (36%)	10 (20%)	41 (27.3%)
Caterpillar no 3	1 (2%)	0 (0%)	2 (4%)	3 (2.0%)
Caterpillar no 4	1 (2%)	4 (8%)	3 (6%)	8 (5.3%)
Caterpillar no 5	3 (6%)	3 (6%)	4 (8%)	10 (6.7%)
Beetle no 1	3 (6%)	0 (0%)	12 (24%)	15 (10.0%)
Beetle no 2	12 (24%)	0 (0%)	0 (0%)	12 (8.0%)
Grasshopper	0 (0%)	4 (8%)	0 (0%)	4 (2.7%)
Aphids	1 (2%)	6 (12%)	6 (12%)	13 (8.7%)
Butterfly	5 (10%)	0 (0%)	0 (0%)	5 (3.3%)
Wasp	1 (2%)	0 (0%)	0 (0%)	1 (0.7%)
ANIMALS				
Duiker	15 (30%)	21 (42%)	15 (30%)	51 (34.0%)
Porcupine	9 (18%)	8 (16%)	6 (12%)	23 (15.3%)
Steenbok	6 (12%)	10 (20%)	8 (16%)	24 (16%)
Goat	6 (12%)	10 (20%)	5 (10%)	21 (14.0%)
Cow	2 (4%)	1 (2%)	3 (6%)	6 (4.0%)
Impala	0 (0%)	0 (0%)	2 (4%)	2 (1.3%)
Baboon	11 (22%)	0 (0%)	1 (2%)	12 (8.0%)
Hare	0 (0%)	0 (0%)	1 (2%)	1 (0.7%)
Ostrich	0 (0%)	0 (0%)	5 (10%)	5 (3.3%)
Rat	1 (2%)	0 (0%)	2 (4%)	3 (2.0%)
Springbok	0 (0%)	0 (0%)	2 (4%)	2 (1.3%)

Table 3: Insect species and animals identified in three villages feeding on morama

3.1.2.2 A survey on pest of mongongo in Okavango

ATTRIBUTES	NUMBER OF RESPONSES			TOTAL
	SHAKAWE	NGARANGE	NXAMASERA	(Average)
GENDER				
Male	19 (38%)	24 (48%)	18 (36%)	61 (40.7%)
Female	31 (62%)	26 (52%)	32 (64%)	89 (59.3%)
MARITAL STATUS				
Yes	8 (16%)	9 (18%)	6 (12%)	23 (15.3%)
No	42 (84%)	41 (82%)	44 (88%)	127 (84.7%)
AGE				
20-29	11 (22%)	14 (28%)	3 (6%)	28 (18.7%)
30-39	13 (26%)	14 (28%)	3 (6%)	30 (20%)
40-49	8 (16%)	3 (6%)	3 (6%)	14 (9.3%)
50-59	7 (14%)	8 (16%)	3 (6%)	18 (12%)
60>	11 (22%)	11 (22%)	38 (76%)	60 (40%)
EDUCATIONAL LEVEL				
Primary	23 (46%)	27 (54%)	42 (84%)	92 (61.3%)
Junior Secondary	19 (28%)	21 (42%)	8 (16%)	48 (32.1%)
Senior Secondary	7 (14%)	1 (2%)	0 (0%)	8 (5.3%)
Tertiary	1 (2%)	1 (2%)	0 (0%)	2 (1.3%)
EMPLOYMENT STATUS				
Employed	11 (22%)	4 (8%)	2 (4%)	17 (11.3%)
Unemployed	25 (50%)	27 (54%)	26 (52%)	78 (52%)
Farmer	14 (28%)	19 (38%)	22 (44%)	55 (36.7%)

Table 4: Farmers socio-economic background in Okavango

Table 4 presents data on socio-economic background of the respondents. 150 people were interviewed from all three villages and 59.3% of the respondents were females and 40.7% males. 40% of the respondents were old aged people above 60 years. All of the respondents said they attended school and out of this majority (61.3%) of them attended school to primary school level. It has been revealed in table 4 that 52% of the respondents from all three villages were unemployed.

Figure 7 above shows the methods of storage used by the respondents for mongongo nuts in the three village of the study. Majority of the respondents indicated that they store their mongongo in bags. 92% of the respondents in Ngarange store mongongo in the bags, compared to 84% and 70%

in Nxamasera and Shakawe, respectively. Only 46 people in all three villages indicated that there do not store mongongo in any container. They stated that they just leave it in open-space to dry and keep it away from wet or moist conditions.



Figure 7: Methods of storage of mongongo nuts



Figure 8: Responses by time about shelf-span for mongongo fruits in storage from the three villages

The responses by time about the shelf-span of mongongo fruits in storage are shown in Fig. 8. A total of 150 respondents (farmers) were interviewed in the three villages of the study. Ninety-six percent of them indicated that mongongo fruits last for years in storage and still keeping its

freshness. Few respondents (4%) indicated that the fruit last for some months before it lose its value. None of the respondents indicated the fruit can last for days. Mongongo fruits have long shelf-span according to the respondents of Okavango region (Fig. 8).



Figure 9: Percentage of respondents' knowledge of control methods used to control pests of mongongo

Figure 9 reveals the percentage of respondents knowledge of control methods used to control pests of mongongo in three villages of the study. 137 of the respondents stated that they do not use any control method in the storage, while 13 of the people use cultural methods to control storage pest. None of the respondents that use chemicals to control pest as they believe that these indigenous plants are resistant to pest attack. In Ngarange all (100%) people indicated that they do not use any control method because mongongo can natural stay for some years without infested by insect pest. Ten of the respondents in Nxamasera use cultural methods such as ash to control caterpillar A and beetle B in the storage in Table 6.

	FREQUENCY %			TOTAL
	SHAKAWE	NGARANE	NXAMASERA	(Average)
PLANT PART				
Leaves	2 (4%)	2 (4%)	0 (0%)	4 (2.7%)
Fruits	46 (92%)	44 (88%)	42 (84%)	132 (88%)
Roots	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Seeds	0 (0%)	4 (8%)	8 (16%)	12 (8%)
Flowers	2 (4%)	0 (0%)	0 (0%)	1 (0.7%)
STAGE OF PLANT				
Early stage	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Flowering	1 (2%)	0 (0%)	0 (0%)	1 (0.7%)
Fruiting	27 (54%)	39 (78%)	40 (80%)	106 (70.7%)
storage	22 (44%)	11 (22%)	10 (20%)	43 (28.7%)

Table 5: Respondents knowledge of parts and growth-stage of mongongo plant attacked by insect pests

Table 5 depicts the response of people in the three villages of the study on their knowledge of parts of mongongo plant that are mostly attacked by insect pests and also the plant growth-stages prone to attack by insects. 88% of respondents from the three villages of the study indicated that the fruits are susceptible to insects attack and the remaining 12% of the respondents stated that other plant parts are attacked by insects (Table 5). 70.7% of the respondents from all the villages of the study indicated that mongongo is attacked by insects during the fruiting stage in the field. 28.7% of the respondents indicated that mongongo has more field insect pests and few storage pests.

		FREQUENCY %		
	SHAKAWE	NGARANE	NXAMASERA	(Average)
INSECT				
Termites	4 (8%)	3 (6%)	7 (14%)	14 (9.3%)
Grasshopper	1 (2%)	0 (0%)	0 (0%)	1 (0.7%)
Beetle A	1 (2%)	0 (0%)	0 (0%)	1 (0.7%)
Beetle B	9 (18%)	3 (6%)	12 (24%)	24 (16%)
Beetle C	6 (12%)	0 (0%)	0 (0%)	6 (4%)
Caterpillar A	16 (32%)	38 (76%)	21 (42%)	75 (50%)
Caterpillar B	3 (6%)	2 (4%)	9 (18%)	14 (9.3%)
Caterpillar C	5 (10%)	4 (8%)	1 (2%)	10 (6.7%)
Caterpillar D	2 (4%)	0 (0%)	0 (0%)	2 (1.3%)
Caterpillar E	3 (6%)	0 (0%)	0 (0%)	3 (2%)

Table 6: Insect pest reported by respondents to attack mongongo in three villages

The insect pests reported by respondents to attack and cause damage on mongongo are listed on Table 6. Instead of identifying the insects by name, the respondents described their features as they did not know the names. The respondents of the three villages gave a description of pests that they had seen attacking mongongo as shown in Table 6. The most commonly cited pests of mongongo were caterpillar A, caterpillar B and beetle B (Table 6). Caterpillar A was considered the most serious pest in all three villages feeding on the fruits. The respondents indicated that there are other insect pests that attack mongongo (Table 6).

Wild animals were cited to browse on mongongo fruits in the field. Elephant is the most important (94.67%) browsers of mongongo, Duiker (54%) and goat (49.33%). Other wild animals were less important browsers of mongongo such as cow, impala, baboon and steenbok (Fig.10).



Figure 10: Identified animals that graze on mongongo fruits

3.1.3 DISCUSSION

It has been shown in this study that majority of respondents are females in all the three areas. This implies that they are ones who are mostly in the frontline involved in the harvesting and utilization of these indigenous edible plants. It has also been revealed in this study that those involved with these indigenous plants are married. Therefore, it can be stated that majority of the people who are actively involved in utilisation of these indigenous plants in all the three rural areas are married females. Ratta (1993) reported that in developing countries women in rural areas are involved in farm production and other activities that can feed their families while men are managing income generating activities. In this regard it can be said that the utilisation of these plants is mainly a family affair. In view of domesticating these plants, women will play a pivotal role. This can be supported by Augustino and Gillah (2005), who argued that domestication is an adaptive strategy for women especially in rural areas due to their marginal economic status and their special interest in plants. In rural communities, women are for the wellbeing of their families. This, therefore, infers the influence of gender on domestication of indigenous plants. It should be noted that

majority of the respondents were unemployed meaning that they supported their families with what they could do for themselves including harvesting and utilising these plants. In addition, the jobs in rural areas are limited. All the three areas of our study are rural lacking economic opportunities. The income per capita in rural areas is relatively low and opportunities for professional development are very limited and in some cases non-existent. Majority of people depend on agriculture sector to support their livelihoods.

It has also been shown from the findings of this study that 61% of the respondents had at-least primary school education and close to a quarter had both junior and senior secondary education with a few having tertiary education. Assuming that primary education is good to inform the decisions one can take, it can therefore, be stated that the respondents interviewed had some form of formal education. Education level is believed to play an important role in decision making to harvest and utilize edible indigenous plants. Those with primary and secondary school education are believed to be educated enough to make well informed decisions and also know the benefits of indigenous plants hence their utilization. A similar observation was reported in a study that was conducted in Nigeria by Oladele (2011) who reported that those who were educated were able to produce and consume indigenous vegetables. This is attributed to the knowledge on aspects of agronomy and nutritive value.

The findings of this study depicts that most respondents for morama study aged between 20-39 years. This indicates that they are still in the productive age group and these are expected to actively participate in the livelihood activities such as harvesting and utilization of edible indigenous plants. Their interest and involvement in these plants can be manipulated so that they can actively assist and take part in the domestication process of these plants in their areas. This has

similarly been seen in a study that was conducted in Gweta (Botswana) by Badimo *et al*, (2015) and Onim and Mwaniki (2008), who indicated that those at young age participate in farming activities and utilisation of plants for their livelihoods. Age is one of the factors which can affect and influence participation in harvesting and utilisation of edible indigenous plants. This is because age affects the efficiency of carrying out activities (Nyaruwata, 2019). Young people can make informed decisions because they are networked. In contrary, a study that was done in Cameroon (Godswill *et al.*, 2014) reported that younger people are influenced by modernization and globalisation. Therefore, there is likelihood for them to shun the harvesting and utilisation of indigenous plants and have preference for exotic plants. In light of this, in most rural communities, the indigenous plants are mostly consumed by older people than younger ones.

Majority of the respondents indicated that morama and mongongo nuts have long storage life span. This finding is in agreement with Van der Maeson (2006) who stated that raw mature morama seeds store well and remain edible for years under dry storage conditions. Ronne and Joker (2006) reported that mongongo seeds remain viable for up to 2 years if stored at 10°C. Both the respondents from Kweneng region and Ngamiland region interviewed in this study indicated that they store mongongo and morama nuts in open weaved and polypropylene bags for future use. Similar method of storage was reported by Hodges and Stathers (2015) who stated that polypropylene bags are used for grain and pulse storage by more than 80% of smallholder farmers in Malawi, Tanzania, Kenya and Zimbabwe. At a small scale, grain and pulse are stored traditionally in different styles of containers, depending on the farmer's socio-economic status and his environment (Sallam, 2013). Structures used traditionally are often inexpensive and environment motivated. Hodges and Stathers (2015) reported that polypropylene bags are not as

very expensive by rural households, and are typically re-used a couple of times. Still in the same regions few respondents indicated that they do not store in any containers.

The nuts are left to dry in an open space by solar radiation. Respondents also stated that humid and warm temperatures have been reported as the principal conditions that favour establishment of pests (Landston and Eaker, 2009). Majority of the respondents from both regions indicated that in this study, they do not use any control methods to protect their products from pest. They further explained that the reason for not controlling the pests is that mongongo and morama are wild crops which are resistant to pest and diseases.

In this survey respondents indicated that there are some insect pests that are major constraint to the production of mongongo and morama. Their level of education had an effect in identifying the insects by names. Respondents in all areas generally gave description of the insect (pest) to Order level. Descriptions given also included general local names for the insects which enabled the study to capture them into orders. Two insect orders namely, Coleopteran (Dikhukhwane) and Hymenoptera (Mehu), were highly ranked to cause damage to the fruits of mongongo and morama. White larvae (Diboko) which could be either Lepidoptera or Coleoptera from the description were identified as a major constraint for both mongongo and morama fruits followed by brown beetles. This identification by Respondents is consistent with the report of Agro-forestry tree data base, (2006) that fallen fruits of mongongo are attacked by unspecified moth and larvae which eventually eat all the fleshy parts. Jackson *et al.* (2010) reported that insect pests have been observed causing seed damage of morama but the identification of these insects was not confirmed. It has been reported that the level of resistance in most of the varieties released for cultivation was moderate while high levels of resistance have been reported in their wild relatives (Sharma *et al.*, *e*

2003). This can imply that the reason why respondents did not mention or observe many insects on these plants could be the factor of resistance of these plants to insects.

In this study the majority of the respondents stated the names of vertebrate pests which are major constraints to mongongo and morama production. Respondents showed that both wild and domesticated animal feed on mongongo and morama. Elephant was highly ranked as the major pest for mongongo. Other wild animals such as kudu, Duiker, monkey and domesticated animals such as a cow and goats were reported to feed on the fruits of mongongo by the respondents in Okavango region. The findings of Graz, (2002) report that Loxodonta africana (Elephant), Hystrix africaeustralis (Porcupine) and Tragelaphus strepsiceros (Kudu) feast on the sweet fruits of mongongo and produce a clean nut. Chimbelu (1983) reported that Elephant and Kudu are involved in the seed dispersal through the discarded pits. He further elucidated that livestock chew the soft part of the fruit and discard the stone. Mongongo fruit pulp and the seed meal which are rich in protein can be fed to cattle, however, the feed is suspected to cause a discolouration of beef (Agro-forestry tree database, 2006). National Academy of Sciences (1979) reported that grazing animals especially cattle feed on morama stems and leaves, limiting the occurrence of the plant around the waterholes. The foliage of the morama plant serves as forage for livestock and wildlife in southern Africa because the leaves are highly palatable (Dakora, 2013).

3.1.4 Conclusion

Based on the findings of this study, it can be concluded that farmers had seen some insects attacking both morama and mongongo plants. Therefore, it can be concluded that they knew the pests of these two plants. However, they were not able to identify the insects by name but could only describe their features. The survey revealed that both the plants under the study are attacked

by insect pests at field and in the storage. The results of the survey are consistent with the alternative hypothesis that insect pests attack indigenous plant species (mongongo and morama). Wild animals and domesticated animals they browse on morama and mongongo which may affect their production. Wild animal such as elephant which are the main pest of mongongo may cause serious damage on the domestication of mongongo plants since the elephant are difficult to control.

3.2 Experiment 2: Diversity of insect pests and natural enemies of wild morama and mongongo plants in the Kweneng and Okavango regions.

3.2.1 Materials and methods

3.2.1.1 Sampling sites

Locations within Botswana, where Mongongo and the Morama bean plant grows in natural abundance, were chosen as sampling sites for this study of the field insect pest of Morama and Mongongo. Two villages were selected in Kweneng district for morama study namely: Malwelwe (S23.99166°; E023.20027°), Maboane (S24.09500°; E024.55607°). Mongongo areas which were selected for the study included Shakawe (Shaikarawe and Ukhusi) in the Northern part of the country (S18.30377°; E021.73665°) and BUAN garden (S24.5914°; E025.9415°) in Gaborone. For this study, the research areas are different from those of experiment one because during the survey it was revealed that people in Nxamasere and Ngarange (For experiment 1) collect mongongo from other Shakawe surrounding areas such as Ukhusi and Shaikarawe where it is present in abundance. Therefore, this is why experiment 2 was done in Ukhusi and Shaikarawe. It should be noted that Nxamasere, Ngarange, Ukhusi and Shaikarawe settlements are all close to each other and surround Shakawe.

3.2.1.2 Experimental procedure

Sampling was done during rainy season (October - May) for the insects that attack the morama and mongongo vegetation stages. Therefore, field visit to the areas of study, where mongongo and the morama bean plant grow in natural abundance, was done during the SASSCAL #335 trips as this was part of this project. Three field trips were taken to those areas. The first trip was done

immediately after the trees started to shoot in spring for both morama and mongongo from October to December. The second trip was from January to February for morama bean only when the plants had fruits or pods. The last trip was done from April to May for mongongo when plants started to fruit and also when fruits dropped off. The insects were captured live from the trees. Sweep nets and yellow sticky traps were used to catch flying insects from the plants and crawling insects were handpicked using protective gloves. Three to five same insects were collected per unit area where possible and the containers were labelled by writing the date, location, and name. The activities of insects such as during feeding on the plants were captured using a digital camera by taking photos. Insects are easy to identify when they are adults, therefore where insects captured are immature stages such as larvae, they were reared in rearing bottles under laboratory conditions to allow the insect to complete its life cycle to adult stage.

The collected insects were killed using ethyl acetate and put in a container with a relaxer to avoid drying. Some of the insects sample especially the ones that are very small in size (Aphids and Whiteflies) and those at larval stage were stored in a container with 95% alcohol and put in -80° C freezer for preservation. Moths can easily lose identification features like body colour and scales were left in collection relaxing containers for later identification.

3.2.1.3 Insect identification preparation

The morphological identification of insect samples was done. The insect samples preserved in alcohol were prepared for morphological identification by removing them from alcohol and air drying them by placing them on the paper towel under laboratory conditions for two minutes. Those in collection and storage containers were also removed and prepared for the identification process. Large insects were pinned while the small specimens were mounted by double mounting method. Pinning of insects was done by removing specimens from the relaxing container, to ensure

the appendages moved freely and were spread to the desired shape and posture. Size 2 and 3 pins were used to pin large specimens and standard methods of pinning were applied according to types of insects.

Stink bugs and other large Hemiptera were pinned through the scutellum to the right of the middle line. Wasps (parasitoids) were pinned through the thorax slightly behind the bases of the forewings and to the right of the middle line. Beetles were pinned through the right wing cover near the base. Moths were pinned through the middle line of the thorax at the thickest point, between and slightly behind the bases of the forewings. The small specimens were double mounted using a 0.0001 pin, which passing through the thorax to the paper on a pin. This was done under 2× magnifying lens using forceps to pin or glued to paper point on a pin. After mounting of the insect specimens, morphological identification was done using identification keys under the dissection microscope. Insect identification was done to family level and then specimens were sent to Pretoria at ARC Biosystemic for further identification to species level. After identification to species level at the end of the study the specimens were stored and added to BUAN entomology collection.

3.2.1.4 Data collection

The data on insect species composition on both morama and mongongo in the field were recorded. However, the total number of insects present in the samples was not collected consequently detailed analysis of the abundance of each species was not done.

3.2.2 RESULTS

Table 7 below depicts the diversity of insects which were collected from mongongo plants from outskirts of Shakawe village (Ukhusi and Shaikarawe) and BUAN. Insects for morama were collected from BUAN and Malwelwe village. Insect diversity data showed different individuals from five insect Orders of Coleoptera, Isoptera, Hemiptera, Diptera and Lepidoptera were collected from two different host plants morama and mongongo (Table 7). These insects were found feeding on the host plant during its vegetative stage in the forest. The number of insect Orders, Families, Superfamilies, Genus and Species of each host plants morama were presented in Table 7 below.

ORDER	FAMILY	SUPERFAMILY	GENUS	SPECIES	HOST	LOCATION
Coleoptera	Tenebrionidae	Coelemetopinae	Alobates sp		Mongongo	Ukhusi
Coleoptera	Nitidulidae	Carpophilinae	Carpophilus	hemipterus	Mongongo	Ukhusi
Coleoptera	Silvanidae	Silvaninae	Silvanus sp		Mongongo	Shaikarawe
Diptera	Agromyzidae		_		Mongongo	BUAN
Hemiptera	Pentatomidae		Dorycoris sp		Mongongo	Ukhusi
Hemiptera	Pentatomidae		Pseudatelus	natalensis	Mongongo	BUAN
Hemiptera	Pentatomidae		Menida sp		Mongongo	BUAN
Hemiptera	Psyllidae				Mongongo	Ukhusi
Hemiptera	Alydidae		Tupalus	faciatus	Mongongo	BUAN
Hemiptera	Lygaeidae				Mongongo	Shaikarawe
Hemiptera	Pseudococcidae		Paracoccus sp		Mongongo	BUAN
Lepidoptera					Mongongo	Shaikarawe
Lepidoptera					Mongongo	Shaikarawe
Lepidoptera					Mongongo	Ukhusi
Coleoptera	Chrysomelidae	Galerucinae	Afropachylepta	nigrotibialis	Morama	Malwelwe
Hemiptera	Cicadellidae		Afrosteles sp		Morama	Malwelwe
Hemiptera	Scutelleridae		Callidea	dregii	Morama	BUAN
Hemiptera	Aleyrodidae		Bemisia	tabaci	Morama	BUAN
Hemiptera	Aphididae		Aphis	gossypii	Morama	BUAN
Isoptera	Termitidae	Macrotermitinae			Morama	BUAN
Lepidoptera					Morama	Malwelwe

Table 7: Summary of field insect pests of mongongo and morama

Lepidoptera were collected and were not identified because were unknown.



Figure 11: Field pest of morama *Afropachylepta nigrotibialis* (A), Unknown moth (B), *Afrosteles sp* (D) and *Macrotermitinae* (E).

Figure 11 shows pictures of field insect pests of morama mentioned in Table 7 which were identified to feed on morama leaves. The other three herbivores *Calidea dreggi, Bemisia tabaci* and *Aphis gossypii*, their pictures were not captured because they were stored in alcohol. White flies and aphids were found sucking the sap under the leaves surface of morama. *Calidea dreggi* was found on the flowers of morama. *Afrosteles sp* were found feeding under the leaf surface by sucking the sap and cause the leaves to dry and die. *Macrotermitinae sp* were found attacking morama plant parts above the ground, causing the leaves and vines to die off. Unknown moth (B) was found feed under the leaf surface and also boring holes on fresh pods and seeds of morama. The larvae was described by the respondents during the survey study in Malwelwe and Maboane villages, as the pest that cause damage to the fresh pods and seeds of morama in the field. Few numbers of the larvae of unknown moth B were observed during collection (Fig. 11).



Figure 12: Field pest of mongongo; Unknown moth (A), Unknown moth (B), Unknown moth (C), *Carpophilus hemipterus* (E), *Alobates sp* (F), *Silvanus sp* (G), *Psyllidae sp* (H), *Agromyzidae sp* (I) *Monomorium sp* (J), *Dorycoris sp* (K), *Paracoccus sp* (L), *Tupalus faciatus* (M), Unknown beetle (N) and Unknown wasp (O).

Figure 12 above present the pictures of identified and unidentified field insect pests hosted by mongongo plant in the forest that were presented in Table 7. A high incidence of larvae was recorded feeding on the fruits than on the leaves of mongongo. Three species of unknown moths were found feeding on the flesh of mongongo fruits as larvae or during their larval stage. Unknown moth A was found its larvae feeding on fruit flesh of mongongo. The larvae of moth A feed and pupate inside the fruit, and emerge out of the fruit as an adult moth. Massive numbers of larvae and moths A were observed during collection (Fig. 12). Unknown moth B larvae also feed on the fruit flesh of mongongo, but when it comes time for the larvae to pupate, it emerges out of the fruit. The larvae dug the soil and pupate under the soil. The larvae of moth C (Fig. 12) it also feed on the flesh of the fruit and pupates inside the fruit. Both moth B and moth C were not found in massive number during collection. Beetle species (E, F, G and N) on Figure 12 are the herbivores

feeding on fresh mongongo fruits on the tree and dropped fruits on the ground. *Carpophilus hemipterus* was found feeding on dried mongongo fruits that has fallen down on the ground. Massive numbers of *C. hemipterus* were observed during collection. Beetle F was found feeding on mongongo decomposed fruits on the ground. According to the observation during collection beetle G and N were not in massive numbers during collection at the field. Insect L and H (Fig. 12) were found sucking the sap under mongongo leave surface and cause the leaves to wilt and dry. Other insect were found on the leaves and fruits of mongongo.

3.2.3 DISCUSSION

Different species of insect pests which were collected from mongongo and morama plants in the wild were sent for identification at Agricultural Research Council (ARC) in South Africa. No literature has documented the pest of mongongo and morama. This document is the first to identify the insects that are hosted by morama and mongongo.

3.2.3.1 Morama field insect pests

Four Orders of insects were identified to be hosted by morama plant at the wild. These Orders were Coleoptera, Hemiptera, Isoptera and Lepidoptera. The identified insect species were the herbivores *Afropachylepta nigrotibialis, Afrosteles sp, Callidea dreggi, Bemisia tabaci, Aphis gossypii, macrotermitinae* and unknown moth. These were found feeding on morama leaves. *Callidea dreggi* was found feeding on the flowers of morama plants. In Ghana, Kaufmann (1966) reported that oviposition by *Callidea dreggi* occurs throughout the year, on the flower parts and uncommonly on the stems of its host plant *Jatropha podagrica hooker* (Euphorbiaceae). Giliomee, (1997) reported during 1995 and 1996 the iridescent stink bug *Callidea dreggi* (Hemiptera:

Scutelleridae) was found at Western cape Provence in such large numbers that they caused damage to garden plants. According to Enlitz *et al.* (1989) it is widely distributed in the eastern parts of Africa, Madagascar and Arabia where it is a pest of various crops, including sunflower and cotton. *Callidea dreggi* can be a serious pest for morama production if it is not monitored and increase in large population. Females mate immediately after emergence as adults and the first batch of eggs is usually laid about 10 days after the first mating (Kaufmann, 1966).

This study discovered that *Bemisia tabaci* was hosted by morama plant in greenhouse at BUAN. The new germinated morama seedlings and fresh leaves or shoots were observed to be susceptible whitefly attack. Similar observation were reported by Oliveira and Silva, (1997) that the whitefly is a very adaptive insect with a high oviposition rate, laying its eggs in the abaxial surface of young leaves. After hatching the first instars move along the leaf until they find a favourable site for their development, where they insert their stylet into phloem and remain sessile until the emergence of the adult (Byrne and Bellows, 1991). Cock, (1993) indicated that *Bemisia tabaci* collectively colonizes over several hundred plant species. They have a high potential to cause economic damage in tropical and subtropical regions due to the direct and indirect injuries inflicted on numerous crops (Vieira *et al.*, 2012). Direct damage is characterised by sap sucking and injection of toxins into the plant, whereas indirect damage includes virus transmission and excretion of honeydew, which serves as a substrate for the growth of sooty mold fungi (Suekane *et al.*, 2013). Since 1995, the losses caused by *B. tabaci* have increased in the crops of beans, cotton, melon and vegetables totalling in amount of 3 billion USD (Czepak, 2010).

Aphis gossypii was also observed feeding on morama seedling in the greenhouse at BUAN. The affected leaves of morama showed some excretion of honeydew on the surface. According Zamani *et al.* (2012) stated that *Aphis gossypii* infest important crops in Iran including greenhouse plants.
Alizadeh *et al.* (2016) described *Aphis gossypii* as small, soft-bodied insects feeds on the underside of leaves sucking out plant sap. This species has a very high rate of development and is able to increase up to 12 times per week (Roistacher *et al.*, 1984). Henneberry and Forlow, (2001) reported that high population of *Aphis gossypii* can reduce the vigour of the plant, making it susceptible to other pests. The previous studies indicate that *Aphis gossypii* will be a problem pest of morama if is not controlled due to its high rate of development. The honeydew that aphids excrete will reduce the quality of morama pods because of the development of a black sooty mold on the substrate. Rondon *et al.* (2005) reported that sooty mold reduces photosynthate production and reduces the quality of the plant causing considerable economic injury.

3.2.3.2 Mongongo field insect pests

Five insect Orders were identified to be hosted by mongongo plants in the wild. The identified insect species were the herbivores *Carpophilus hemipterus, Silvanus sp, Dorycoris sp, Tupalus faciatus, Pseudutelus natalensis, Menita sp, Paracoccus sp* and *unknown moths*. This study has revealed that mongongo is susceptible to insect pest that feed on the fruits. It has been reported that the exocarp is often eaten by insects (Peters, 1987). The identified insect pests that are hosted by mongongo plant only attack the outer layer of the fruit, failing to penetrate the thick hard shell (kernels) containing the nut. Similar observation were reported by Graz, (2007) that fallen fruits are susceptible to attack by unspecified moth larvae which eventually eat all the fleshy parts. Mongongo has a brown fruits when ripen, containing a thin layer of edible flesh around thick hard shell and inside the shell is a highly nutritious nut used to extract oil.

Carpophilus hemipterus is one of the insect pests hosted by mongongo plants in the wild. Many *Carpophilus hemipterus* beetles were found feeding on the flesh of mongongo fruits and causing the fruits to rot. The rotten fruit were covered with white powdery substance. Klein *et al.* (2007)

described *Carpophilus hemipterus* as a cosmopolitan species that is frequently found in stored products and rotting fruit. Yeasts growing on rotting fruit form an important part of its diet and some of these yeast species have been characterised (Miller and Mrak, 1953). In Hawaii these beetles breed in large numbers in knocked down pineapple fields where they feed upon the rotting stem (Gerling and Mordechai, 1981). They further stated that once their food source has been exhausted, the beetles migrate to other places looking for new food sources. Previous studies about *C. hemipterus* indicated that the beetle can be destructive to mongongo fruits if it is not monitored, because the beetles are always migrating at large swarms that feed on the flesh of mongongo fruits.

3.2.4 Conclusion

The findings of this study revealed that both mongongo and morama in the wild is attacked by insect pests especially during fruiting period. The findings agree with alternative hypothesis that insect pests affect indigenous plant species (mongongo and morama). This study reveals that majority of identified insect pests of morama and mongongo were found to target mostly the fruits and pods. If insect pests are not monitored or controlled they may cause problem for domestication of indigenous species.

3.3 Experiment 3: Diversity of storage pests and natural enemies of morama and mongongo nuts in storage

3.3.1 Materials and Methods

3.3.1.1 Study area

The experiment was conducted at the University of Agriculture and Natural Resources (BUAN), Sebele, Entomology laboratory. The University lies on latitude 24°33'S and longitude 25°54'E elevated at 993m above sea level. The climate of the area is semi-arid with an average rainfall of 538mm.

3.3.1.2 Source of test material

Morama and mongongo seeds and fruits that were used in this study were collected from 2014 to 2016, through Southern African Science Service Center for Climate Change and Adaptive Land Management (SASSCAL) # 335 project. SASSCAL # 335 was a project researching on cultivation, value addition and marketing of climate smart emerging crops to improve food security in Botswana. The overall objective of the project was to cultivate, add value and market selected indigenous species (*Tylosema esculentum, Citrullus lanatus, Schinziophyton rautenenii* and *Bauhinia petersiana*), in order to contribute towards enhancing food security and poverty eradication in Botswana. The seeds were stored in Food Science and Technology laboratory storeroom in BUAN. These seeds and fruits were obtained where mongongo and the morama bean plants naturally grow in abundance. The mongongo seeds and fruits were collected from Shaikarawe areas in northern part of the country (S18.30377°; E021.73665°) whilst morama seeds were collected from both Maboane (S24.09500°; E024.55607°) and Malwelwe (S23.99166°;

E023.20027°) villages in Kweneng district. For this study, seeds were not separated by location because of mix-up of bags in storage.

3.3.1.3 Experimental procedure

This experiment was done to determine if seeds of morama and mongongo attract and get attacked (susceptible) by insect pests. It should be noted that the source of infestation could be from where the seeds were collected or where the seeds are kept. But since the interest of the study is to determine their susceptibility to insect pests attack and also determining which insects attack them, the source of infestation was assumed insignificant. For this experiment, 200g of seeds were weighed using Top pan balance scale and these seeds were put in maize meal sacks and kept under laboratory conditions on top of the laboratory bench for six months to allow any insect activity to happen. This was replicated 4 times. Any insects observed on the seeds were then collected and kept for identification. The insects collected were killed using ethyl acetate and then preserved by keeping them in 95% alcohol in small vials while awaiting identification. This also included the immature stage (larvae and the nymphs). Only the Lepidoptera adults were preserved by pin mounting them to preserve scales which can assist in the identification.

3.3.1.4 Data collection

The different types of insects that developed from the seeds and fruits were collected weekly until the end of the experiment. The insects found were observed and counted from all the replicated sacks. Before opening and counting the insects found in each sack, the sacks were put in a freezer at -20c for 30 minutes to anaesthetically treat the insect to avoid flying insects to escape. The insects were counted under a magnified double lens on the laboratory benches.

3.3.1.5 Insect identification preparations

The insects collected were mounted using different methods depending on the type of the insect. Big insects were mounted on pins. A spreading board was used to spread the wings and legs of insects into the desired position, and pin the specimen on the board. Card point mounting was done for very small insect specimens by gluing them to small cardboard points mounted on pins. After mounting of the insect specimens, morphological identification was done using identification keys under the dissecting microscope. Insect identification was done to family level in the laboratory. Afterwards the specimens were again prepared for verification process. The identified specimens were given codes and put inside packaging box and taken to Dalsey Hillblom Lynn (DHL) couriers. The specimens were sent to Agricultural Research Council (ARC) in South Africa for Biosystematics verification and further identification to species level.

3.3.1.6 Data analysis

The pattern and structure of data on species diversity and composition was detected using multivariate analysis. Data on insects were analysed as repeated measure designs using a repeated statement in a mixed model procedure (PROC MIXED) (SAS Institute, 2004). However, the total number of insects present in the samples was not collected consequently detailed analysis of the abundance of each species was not done.

3.3.2 RESULTS

		Super			Host
Order	Family	family	Genus	Species	plant
Coleoptera	Ptinidae	Xyletininae	Lasioderma	serricorne	Mongongo
Coleoptera	Tenebrionidae	Tenebrioninae	Tribolium	confusum	Morama
Lepidoptera	Pyralidae		Plodia	interpuctella	Morama
Lepidoptera	Pyralidae		Plodia	interpuctella	Mongongo

Table 8: Summary of identified storage insect pests of mongongo and morama

Table 8 shows the insect pests observed on morama and mongongo in storage. Insect in two orders were identified attacking both mongongo and morama nut in storage. The insects were found to belong to Orders Coleoptera and Lepidoptera. Coleopteran family, Ptinidae was found to attack mongongo whilst family Tenebrionidae was recorded attacking morama. (Coleoptera: Ptinidae), *Lasioderma serricorne* were hosted by mongongo nuts, while (Coleoptera: Tenebrionidae), *Tribolium confusum* was only hosted by morama nuts. *Plodia interpunctella* was found to attack both mongongo and morama nuts in massive numbers.



Figure 13: *Tribolium confusum* (A), *T. confusum* feeding on deshelled morama (B), *Plodia interpunctella* (C) and *P. interpunctella* larvae feeding and making webs (D).

Figure 13 shows the pictures of two storage pest of morama and the damage they cause to morama nuts in storage. Both the larva and adult beetle of *Tribolium confusum* feed on morama nuts by causing channels inside the seed (B). However, the only the larvae of *Plodia interpunctella* moth

feed on the seed of morama and crash them in to powdery form (D). A numerous number of *T*. *confusum* and *P. interpunctella* were observed feeding on morama seeds in storage.



Figure 14: *Plodia interpunctella* adult (A), *P. interpunctella* larvae making a web (B), *L. serricorne* larvae pupating (C) and *Lasioderma serricorne* adult (D).

Figure 14 shows pictures of identified storage pests of mongongo presented in Table 8. Two species *Lasioderma serricorne* and *Plodia interpunctella* were identified feeding on the pulp of mongongo dried fruits in storage. Both the species were observed to eat the fruit flesh between the hard nut and the out skin.

Table 9: Summary of identified natural enemies of pest of mongongo and morama

Order	Family	Super family	Genus	Species	Host plant
Hymenoptera	Braconidae:	Braconinae	Bracon	Hebetor	Morama
Hymenoptera	Chalcididae:	Haltichellinae	Psilochalsis sp		Morama
Hymenoptera	Chalcididae	Haltichellinae	Antrocephalus sp.		Morama
Hymenoptera	Formicidae	Myrmicinae	Monomorium sp		Morama
Hymenoptera	Bethylidae				Morama

Table 9 depicts the natural enemies observed on pests of mongongo and morama in storage. The natural enemies are all in insect Order Hymenoptera and these were observed only on morama. In this Order Hymenoptera four different families were recorded namely; Braconidae, Chalcididae, Formicidae and Bethylidae. A parasitoid *Bracon hebetor* (Braconidae) was seen laying eggs on the larvae of the *Plodia interpunctella*, a storage pest of morama nuts. *Psilochalsis sp*

(Chalcididae) was found attacking the larvae of unknown moth that feeds on the leaves of morama. A parasitoids *Antrocephalus sp* and Ants *Monomorium sp* were found in a storage bag with morama seeds infested with *P. interpunctella*.



Figure 15: Bethylidae sp (A), Psilochalsis sp (B) Bracon hebetor (C), and Antrocephalus sp (D).

Figure 15 shows pictures of natural enemies that are presented in Table 9 which are parasitoids of pests attacking morama. Parasitoids A and B (Fig. 15) were found around unknown moth (B) larvae presented in Figure 11 that was feeding on morama leaves at the forest. Few numbers of unknown moth (B) larvae were observed feeding on morama leaves and I assumed parasitoid A and B (Fig. 15) managed to control the population growth of the unknown moth B larvae (Fig. 11). Parasitoid C and D (Fig. 15) were found attacking *P. interpunctella* that feed on morama nuts in storage. *B. hebetor* paralysis the larva of the *P. interpunctella* moth and deposits its eggs inside the paralysed moth. Parasitoid *Antrocephalus sp* were found bag of stored morama seeds which were attacked by *P. interpunctella* larvae. Few numbers of *P. interpunctella* larvae and moths were observed inside the storage bag of morama. I assumed the population growth of *P. interpunctella* was controlled by the parasitoid *Antrocephalus sp* to reduce their numbers compared to other bags where there was no parasitoids found in the storage bags.



Figure 16: *Bracon hebetor* (A), *B. hebetor* depositing eggs in paralysed *P. interpunctella* larvae (B), Unknown parasitoid (C), Parasitoid C larvae pupates outside unknown mongongo moth larvae (D), Unknown parasitoid (E), Unknown parasite (F) and unknown fruit parasitoid (G).

Figure 16 above present the pictures of natural enemies of insect pests that attack mongongo in the wild and storage. Fig. 16, unknown parasitoid C, was observed attacking the larvae of unknown moths B (Fig. 12) and deposit eggs inside the larvae. Parasitoid C has demonstrated different behaviour compared to *B. hebetor*. The parasitoid C does not paralyse the host larvae (Fig. 16, D which is the adult moth B, Fig. 12) and deposit eggs like *B. hebetor* does. It was observed that the parasitoid C deposit eggs and they survive inside the host without killing the larvae. The parasitoid larvae is the one that kills the host larvae when it is about to pupate and the late instar parasitoid larvae will later come outside and pupate. The population number of host larva D (Fig. 16) and adult moth B (Fig. 12) according to the observation when collecting samples only few larvae were found. I assume parasitoid C (Fig. 16) has influence on the low number of larva D that where observed. Parasitoids E and G were found pupating inside mongongo fruits. Their adults (wasps) were observed coming out of the fruits. We assume that they parasitize their host inside the fruit. Parasite F was found around mongongo fruits infested with moth larvae A (Fig. 12).

3.3.3 DISCUSSION

Insects that infest stored products of morama and mongongo were identified in the order Coleoptera and Lepidoptera. Two species of beetles and specie of moths were causing destruction to the stored products of mongongo and morama. This study was able to identify storage pests of morama. The most abundant insect species were the herbivores, *Tribolium confusum* and *Plodia interpunctella*. The larvae of the moth *Plodia interpunctella* were observed to feed on de-shelled morama seeds and spin the web as they feed. This is supported by what was reported by Mohandass *et al.* (2007) who stated that the larvae continuously spin a silken web both inside and on top of food surface and feed within the web. The observation from this study was that the infested seeds of morama were producing unpleasant smell covered with a dust web (Fig.13 D). Phillips *et al.* (2000) explain that the webbing contains larval excreta (frass) and exuvia (cast skins), and gives unpleasant odour to the infested commodity. He further stated that the commodity is sometimes covered on the surface with a thick mat of silken webbing causing direct product loss and indirect economic costs through pest control costs, quality losses, and consumer complaints.

Scientific studies undertaken by Hamlin *et al.* (1931) categorised *P. interpunctella* as a pest of grain, fruits, nuts and Candice from agricultural system in California. Phillips and Strand, (1994) found that adult *P. interpunctella* oriented towards food odours and laid more eggs on substrates containing food than on those without food. Silhacek *et al.* (2003) stated that the adults may lay their eggs near the food surface when the food is inaccessible due to packaging or other barriers, or when food odours are weak. Observations on this study were that *P. interpunctella* adults and larvae were found in de-shelled morama nuts than in shelled morama storage bags. Arthur, (1994) found that one stored oil seed crop that is particularly vulnerable to *P. interpunctella* is stored in-

shell peanuts. Arthur, (1995) that the larvae of *P. interpunctella* will not penetrate a solid pod, but enter through a crack or split in the shell, and then feed on the kernel.

Tribolium confusum beetles and larvae were present in large numbers infesting de-shelled seeds of morama causing the seed to turn in powdery form as they feed. This is in agreement to what was reported by Baldwin and Fasulo, (2003) that the beetles infest grains in large numbers but are unable to attack sound or undamaged grains. It spends its entire life in various pulverized grain such as flour. T. confusum was found in the same storage bags with P. interpunctella feeding on morama nuts during insects collection on this study. Pires et al. (2017) stated that this beetle is categorized as a secondary pest because the adult and immature form feed on pre-cracked or broken grains, which were damaged by primary pests. However, reports in the literature describe the ability of this insect to survive even in the undamaged grains (White, 1982). T. confusum beetles were found causing channels inside de-shelled morama seeds (Fig. 13 B). Pire et al. (2017) described adult and immature T. confusum as builder of galleries which may evolve into larger lesions. The attack of T. confusum caused evident loss of dry matters resulting in loses of bulk, mass, concurring with problems associated with pest attack in other stored grains (Caneppele et al., 2003). Losses caused by T. confusum to stored products have been assessed in Botswana by other scientist. Mohale et al. (2010) reported that increased weight loss and damage caused to groundnuts were related to the T. confosum adult and larvae populations. The observation by Mohale et al. (2010) on correlation between T. confusum numbers and increased weight loss in the groundnuts samples agrees with other observations done by Allotey and Goswami, (1994).

This study was also able to record the identified storage pests of mongongo. Two species *Lasioderma serricorne* and *Plodia interpunctella* were identified feeding on the pulp of mongongo

dried fruits in storage. Both the species were observed to eat the fruit flesh between the hard nut and the out skin. Poderoso *et al.* (2013) describe *L. serricorne* as the important pest of stored products such as dried fruit, nuts, cereals and tobacco. The cocoons, larvae and beetles were found inside the mongongo fruits after feeding on the fruit flesh. This shows that this insect might be completing its developmental stages in the fruit. The adult beetles make some holes on the way out of the fruit. This finding is consistent with Cabrera, (2001) report that mature larvae create small cocoon with particles of the substrate and pupate within it. Adult beetles and grubs bore through and leaving holes. Saglam *et al.* (2015) stated that adult beetles live only 1-2 weeks, during which time they mate, locate suitable larval food, lay eggs and then die. The larvae cause damage to products by consuming and degrading the quality of the product to a point that there is an economic loss (CORESTA, 2013).

P. interpunctella larvae were feasting on the flesh of mongongo fruits. The mature larvae make holes and leave the fruit to pupate outside the fruit by making a silk web. The same characteristics were reported by Phillips *et al.* (2000) who stated that the larvae continuously spin a silken web both inside and on top of the food surface and feed within the web. Natural enemies of pests attacking both mongongo and morama were also identified in this study. Few predators were identified attacking the pest hosted by morama and mongongo plants. For morama seed in storage parasitoid *Bracon hebetor* was identified to be hosted by *Plodia interpunctella* in storage. The parasitoid *B. hebetor* was paralysing the larvae and deposit eggs direct inside the larvae of the moth *P. interpunctella*. The eggs hatch inside dead larvae and the mature larvae of the *B. hebetor* will come out of the dead moth larvae and pupate outside the host. This characteristic is consistent with the report of Antolin *et al.* (1995) who stated that *B. hebetor* females first paralyze their host larvae by stinging and then laying various numbers of eggs singly in the paralyzed host. Once a

host is located the female *B. hebetor* inject venom that induces complete paralysis of the host (Hagstrum and Smittle, 1978). The venom blocks neuromuscular transmission at a pre-synaptic site (Petters and Stefunelli, 1983). Paralyzed host larvae are then used as food source for developing wasp and adult females. *B. hebetor* is primarily known as a parasitoid of pyralid moth larvae that are associated with durable stored food products, and include the *P. interpunctella, Ephestica kuehniella, Ephestia elutella, Ephestia cautella* and *Vitula edmansae* (Brower *et al.,* 1996). Ahmed, (2012) stated that *B. hebetor* is currently used potential biological control agent against *pyralid* and stored grain products. Drenth, (1969) reported that only Lepidoptera species are susceptible to its venom. For instance it is used on *Plodia interpuctella* in USA (Brower and Press, 1990).

An *Antrocephalus sp* was found in a storage bag of morama infested with *P. interpunctella*. Literature shows that some chalcidids of genus *Antrocephalus* are natural enemies of stored product moth pests (Konoshi *et al.*, 2004). Parasitoid *Psilochalis sp* were found around unknown moth B (Fig. 11) larvae that were feeding on morama leaves. Obopile and Mosinke, (2003) recorded *Psilochalis soudanensis* parasitoid attacking stemborer *Chilo partellus* at Chadibe and Parakarungu in Botswana. The success of a parasitoid as an efficient bio-control agent depends on its fitness (survival, fecundity, development duration and sex ratio) over generations. The intrinsic rate of natural increase is a measure of the biotic potential of the species and the advantage of using this measure is that it integrates the effect of the fertility factors into a single value (Saini *et al.*, 2019).

3.3.4 CONCLUSION

Based on the findings of this study, it can be concluded that mongongo is a major host for *P*. *interpunctella* and *L. serrecorne* in storage. These pests feed on dried fruits of mongongo in massive numbers. Morama nuts also indicated to host massive number of *T. confusum* and *P. interpunctella* in storage. *B. hebetor* was identified as a biological control agent to reduce the population of *P. interpunctella* in storage.

3.4 Experiment 4: Determination of the sex ratio and number of parasitoid larvae *Bracon hebetor* hatching from larvae of morama storage pest *Plodia interpunctella*

3.4.1 Materials and methods

3.4.1.1 Study Area

This was a laboratory experiment that was conducted at Botswana University of Agriculture and Natural Resources (BUAN), Department of Crop and Soil Sciences Entomology laboratory in Sebele. It is worth noting that, before the start of the experiment, it was reported that some seeds which were kept in the Department of Food and Science Technology (BUAN) storeroom were attacked by insect pests and these pests were found to be attacked by *Bracon hebetor* parasitoid. This experiment was planned and done to assess the quality and suitability of host species (*Plodia spp*) on the development and reproduction of parasitoid *B. hebetor* reared on morama beans.

3.4.1.2 Collection and preparation

3.4.1.2.1 Rearing of the host

Indian meal moth (*Plodia interpunctella*) was collected from the seeds from Department of Food Science Technology (FST) laboratory, BUAN. These insects were reported to be feeding on deshelled seeds of stored morama by the FST Department staff. At the larval stage, the insects were collected and reared under laboratory conditions at BUAN entomology laboratory. After reaching the adult stage, the moths of two days old (male and female) were collected from the reared stock and held in jar filled with clean morama seeds (not infested by pests) for them to mate and oviposit. The sexes of the moths were differentiated using their sizes, male are smaller than female when freshly emerged. In addition male *P. interpunctella* is tapered at the apex of the abdomen of the female is truncated (Richards and Thomson, 1932).

These mated and laid eggs which later hatched to larvae. The resulting fifth instar larvae from the hatched laid eggs were used for the experiment.

3.4.1.2.2 Rearing of parastoids

B. hebetor parasitoids were collected from the same bag of morama seeds which had a stock culture of Indian meal moth. The parasitoid was then reared by letting it lay eggs in the Indian meal moth larvae. The parasitized larvae were kept in rearing jar container with a muslin cloth cover held in position by an elastic rubber band. The resulting adult parasitoids were used in the experiment. Therefore, *B. hebetor* females within 24 hours of emergence were kept with males for another 24 hours in 100 ml glass jar for them to mate because 80% of virgin *B. hebetor* females mate within the first 15 minutes of being in the presence of male as reported by Ode *et al.* (1995).

3.4.1.2.3 Inoculation

After 24 h, *B. hebetor* females were isolated from the males and introduced individually into experimental arenas containing a full grown host larva (fifth instar). The isolation of the females and males were done by firstly anaesthetically treating them by placing the samples in a freezer for 5 minutes to prevent them from escaping and flying away. The presence of the ovipositor on the females was used as an indicator for different sex.

3.4.1.3 Experimental procedure

Experiments were conducted in the laboratory in a no-choice design in a completely randomised design using 100mm bottles as experimental arenas with full grown wandering stage larvae (fifth instar). According to Hagstrum and Smittle, (1977), *B. hebetor* females attack wandering larvae at a rate 10-fold more than they attack young larvae. There were three treatments based on the number

of larvae. Treatment one had one fifth instar larvae of *P. interpunctella*; treatment two had five fifth instar larvae of *P. interpunctella* and treatment three had ten fifth instar larvae of *P. interpunctella*. Each of these treatments was replicated four times. These were introduced into a 100ml rearing bottles with an open top which were covered with a muslin cloth. The parasitoids were allowed to mate for 24 hours. After 24 hours of mating, a single female was carefully introduced to each treatment of experimental arenas which had larva of a host species. Parasitoids were allowed to attack, paralyse and deposit eggs until the larva (host) die. The parasitized hosts were incubated at room temperature. Observations and monitoring were done until the female died. The number of eggs laid inside each host was recorded by counting the number of emerging larvae of the parasitoids from the host. The number of parasitoids hatching and emerging per host, and the progeny sex ratio per host (presence of ovipositor as indicator) was recorded.

3.4.1.4 Data collection

The number of hosts parasitized, number of parasitoids larvae emerging per host, and the progeny sex ratio per host (presence of ovipositor as indicator) were recorded after 5 days for all the treatment.

3.4.1.5 Data analysis

Analysis of variance (ANOVA) was used to analyse data on response variables using the General Linear Model procedures in SAS 9.2 statistical software (SAS Institute, 2004). Pearson correlation analysis (r) was done to determine the strength and the relationship of number of larvae and number of eggs laid. Similarly, it was also done between progeny sex ratio and the number of *B. hebetor* larvae per host. The means were separated using Tukey's Honestly significant difference test (Zar, 1984) at P<0.05.

3.4.2 RESULTS

Table 10: The average egg counts deposited by single parasitoids depending in relation to host (*P. interpunctella* larvae) number

Number	of	larvae	Average number	of eggs laid	l by	parasitoid \pm SE
(Treatme	nt)					

One	$2.500^{b} \pm 1.50$
Five	$12.25^{ab} \pm 3.42$
Ten	$17.25^{a} \pm 4.96$

* Means within a column followed by the same small letter are not significantly different at P=0.05, Tukey's test *SE means standard error

Table 10 above shows the average number of eggs which were deposited by a single parasitoid depending on the host number. The experiment was done to determine if the host number has an effect in the parasitoid's ability to lay eggs. The number of host larvae *Plodia interpunctella* had a significantly impact on the eggs deposited by the parasitoid *Bracon hebeter* ($F_{6, 2}$ =5.21; P=0.0488). The results presented in the Table 10 show that when one larva and five larvae are used there is no difference in the number of eggs laid by the parasitoid on these larvae. Similarly when five larvae and ten larvae were provided to the parasitoid, there was also no significant difference in the number of eggs laid by these parasitoids on the larvae. However, when comparing the number of eggs laid by parasitoid on the one larva and ten larvae there was significant difference (Table 10).

Table 11:	Comparison	of the	number	of	male	and	female	parasitoids	hatched	from	all	larval
treatments												

Sex	Average number of parasitoids hatched \pm SE
Male	$5.42^{a}\pm1.55$
Female	$5.25^{a} \pm 1.82$

* Means within a column followed by the same small letter are not significantly different at P=0.05, Tukey's test *SE means standard error

Table 11 depicts the comparison of the number of male and female parasitoids hatched from all larval treatments. The table reveals that there was no significant difference of the number of male and female parasitoids hatched among all larval treatments ($F_{1, 17}=0.01$; P=0.9360).

Table 12: The average number of parasitoids hatched from one, five and ten larvae

Number of larvae (Treatment)	Average number of parasitoids hatched ± SE
One	$1.25^{a} \pm 0.65$
Five	$6.13^{ab} \pm 1.44$
Ten	$8.63^{b} \pm 2.63$

* Means within a column followed by the same small letter are not significantly different at P=0.05, Tukey's test *SE means standard error

Table 12 shows the average number of parasitoids hatched from one, five and ten larvae. The experiment was done to determine if number of the host has the influence on the number of parasitoid hatched. The results in the table revealed that there was a significant difference in the number of parasitoid hatched among each treatment ($F_{2, 17}$ =4.49; P=0.027). The table shows that the number of parasitoids hatched in one host larva and five host larvae there is no difference. Similarly with the number of parasitoids hatched from five host larvae and ten host larvae were

not different. However, there was significant difference in the number of parasitoids hatched between one host larva and ten host larvae (Table 12).

Parasitoid sex					
Number of host larvae (Treatment)	Male	Female	Statistics		
One	$1.00^{\mathtt{aA}} \pm 1.00$	$1.50^{aA} \pm 0.96$	F _{2,15} =0.12; P=0.896		
Five	$5.50^{aAB} \pm 2.60$	$6.75^{aA} \pm 1.65$	F _{2.15} =0.12; P=0.514		

 $8.00^{aA} \pm 3.85$

F_{2.15}=0.12; P=0.743

Table 13: Comparison of the average number of parasitoids hatched (males and females) within each treatment and the comparisons of male and female parasitoids across the treatments

* Means within a row followed by the same small letter are not significantly different at P=0.05, Tukey's test

* Means within a column followed by the same capital letter are not significantly different at P=0.05 *SE means standard error

 $9.25^{aB} \pm 4.15$

Ten

Table 13 shows the comparison of the number of parasitoids hatched within each treatment and the comparisons of males and the females across the treatments. The table reveals that there was no significant difference between males and females when the parasitoid (adult) was introduced to one host larva ($F_{2, 15}=0.12$; P=0.896), five host larvae ($F_{2, 15}=0.12$; P=0.514) and ten host larvae ($F_{2, 15}=0.12$; P=0.743). When comparing the average number of male parasitoids (progeny) across the treatments (larvae number), it has been shown that when a parasitoid adult is introduced to a single host larvae, it resulted in small number of male larvae (1.00 ± 1.00) and this was not significantly different from parasitoid adult introduced to a single host larvae (5.50 ± 2.60) (Table 13). However, when a parasitoid adult is introduced to a single host larvae (1.00 ± 1.00), it was revealed that it was significantly smaller than when a parasitoid adult was introduced to 10 host larvae (9.25 ± 4.15). More so, there was no significant difference between five host larvae and ten host larvae treatments in the number of male parasitoids progeny.

Interestingly, when comparing the average number of female parasitoid progeny across the treatments (larvae number), it has been shown that when a parasitoid is introduced to a single host larvae, five host larvae and ten host larvae, they did not differ significant in the number of female parasitoids (progeny).

Table 14: Pearson coefficient correlation (r-values) value between number of host larvae (*P. interpunctella*) and number of eggs laid by parasitoid and also between parasitoid progeny sex ratio and number of host larvae (*P. interpunctella*).

Variables	r-value	
Males	0.556	
Females	0.504	
Number of eggs laid	0.96	

R = 1.0-0.9 (Very Strong Correlation), r = 0.89-0.7 (Strong Correlation), r = 0.69-0.4 (Moderate Correlation), r = 0.39-0.1 (Weak Correlation) (Drakou *et al.*, 2020)

Table 14 depicts the correlation between number of host larvae (*P. interpunctella*) and number of eggs laid and also between progeny sex ratio and number of parasitoid larvae. This table shows that there was very strong correlation between number of host larvae (*P. interpunctella*) and number of eggs laid by parasitoid (r=0.96). However, there was moderate correlation between males and number of host larvae (r=0.556) and also between females and number of host larvae (r=0.504).

3.4.3 DISCUSSION

This study revealed a correlation between host number and the number of eggs laid by a single parasitoid. It has been shown that the density of host P. interpunctella had an influence on the number of eggs deposited by parasitoid *B. hebetor*. The study revealed that *B. hebetor* laid fewer eggs on a single host larva P. interpunctella compared to more eggs laid on a ten larvae host. The number of eggs can be seen as the increase in size or availability of the host. Therefore it implies that if the eggs are of high quality and many, there will be ample quality of food that will enable the parasitoid to reproduce more. A study by Taylor (1988b) on *Bracon hebetor* attacking larvae of *P. interpunctella* also showed similar oviposition behaviour related to host size, where it deposited more eggs on larger hosts than smaller ones. Previous studies reported that when a female B. hebetor was supplied with only one host larva of Adoxophyes orana every day, it never lays more than 7 to 12 eggs per host (Isitan et al., 2011). Van Alpen and Jervis, (1996) explained that some gregarious parasitoids can optimise their reproductive potential by regulating the number of eggs on a host (clutch size). Yu et al. (2003) suggested that parasitoids regulate clutch size based on the host quality. While Taylor, (1988a) reported that the parasitoid is adjusting clutch size to the nutritional value of the host there by avoiding larval competition among progeny. However, Taylor, (1988b) reported that the total number of eggs laid by B. hebetor was independent of the host density, which was opposite to our findings. Yu et al. (2003) Suggested that this was probably due to differences in strains or experimental procedure between the two studies.

This study showed that the size of the population of the host *P. interpuctella* had influence on the gender of the parasitoid *B. hebetor* offspring. This study reveal that when a parasitoid adult is introduced to a single host larvae (1.00 ± 1.00), it was revealed that it was significantly smaller than when a parasitoid adult was introduced to 10 host larvae (9.25 ± 4.15). These results were not

different from the findings reported by Yu *et al.* (2003) who found that the results in male-biased sex ratio in *B. hebetor*, and that the progeny sex ratio (male/total) increased as the host/parasitoid ratio decreased. This study showed that sex ratio (female/total) increased as the number of the host increases or the number of eggs laid in the host increases. This suggests that sex ratio of *B. hebetor* changed from male bias to female bias as the number of eggs on a host increased. These results with *B. hebetor* agree with the models proposed by King (1994), in which the ovipositing female parasitoid controls the sex of the eggs she is laying depending on the host size. However, Eliopoulos and Stathas, (2008) found that sex ratio of *B. hebetor* was unaffected by host density. Bull, (1983) reported that arthropods sex determination is linked to fertilization. Heimpel *et al.* (2008) stated that among insects most species in the Order Hymenoptera have haplo-diploid sex determination.

3.4.4 CONCLUSION

This study finding showed that the numbers of eggs laid by a single parasitoid *B. hebetor* were affected by the density on population of the host larvae *P. interpunctella*. The study revealed that *B. hebetor* laid fewer eggs on a single host larva *P. interpunctella* compared to more eggs laid on a ten larvae host. The study agrees with the alternative hypothesis natural enemies supress potential population of insect pests. Our results indicated that *B. hebetor* reproductive potential is strongly influenced by density population of the host *P. interpuctella*. Furthermore the size of the population of the host *P. interpuctella* had influence on the gender of the parasitoid *B. hebetor* offspring. *B. hebetor* is a commonly used biological control agent in moth pests. We have theoretically verified this fact by showing that the wasp is intrinsically capable of suppressing its host as it is revealed the number of males and females ratio compared to host density.

3.5 Experiment 5: Effect of different storage methods in managing population growth of storage pests of morama and mongongo.

3.5.1 Materials and methods

The experiment involved a comparison of the performance of a Jute bag, woven polypropylene bag (maize meal bag) and bottle container. This research work was meant to determine which control method supports the life of the pest in terms of reproduction by producing suitable conditions for the pest to complete their life cycle. The experiment was conducted for six months and the data was collected at the end of experiment.

3.5.1.1 Experimental Area

The experiment was conducted in entomology laboratory, BUAN under laboratory conditions.

3.5.1.2 Experimental design and procedure

A randomized complete block design was used to test how different storage methods affect population levels of storage pests. Seeds of morama and mongongo were observed to identify pests infesting them. 200g of these seeds were weighed using top pan balance scale (morama was deshelled) and put in three different storage containers which are jute bag, maize meal bag and bottles which are treatments. The containers were placed on a laboratory bench throughout the study. The treatments were replicated four times. Each set-up was left undisturbed for six months to determine the insect species diversity (number of species) and abundance (number of individuals) in each storage container which was recorded at the end of the study.

3.5.1.3 Data collection

Seeds were selected randomly from each treatment and assessed for pest infestation. From each treatment, observations were done at the end of 6 months and this included:

- The final weight of the seeds (possible damage done)
- Number of holes per seed
- Insect diversity per bag and also insect counts per group
- > This included also the natural enemies such as parasitoids

At the end of the study, the seeds in each storage container were weighed and the initial weight was compared with the final weight to show how much damage possibly could have been done. The resultant data of weight was then substituted in the equation as shown below:

- Weight Loss = initial seed mass final seed mass
- Percentage grains weight loss was calculated using the following formula:

Percent weight loss [% WT Loss] = (IM - FM)/IM*100 Where WT is seed weight; IM is initial seed mass; FM is the final seed mass

Exit holes were counted to determine % damage of seeds (Tegegne, 2017)

> % Damage (PD)= (number of seeds with holes/Total number of seeds) *100

3.5.1.5 Data analysis

Analysis of variance (ANOVA) was evaluated using the SAS 9.2 statistical software (SAS Institute, 2004) using the PROC GLM procedures. Treatment means were compared using Turkey test at probability level of 5% (P \leq 0.05).

3.5.2 RESULTS



Figure 17: Weight of mongongo fruits after natural infestation

Figure 17 illustrate the weight of mongongo fruits after natural infestation for 6 months period. After the experimental period, the seeds were found to have been attacked by *Plodia interpunctella* larvae. The fruits were weighed before start of the experiment to get the initial weight (200g). Then, Six months after infestation, the final weight was recorded to be able to get weight differences. This experiment has revealed that the weight of mongongo fruits significantly differed by storage method ($F_{2, 6}$ =67.46; P=0.0001) and this was influenced by the pests that attacked the seeds. It was shown that the weight of the mongongo fruits decreased in maize meal bag. While the final weight of mongongo fruits in Bottle storage had increased to 268g and jute bag storage 222g (Fig. 17). This means each storage method has differently controlled insect pests and the damaged caused by the pests in each storage method was different.

The average numbers of holes per fruit of mongongo after attack by pests are shown in Fig. 18. It is depicted that there was no significant difference in number of exit holes ($F_{2,6}=1.58$; P=0.2802). There was no difference in the number of holes exhibited on bottles, maize meal bag and jute bag

treatment statistically. A high average mean of holes in jute bag (88) was observed compared to lower average means in maize meal bag (66) and bottles (52).



Figure 18: Average number of holes found in the fruits of mongongo



INSECT TYPE

Figure 19: Distribution of insect per insect type

Figure 19 shows the distribution of insects for mongongo according to insect type. It has been revealed that average number of insects among the insect types are not significantly different ($F_{4,42}$ =1.32: P=0.2768).

Number of insects per storage method ± SE						
Insect type	Bottles	Jute bags	Maize meal bag	Statistics		
Beetles	$0.25^{\mathrm{aA}}\pm0.25$	$0.00^{\mathrm{aA}}\pm0.00$	$0.00^{abA}\pm0.00$	F _{2,42} = 1.49: P=0.9815		
Unclassified larvae	$9.75^{aA}\pm5.36$	$0.50^{aA}\pm0.29$	$5.75^{abA}\pm1.44$	F _{2,42} = 1.49: P=0.1432		
Moth	$17.00^{\mathrm{aA}} \pm 14.37$	$0.00^{\mathrm{aA}}\pm0.00$	$0.75^{\mathrm{aA}}\pm0.48$	F _{2,42} = 1.49: P=0.1054		
Parasitoid	$13.00^{aA}\pm9.43$	$0.75^{\mathrm{aA}}\pm0.48$	$5.50^{abA}\pm5.17$	F _{2,42} = 1.49: P=0.0799		
Shell	$13.00^{aAB}\pm9.43$	$0.00^{aA}\pm0.00$	$22.00^{\text{bB}}\pm12.40$	F _{2,42} = 1.49: P=0.0022		
Statistics	F _{4,42} = 2.46: P=0.0714	F _{4,42} = 2.46: P=0.9286	F _{4, 42} =2.46; P= 0.0183			

Table 15: Average number of insects found in each storage method of mongongo

* Means within a row followed by the same small letter are not significantly different at P=0.05, Tukey's test

* Means within a column followed by the same capital letter are not significantly different at P=0.05

*SE means standard error

Table 15 shows the average number of insects found in each method in mongongo. This was done to determine the effect of storage method on increase in insect population. The study revealed that storage methods were not significantly different to reduce moth populations ($F_{2, 42}=1.49$; P=0.1054), beetle population ($F_{2, 42}=1.49$; P=0.9815), larvae population ($F_{2, 42}=1.49$; P=0.1432) and parasitoid population ($F_{2, 42}=1.49$; P=0.0799). However, there was significant different on shell numbers ($F_{2, 42}=1.49$; P=0.0022). Jute bag, maize meal bag and bottle containers methods managed to reduce the number of pest infestation (beetles, larvae, moths, and parasitoids) the same. Jute bags and maize meal bags had no difference on the number of shells. However, there was a difference in the number of shells between bottle storage and other storage methods (jute

bag and maize meal bag). However, in bottle storage moth numbers (17.00 ± 14.37) , it was revealed that it was greater than the numbers of moths observed in jute storage (0.00 ± 0.00) and maize meal storage (0.75 ± 0.48) . Similar trend was observed on the number of unclassified larvae in bottle storage (9.75 ± 5.36) was higher than in jute storage (0.50 ± 0.29) . Also more numbers of parasitoids in bottle storage (13.00 ± 9.43) were more than in jute storage (0.75 ± 0.48) .

The results presented in Table 15 also reveal that there was no difference in insect type within bottle storage method ($F_{4, 42}=2.46$; P=0.0714) and jute bag ($F_{4, 42}=2.46$; P=0.9286). Insect type (beetles, larvae, moths, parasitoids and shells) did not differ in population number when are compered to each other in bottle storage. Similarly when we compare insect type in jute bag storage there was no difference in insect population within the storage when compared to each other. However, there was significant difference in insect type within maize meal bag storage ($F_{4, 42}=2.46$; P=0.0183). Insect type population within maize meal bag between moths and shells were different. An average of 22 shells was found in maize meal bag compared to 0.7 moths that were discovered in the same storage bag.

Figure 20 illustrate the effect of pests in terms of weight loss, number of eggs laid and number of exit holes on morama stored seeds among the three storage methods. Comparisons were made for weight, holes and eggs among the storage method and not within the storage method. The experiment showed that there was no significant difference between the three methods of storage on the weight loss due to pest infestation ($F_{6, 2}=0.77$; P=0.5052). Same trend was observed on the number of eggs laid ($F_{6, 2}=0.63$; P=0.5641) and number of exit holes ($F_{6, 2}=2.39$; P=0.1726) on the fruits of morama. Figure 20 reveal that the weight among storage methods jute bag, bottle and maize meal bag was the same. Comparing the holes damage caused by insect among storage

methods, the results reveal that the damage was the same. Similarly on comparing the eggs found among the storage methods, there was no difference in the number of eggs laid. According to the results presented on Figure 20 above all the three storage methods produced the same environmental conditions for the host pest of morama to survive.



Figure 20: Average performance in weight loss, number of eggs laid and number of exit holes among the three methods on morama stored seeds



INSECT TYPE

Figure 21: Distribution of insect per insect type

Figure 21 shows the distribution of insects for morama according to insect type. It has been revealed that average number of insects among the insect types are not significantly different ($F_{4,42}$ =1.14: P=0.3527).

The average numbers of insects found under each storage method of morama were investigated (Table 16). The findings of this study showed that the methods of storage were not significantly different in moth counts ($F_{2, 42}$ = 2.64: P=0.7284). Same trend was observed for the beetles ($F_{2, 42}$ = 2.64: P=0.3872), larvae ($F_{2, 42}$ = 2.64: P=0.1233) and parasitoid ($F_{2, 42}$ = 2.64: P=0.6028). Therefore, there was no difference on how all the three methods of storage managed to control the number of pest infestation (moths, beetles, larvae and parasitoids). However, there was significant difference on the number of shells ($F_{2, 42}$ = 2.64: P=0.0049). The number of shells in bottle storage was different from other storage method (jute bag and maize meal bag). The number of shell between jute bags and maize meal bag were not different. Zero number of shells was observed on bottle storage method compared to number of shells of the insects that escaped from the storage methods jute bag and maize meal bag.

The table also present the results of insect types (beetles, moths, larvae, parasitoids and shells) within the storage method. The number of insects type within the bottle storage method were not different ($F_{4, 42}$ = 1.14: P=1.000). Same trend was observed within the maize meal bag storage method ($F_{4, 42}$ = 1.14: P=0.1118). Comparing the insects type within the bottle storage, number of beetles, larvae, moths, shells and parasitoids were the same. Also in maize meal bag similar trend was observed. However, the number of insect types within jute storage method was significant difference ($F_{4, 42}$ = 1.14: P=0.023). Insect type population within jute bag between moths and shells were different. Similar trend was observed between parasitoids and shells within jute bag.

Number of insects per storage method ± SE							
Insect type	Bottles	Jute bags	Maize meal bag	Statistics			
Beetles	$0.00^{\mathrm{aA}}\pm0.00$	$1.25^{abA}\pm0.75$	$0.00^{\mathrm{aA}}\pm0.00$	F _{2,42} = 2.64: P=0.3872			
Unclassified larvae	$0.00^{aA}\pm0.00$	$1.50^{\text{abcA}} \pm 0.96$	$0.75^{aA}\pm0.48$	F _{2,42} = 2.64: P=0.1233			
Moth	$0.00^{\mathrm{aA}}\pm0.00$	$0.00^{\text{abA}}\pm0.00$	$0.50^{\mathtt{aA}}\pm0.29$	F _{2,42} = 2.64: P=0.7284			
Parasitoid	$0.00^{\text{aA}}\pm0.00$	$0.00^{\text{abA}}\pm0.00$	$0.75^{\text{aA}}\pm0.75$	F _{2,42} = 2.64: P=0.6028			
Shell	$0.00^{aA}\pm0.00$	$3.25^{\text{cB}} \pm 2.63$	$1.00^{\mathrm{aB}}\pm1.00$	F _{2,42} = 2.64: P=0.0049			
Statistics	F _{4,42} = 1.14: P=1.000	F _{4,42} = 1.14: P=0.023	F _{4, 42} =1.14; P= 0.1118				

Table 16: The average number of insects found in three storage methods of morama

* Means within a row followed by the same small letter are not significantly different at P=0.05, Tukey's test

* Means within a column followed by the same capital letter are not significantly different at P=0.05

*SE means standard error

3.5.3 DISCUSSION

It is generally accepted that 5-15% of total weight of all cereal, oil seed and pulses is lost after harvest (Padin *et al.*, 2002). Weight loss of stored products may results from the feeding by insects, rodents and microorganisms all of which are influenced by environmental conditions. In this study, interestingly there was increase in mongongo seed weight observed in all storage methods instead of weight decreasing due to pest feeding. Bottle container had the highest weight increment after infestation (Fig. 17). The difference between initial weight and final weight could be influenced by additional weight from moisture content as well as insects inside the seed. However, moisture content was not assessed in this study. Tareq *et al.* (2015) reported that moisture percent was increased with passing of time after storing on seeds in jute bag. It was found that increasing moisture content is the cause of increasing seed weight (Pulok *et al.*, 2014). The weight increase could have resulted from the number of larvae which were feeding inside the fruits, which is the more the number of the larvae the higher the weight. This can also imply that the storage methods with higher weight records were able to support the larval development and not support parasitoid host accessibility. Based on this reasoning, the polypropylene bags enable functioning and activity of parasitoid to attack their host (pest). Maize meal bags (polypropylene material) had a decrease in weight lower than initial weight before infestation by pest. The weight reduced because the larvae feeding on the fruits of mongongo were attacked by parasitoids. The bottle containers prohibited large numbers of parasitoids to have access to insect larvae feeding on mongongo fruit flesh. While maize meal bag created the environment that allowed the parasitoids to parasitize the insect larvae feeding inside the bag through puncturing from the outside of the bag.

All the storage methods which were used to keep morama seeds did not differ in terms of weight after feeding by the pest (Fig. 20). However, they all exhibited depressed weight loss after feeding by the pest. Although seed damage measured in maize meal bag was slightly higher numerically than that measured in Jute bag and Bottle container, the weight loss in three storage methods did not differ statistically. In other study reported by Nganga *et al.* (2016), it was demonstrated that great grain damage and weight loss in maize stored in Polypropylene bag (Maize meal bag) and jute bags compared to that of PICS (Purdue improved crop storage) bags. He further explained that the high levels of grain damage and weight loss in polypropylene bag and jute bag may be attributed to high rate of respiration and insect pest multiplication as a result of presence of conducive environment particularly high oxygen concentrations within the bags or moisture loss of the seeds.

This study revealed that all the three method of storage in mongongo has the same conducive environment for insects population growth except on moths in bottle containers. High numbers of moths, parasitoids and unclassified larvae were observed in bottle storage compared jute and maize meal storage. These results suggest that the fruits in mongongo were infested prior to being put into bottles, that's how could the insects get into the bottle. In addition, the lids for some of the bottle containers were found opened and this could have also disturbed the experiment. That's how the parasitoids had gained access into the seal bottle to attack the larvae. More shells of parasitoids, moths and beetles were observed on maize meal bag, it means other insects manage to complete their life cycle and escape the storage bag. In morama no insects were observed in bottles and few insects were observed between jute bag and maize meal bag. Therefore statistical there was no difference in all the three methods of storage to control the number of pest infestation. These findings were different from Mutambuki et al. (2019) who reported that a higher increase in live insect population occurred in polypropylene and jute bag. He further reported that lower number of insects recorded in PICS bags compared to polypropylene bag and jute bags, which were reported to be conducive for build-up insect population. Bottles storage in morama showed zero infestation after 6 months. Therefore it's the best method for storage of morama seeds, reproduction of insects which could pre-infested the seeds before storage was suppressed to unfavourable conditions of low oxygen and high carbon dioxide.

Grain damage which is evident by exit holes or tunnelling made by adult or larval forms of insect influences the pricing of the grain at the market (Meikle *et al.*, 2002). Statistical this study revealed that there was no difference in number of holes caused by insects in all three storage methods. However, more holes were observed in jute bag compared to maize meal and bottle being the lowest hole numbers (Fig. 18). A report on the markets in sub-Saharan Africa discount insect-

damaged maize showed that the discount was 3% (Compton *et al.*, 1998). Although this study did not cover pricing of different grain damage levels, such information would be a great interest to producers of morama and mongongo, thus motivating them to store uninfected seeds of mongongo and morama in bottle storage. High infested, high number of holes and grain quality perception determines whether the grain is could be used for human consumption or animal feed or destroyed (Kadjo *et al.*, 2016).

3.5.4 CONCLUSION

The study showed that there was no difference in the insect numbers among the three storage methods that is jute bag, bottle container and maize meal bag (polypropylene bag). All the three methods of storage manage to suppress the population of insect pests because only low numbers insects were found inside the storage methods. These findings agree with the alternative hypothesis that storage method supress population of insect pest of mongongo and morama in storage. However, polypropylene bag allow natural enemies such as parasitoids to access their host while outside the storage bag.

CHAPTER FOUR

CONCLUSION AND RECOMMENDATIONS

4.1 CONCLUSION

Our knowledge about the insect pests of mongongo and morama is largely incomplete. Diverse numbers of insect pests were identified to be hosted by mongongo and morama in the natural forest and in storage. Accordingly, pests of mongongo and morama were mainly damaging the fruit and the seed. A variety of insects such as the beetles, flies and caterpillars were involved in the destruction of the fruits, pods and seeds. A survey revealed that farmers or users of mongongo and morama do not use control methods or do post-harvest treatment, but the study revealed that mongongo and morama are attacked by destructive insect pests which can cause huge damage if not controlled. Both mongongo and morama have a long shelf life, but the study showed that insects such as *P. interpunctella* and *T. confusum* may lead to post harvest deterioration of fruit and shortening of the shelf life. According to the survey both morama and mongongo users indicated that they store collected morama and mongongo in open weave bags. The study showed that both the jute bag and maize meal bag (woven polypropylene bag) have no significant difference in reducing the natural infestation population of the insect pests. Natural enemies of the pests of morama and mongongo were identified and have proven to control the population of the P. interpunctella moth on storage products of morama. Different species of natural anemies of insect pests of mongngo and morama were identified in this study, which can be used as biological control agent to supress their population.
4.2 RECOMMENDATION

Polypropylene bag is recommended for use by farmers to store morama and mongongo because it showed that it allows the natural enemies to attack their host pest to reduce the population.

CHAPTER FIVE

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