BIOLOGY AND CONTROL OF *Planococcus citri* (RISSO) (HEMIPTERA: PSEUDOCOCCIDAE) ON FIVE YAM VARIETIES IN STORAGE

BY

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MARCH 2016

BIOLOGY AND CONTROL OF *Planococcus citri* (RISSO) (HEMIPTERA: PSEUDOCOCCIDAE) ON FIVE YAM VARIETIES IN STORAGE

A THESIS SUBMITTED TO THE BOARD OF GRADUATE STUDIES

KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY,

KUMASI, GHANA

IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF

DOCTOR OF PHILOSOPHY DEGREE (ENTOMOLOGY)

BY

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MSc. CROP PROTECTION (ENTOMOLOGY)

MARCH 2016

DECLARATION

I hereby declare that no part of this dissertation, which I have submitted to the Board of Graduate studies, KNUST, Kumasi has been published or copyrighted before in Ghana or elsewhere, except publications in peer reviewed journals, which emanated out of the research.

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ABSTRACT

The Citrus mealybug, *Planococcus citri* (Risso) seriously infests yams in storage. However, there is little information on the biology and control of *P. citri* on stored yam locally and the West African sub-region. Incubation and developmental periods, adult longevity, total number of eggs laid and life span of *P. citri* on stored yam (*Disocorea* species) were studied on five yam varieties stored in the laboratory with an ambient temperature of 26.0 - 30.0 ^{0}C and relative humidity of 70.0 -75.0 %. The mean life span values of female *P. citri* on the *Dioscorea rotundata* varieties Pona, Labreko and Muchumudu and the *D. alata* variety Matches and *Dioscorea rotundata* var. Dente were 62.4, 63.5, 66.8, 67.3 and 69.8 days, respectively and those of the male were 23.8, 25.3, 29.6, 30.2 and 33.5 days, respectively. Generally, the adult female lived longer than the adult male. The citrus mealybug was found to prefer *D. rotundata* var. Pona to the rest of the varieties because it developed faster and laid more eggs (497.0 eggs) on it. The least number of eggs (257.0 eggs) was laid on *Dioscorea rotundata* var. Dente and the mealybug development on it was the slowest. It should therefore be the preferred yam for long-term storage. Six chemicals were evaluated for their effect on the third instar and adult *P. citri* in both the laboratory and in improved yam barn storage conditions. In the laboratory, the topical method was used and in the barns, yams were sprayed with a small hand-held sprayer. In the bioassay test, observations on mortality were recorded at 12, 24 and 48 h and subjected to Probit analysis to obtain LD_{50} and LD_{90} values. It was observed that in both stages of the insect pest, the LD_{50} and LD_{90} decreased with exposure period. At the same exposure period, the lethal dosage for the third instar was lower than that of the adult insect. In all the experiments, cypermethrin recorded the lowest LD_{50} and LD_{90} values for both the third instar and adult stages with key soap solution recording the highest LD_{50} and LD_{90} values. In the improved yam barns 0.05 % cypermethrin, 0.07 %

imidacloprid, 1.0 % groundnut oil emulsion, 1.2 %, soyabean oil emulsion, 1.9 % sunlight dishwashing detergent, 2.5 % key soap solution as well as water (control) were sprayed four times (seven days apart) onto the mealybug infested yams. The results showed that a second application of cypermethrin, imidacloprid and groundnut oil to the yams caused over 95 % mortality and was significantly ($P < 0.05$) higher than mortality caused by soyabean oil, sunlight detergent and key soap solution. Therefore, for rapid control of mealybug infestation of yam setts in storage, cypermethrin is recommended and it should be applied at the concentration of 0.049 %. However, the following chemicals, groundnut oil emulsion 0.963 %, soyabean oil emulsion 1.231 %, sunlight dishwashing detergent 1.948 % and key soap solution 2.475 % could be used to effectively manage *P. citri* on ware yam**.**

DEDICATION

This work is first and foremost dedicated to the Almighty God for His protection from the beginning to the end of this research work. Also, I dedicate this work to my family, lovely mother Madam Juliana Wiafe and my late beloved father Mr. Samuel Yaw Asiedu.

ACKNOWLEDGEMENT

My special thanks go to the Almighty God, by whose grace and mercy this research project has come to a successful completion. I wish also to express my heartfelt thanks to my supervisor, Prof. J.V.K. Afun, Associate Professor of Entomology and my Co-Supervisor, Dr. Charles Kwoseh, Senior Lecturer in Plant Pathology and Nematology, Department of Crop and Soil Sciences, College of Agriculture and Natural Resources, KNUST, Kumasi, for their suggestions, contributions, guidelines and time spent in reading the thesis. I really value the efforts these two wonderful scholars invested in this work, without which I would not have come this far.

Special gratitude also goes to my family for their words of encouragement and support throughout my study. I really appreciate them and thank God for their lives.

I also thank Mr. Francis Asamoah Barnieh a retired educationist at Ejisu for his moral support and encouragement. Finally, my gratitude goes to the technical staff of the Insect Laboratory of the Faculty of Agriculture, KNUST, Kumasi, for their assistance during the practical phase of this study, and all my nice friends in the Graduate School.

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1. 0 INTRODUCTION

Yams are indigenous to West Africa with the exception of water yam *Dioscorea alata* L. and Chinese yam *Dioscorea esculenta* (Lour.) Burk., both of which are of Asiatic origin and introduced to West Africa in the 16th and 17th Centuries (Obeng-Ofori, 2007). Yams are monocotyledonous and of the family *Dioscoreaceae* and genus *Dioscorea* and consist of more than 600 species but only six species are important as staple crops consumed in West Africa (Hahn *et al.,* 1987; Dumont and Vernier, 2010).The economically important species grown are *Dioscorea rotundata* Poir (White Guinea yam), *Dioscorea alata* L. (water yam), *Dioscorea bulbifera* L. (aerial yam), *Dioscorea esculenta* (Lour.) Burk. (Chinese yam), *Dioscorea cayenensis* Lam. (yellow yam) and *Dioscorea dumetorum* (Kunth) (trifoliate yam) (Hahn *et al.,* 1987; Tetteh and Saakwa, 1994).

D. rotundata and *D. alata* together make up about 90 % of world production of food yams (Orkwor *et al.,* 1998). The preferred yam species in West Africa is the white yam (*D. rotundata*) since it generally produces tubers with a high dry-matter content that is good for pounding *fufu* (Dumont and Vernier, 2010). Water yam (*D. alata*) is gaining popularity in sub-Saharan Africa because it is easier to propagate than the native white yam. However, it is less acceptable to many consumers because of the higher water content of the tubers, which means it cannot be pounded to produce *fufu* of the right consistency (Dumont and Vernier, 2010).

In Ghana yam is a very important indigenous subsistence and cash crop that is now the most popular non-traditional export food crop, despite years of scientific neglect (Braimah *et al.,* 2007). Yam is cultivated in almost every part of Ghana except the very dry areas of the Sudan savannah zone (Obeng-Ofori, 2007). In Ghana, *D. rotundata* which comprises about 26 varieties is the principal commercial yam and constitutes about

80 % of the total yam produced (Tetteh and Saakwa, 1994).

For human consumption, the peeled yams may be boiled (ampesi), whole small tubers may be roasted, or pieces may be fried in oil, sometimes after partial boiling. In West Africa, they are usually eaten as *fufu*, sometimes added to stews in the West Indies, or eaten as yam croquettes. Yams also have an important role in the socio-religious life in the yam-growing areas of West Africa (Hahn *et al.*, 1987; Tetteh and Saakwa, 1994). In Ghana, considerable amount of ritualism, tradition and funfair has developed around its production; annual festivals are held at planting and harvesting (Hahn *et al.,* 1987; Tetteh and Saakwa, 1994).

The approximate composition of edible yam tubers is: water 65-75 %; protein 1-2 %; fat 0.05-1.5 %; carbohydrates (starch) 15-25 %; fibre 0.5-1.5 %; ash 0.7-2.0 % and about 810 mg/100 g of ascorbic acid (Jaleel *et al.,* 2007). The loss by peeling is usually 5-15 % (Jaleel *et al.,* 2007). Yams also have medicinal properties and the tuber is said to contain some pharmacologically active substances including dioscorine, saponin and sapogenin (Jaleel *et al.,* 2007).

Yam beetles, of which the widespread species is *Heteroligus meles* (Billb.) are the most destructive pests of yams in West Africa (Hahn *et al.,* 1987). The adult beetles feed on the tubers, causing hemispherical lesions. They can also damage the growing point of newly planted setts, which then fail to grow, or produce small useless tubers (Hahn *et al.,* 1987). Stored yam tubers are susceptible to a myriad of storage pests including insects. The most important insect pests of yam in storage include the moth *Euzopherodes vapidella* Mann. (Lepidoptera: Pyralidae), *Planococcus citri* (Risso) (Hemiptera:

Pseudoccocidae) and *Aspidiella hartii* Cockerel (Hemiptera: Diaspididae) (Forsyth, 1966; Hahn *et al.*, 1987; Williams and Watson, 1988). Sauphanor and Ratnadass (1985) reported that insect pests could cause as much as 25 % weight loss in yam four months in storage.

The citrus mealybug, *Planococcus citri* is a polyphagus species known from all zoogeographical regions (Williams and Watson, 1988). It is one of the most common pests in nearly all greenhouses and nurseries, where it attacks a wide range of ornamentals, citrus and orchard crops in many temperate and tropical regions (McKenzie, 1967; Blumberg *et al.*, 1995). The nymphs and adult females cause damage to host plants with their piercing-sucking mouthparts, which they use to suck sap and remove nutrients. As a result, the plants often become stunted, distorted, or yellowed and show reduced vigour. They excrete honeydew, which provides a medium for the growth of black sooty mould fungi (Al-Ali, 1996; Smith *et al.,* 1997; Heinz *et al.,* 2004). Black sooty mould fungi are detrimental to plants because they cover leaves, thus reducing photosynthesis and inducing plant stress (Heinz *et al.*, 2004). The citrus mealybug is also known as a vector of some important plant viruses such as Banana Streak Virus (Lockhart and
Olszewski, 1993; Su, 1998, 2000; Kubiriba *et al*., 2001; Watson and Kubiriba, 2005).

Infestation of stored yam tubers by *P. citri* results in shrivelling of the tubers, which become light in weight, fibrous and unpalatable and therefore, lose their market value.
Their attack also predisposes the yams to fungal infection leading to rotting of tubers.
Badly affected tubers fail to sprout, if planted (Obeng-Ofori, 2007).

There is, however, not much information on the biology and control of *P. citri* on stored yam either locally or in the West African sub-region. The goal of this study therefore, was

to determine the relative susceptibility of five varieties of yam to *P. citri* and determine the efficacy of suitable chemical(s) for the management of *P. citri* in storage.

The specific objectives of this research were to:

1. Determine the incubation period of *P. citri* on the various yam varieties.
2. Determine the developmental periods and longevity of *P. citri* on the various yam varieties.
3. Determine the total number of eggs laid by *P.citri* on the various yam varieties.
4. Determine the oviposition period of *P. citri* on the yam varieties.
5. Establish the offspring sex ratio of *P. citri* on the yam varieties.
6. Assess the survival rate of *P. citri* on the yam varieties.
7. Determine the mode of reproduction of *P. citri* on the yam varieties.
8. Determine the growth index of *P. citri* on the yam varieties.
9. Determine the lethal dose of suitable chemicals for the third instar larvae and the adult *P. citri.*
10. Evaluate the efficacy of suitable chemicals against *P. citri* on infested yams in improved yam barns.

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CHAPTER TWO

2.0 LITERATURE REVIEW

2.1.0 Morphological features of yam

2.1.1 *Dioscorea rotundata* Poir.

White yam (*Dioscorea rotundata* Poir.) is also known as white yam, Guinea yam, or eight-month yam. *D. rotundata* is the most important cultivated species in West Africa, from where it originated. The main area of cultivation is from the eastern Cote d'ivore through Ghana, Togo and Benin to the Cameroun border of eastern Nigeria (Purseglove, 1976). Many cultivars, which vary in size, shape and palatability of the tubers, occur in West Africa. The tubers are normally cylindrical with rounded or pointed ends, smooth brown skins and white mealy flesh, but they may assume distorted shapes. Normal tubers weigh 2-5 kg, but in good soils, 10 kg is common and weights of 20-25 kg have also been recorded (Purseglove, 1976).

The stems of the yam are cylindrical, 10-12 m long, twining to the right, usually spiny; the leaves are usually opposite, simple, 10-12 x 6-8 cm, broadly cordate, acuminate, usually dark glossy green (Purseglove, 1976). The inflorescences are very similar to *D. cayenensis* (q.v.), often in groups of four (Purseglove, 1976).

In Ghana there are 26 varieties of *D. rotundata* namely Pona, Labreko, Dente, Zung, Puupu, Muchumudu, Kyire Kumasi, Serwaa or Bombatenga, Nemare or Dobidi, Lilia, Nananto, Tede, Tibere, Ntonto, Brass, Maale, Kasante, Obormaale, Punjor, Kprinse, Jatuba-Larbako, Siata, Sakata, Lopre, Maria, Mpuano, Dokoba and Nyereba (Tetteh and Saakwa, 1994). Varieties are given local names, which more or less describe certain attributes they have, or are named after whoever introduced them to the area (Tetteh and Saakwa, 1994).

Three varieties of *D. rotundata*; Pona, Labreko and Dente are cultivated in all yam production areas in Ghana while other varieties are localised (Tetteh and Saakwa, 1994). Factors that influence the choice of variety that farmers grow include consumer taste preference, early maturity, storability, yield, adaptability and availability of planting materials, in descending order of importance (Tetteh and Saakwa, 1994). The varieties, Pona and Labreko are the most popular. Even though farmers claim that Pona is the lowest yielding variety and stores poorly, they still prefer to grow it because it tastes good and matures early (Tetteh and Saakwa, 1994). Consumers are prepared to pay more for it. Its early maturity helps to fill a hunger gap when other yams are not on sale. Dente is another variety that is popular among farmers because it can be stored for a long time and is thus, available when other yams have spoilt (Tetteh and Saakwa, 1994).

2.1.2 *Dioscorea alata* L.

It is also known as greater yam, water yam, winged yam or Asiatic yam and rarely as white yam. *D. alata* is a polymorphic species. Portuguese and Spanish traders carried it in the sixteenth century to West Africa and the New World and it is now pantropical in distribution (Purseglove, 1976). It is the highest yielding of the cultivated yams and is now the preferred species in most parts of the world (Purseglove, 1976). It is less highly regarded than the indigenous *D. rotundata* in West Africa, as it does not produce satisfactory *fufu* (local dish). It is the most popular yam in the eastern Carribbean but *D. rotundata* and *D. cayenensis* appear to be preferred in Jamaica (Purseglove, 1976). The stems are square, winged, twining to the right, green or purplish and spineless. The leaves are variable in shape and size, opposite, ovate, deeply cordate, acuminate and glabrous. Flowers are globose in axillary inflorescences and male flowers in axillary panicles are up to 25 cm long. The female flowers are in simple, solitary, glabrous, short spikes.

Majority of cultivars seldom produce fertile seeds and some are completely sterile. The fruits are transversely elliptic, three winged, about 2-5 cm long and 3.5 cm broad, the seeds are orbicular and winged all round (Purseglove, 1976).

The tubers are variable in size, shape and colour, usually produced singly and they are often large, weighing 5-10 kg. The tubers are mostly cylindrical but vary from long and serpentine to almost globular, but branched, lobed, flattened and fan-shaped tubers occur in some cultivars; the outer cortex and flesh vary from white to deep reddish purple in colour. Axillary tubers or bulbils are formed in leaf axils. A large number of cultivars have been recorded throughout the tropics varying in shape, colour of leaves, stems and tubers. The normal growing period is eight to10 months and the tubers have a dormancy period of three to four months before sprouting (Purseglove, 1976).

2.1.3 The yam tuber and its nutritional composition

The tuber size and shape of yam varies depending on the species and growing conditions and may range from two to three metres in length and over 50 kg in weight (Asiedu, 1986). The tubers of most important cultivars are cylindrical in shape, with some root 'hairs'. The outer part of the tuber forms several layers of cork which constitute effective protection from lesions, water loss and penetration of pathogens from the soil or storage compartments (Asiedu, 1986). The inner part of the tuber is formed by parenchyma tissue which is interwoven with vascular channels. In the tissue is stored carbohydrate, mainly in the form of starch. Even though water and carbohydrate form the bulk of the tuber, it also contains non-carbohydrate components (Asiedu, 1986).

Yam is second to cassava as the most important tropical root crop but from a nutritional point of view, it is better than cassava on account of its higher vitamin C (40-120 mg/g edible portion) and crude protein content (40-140 g/kg dry matter) (Opara, 1999). Information on the nutritive value of yam has been highlighted by several authors (Bradbury and Holloway, 1988; Opara, 1999; Afoakwa and Sefa-Dedeh, 2001).

Generally, the ash content of yam gives an indication of its mineral status (Osagie, 1992). Yam tubers have high contents of moisture, dry matter and starch. They are relatively good sources of some minerals. They contain appreciable amount of potassium, a mineral that helps to control blood pressure (Osagie and Eka, 1998). Yam is also a good source of manganese, a trace mineral that helps with carbohydrate metabolism and acts as a cofactor in a number of enzymes important in energy production and antioxidant defences. It also contains traces of vitamin B- complex (Barquar and Oke, 1977).

According to Bradbury and Singh (1986) total ascorbic acid content of yam tubers is about 50 % greater than that of cassava; values ranging from 200-2100 μg/100 g have been reported for various species. Yam also contains the limiting essential amino acids, isoleucine and sulphur-containing amino acids (Bradbury and Singh, 1986). Moorthy and Nair (1989) reported phosphorus content between 0.011 and 0.015 % for *D. rotundata*. Asemota *et al*. (1992) found phosphorus content to be higher in *D. alata* than in *D. rotundata* and *D. cayenensis*. Table 2.1 shows the nutritional composition of yam from different authors.

2.2.0 Constraints and prospects to yam production

Constraints to yam production include high labour demand for most cultural operations, high cost of labour and other inputs such as planting materials, unreliable sources of credit,

pests, diseases, declining soil fertility and unpredictable rainfall pattern (Degras, 1993; Tetteh and Saakwa, 1994), which separately or jointly can cause severe yield losses. However, yam has great prospects of providing multiple opportunities for poverty reduction and improved nourishment of poor people in Ghana or in the West

African sub-region if efforts are made to overcome the constraints to its production (Tetteh and Saakwa, 1994; Aidoo *et al.,* 2011).

Table 2.1. Nutrient contents of yam species (*Dioscorea* spp.) per 100 g fresh edible tuber portions.

Nutrient (g/100 g)	*D. alata*	*D. rotundata*	*D. cayenensis*	*D. esculenta*	*D. dumetorum*
% Moisture	65.0-78.6	50.0-80.0	60.0 – 84.0	67.0 - 81.0	67.0-79.0
% Carbohydrate	22.0 - 31.0	15.0 - 23.0	16.0	17.0 – 25.0	17’0 – 28.0
% Starch	16.7-28.0	26.8-30.2.0	16.0	25.0	18.0-25.0
% Free sugar	0.5-1.4	0.3-1.0	0.4	0.6	0.2
% Protein	1.1-3.1	1.1-2.3	1.1-1.5	1.3-1.9	2.8
% Crude fat	<0.1-0.6	0.05-0.1	0.06-0.2	0.04-0.3	0.3
% Fibre	1.4-3.8	1.0-1.7	0.4	0.2-1.5	0.3
% Ash	0.7-2.1	0.7-2.6	0.5	0.5-1.5	0.7
Phosphorous(mg)	28.0 -52.0	17.0	17.0	35-53	45-0
Calcium (mg)	28 -38	36.0	36.0	12-62	52-0
Vitamin C (mg/100g)	2.0-8.2	6.0-12.0	-	-	-
Iron (mg)	5.5-11.6	5.2	5.2	0.8	-
Food energy(kcal)	140.0	142.0	71.0	112.0	122.0
β-carotene(μg)	5-10	-	-	-	-
Thiamine (mg)	0.05- 0.10	-	-	0.1	-
Riboflavin (mg)	0.03- 0.04	-	-	0.01	-

Niacin (mg)	0.5	-	-	0.8	-

Sources: Coursey (1967); Eka (1985); Bradbury and Holloway (1988); Osagie (1992); Muzac-Tucker *et al*. (1993); Asiedu *et al*. (1997) and Opara (1999).

Nutritionally, yam is a major staple providing food for millions of people in the world (Aidoo, 2009). In the face of rapid population growth, there is the urgent need for increased production and supply of yam to satisfy domestic and export demand (Asumugha *et al.,* 2009). In Ghana, yam constitutes about 13 % of household food budget in urban centers (Aidoo *et al.,* 2011). Yam exports contribute significant foreign exchange earnings to the Ghanaian economy (Ohene-Yankyera *et al.,* 2011).

2.3.0 Pests of yam in storage

2.3.1 Nematodes

Three types of nematodes are often found in yam tubers, the yam nematode (*Scutellonema bradys* {Steiner & LeHew, 1933} Andrássy, 1958), the lesion nematode (*Pratylenchus coffeae* {Zimmermann 1898} Filipjev and Schuurmans Steckhoven 1941) and the root-knot nematode (*Meloidogyne* spp.) (Bridge *et al.*, 2005). If a yam is not infested the flesh just under the skin is clean. However, if yam is infested with S. *bradys* and *P. coffeae*, the skin become cracked and the flesh immediately under the skin is stained yellow and black/brown. This dry rot beneath the skin gradually spreads into the tuber flesh. Root-knot nematode makes the yam tuber galled with proliferation of root hairs than normal. Yams affected by *S. bradys* and *P. coffeae* will deteriorate very quickly whereas those with root-knot will generally last through storage (Wilson, 1980). Fungal and bacterial rots cause much loss in storage and often the pathogens, which cause these rots, invade the yams through wounds and nematode damage (Wilson, 1980).

2.3.2 Insect pests of yam in storage

Mealybugs such as *Planococcus citri* (Risso), *Planococcus dioscorea* (Will), *Pseudococcus brevipes* (Ckll.), *Planococcus halli* Ezzat & McConnell, *Phenacoccus gossypii* Townsend & Cockerell and *Ferrisia virgata* (Cockerell) and scale insects such as *Aspidiotus destructor* Signoret, *Aspidiella hartii* (Ckll.) attack stored yams (Braimah *et al*., 2007) as they feed, the insects reduce the food reserves in the tubers. Both mealybugs and scale insects reduce the value of the yams as food, and make the yams too weak to sprout vigorously when used as planting setts (Onwueme, 1978; Wilson, 1980; Sauphanor and Ratnadass, 1985). Other insect pests such as the yam moth, *Euzopherodes vapidella* Mann (Lepidoptera: Pyralidae) and ginger weevil, *Elytroteinus subtruncatus* (Fairmaire) damage yams in storage (Onwueme, 1978; Wilson, 1980; Sauphanor and Ratnadass, 1985). They lay eggs on stored yams and when these eggs hatch, the larvae bore into the yam and eat the flesh. As the larvae feed, they leave piles of brown frass that can be seen on the outside of the yam. The larvae mature in the rotten yam (Onwueme, 1978; Wilson, 1980; Sauphanor and Ratnadass, 1985).

2.4.0 Citrus mealybug

2.4.1 Taxonomy

Mealybugs belong to the Class Insecta, Order Hemiptera, Suborder Sternorrhyncha, Superfamily Coccoidea, Family Pseudococcidae, SubFamily Pseudococcinae, Genus *Planococcus* and Species *citri* (Cox, 1989). Insects belonging to the family Pseudococcidae are scales that are soft bodied, oval, flat and distinctly segmented. They are covered with a white mealy wax, which extends into spines along the body margin and the posterior end. The presence of legs is a characteristic of the Subfamily Pseudococcinae. The female of Pseudococcinae discharge abundant honeydew on which sooty mould can

grow (Ortu *et al.,* 2002). The species mainly differ by the thickness and the length of the waxy spines (Ortu *et al.,* 2002). *Planococcus citri* is commonly known as citrus mealybug.

2.4.2 Distribution and host range of citrus mealybugs

The citrus mealybug, is a highly polyphagous and a cosmopolitan species**.** In Africa, it is known to occur in Angola, Cote d'ivore, Eritrea, Ethiopia, Ghana, Kenya, Malawi, Nigeria, Principe, Sao Tome, Senegal, Sierra Leone, South Africa, Sudan, Swaziland, Tanzania (including Zanzibar), DR Congo, Zambia and Zimbabwe (Watson and Kubiriba, 2005). It has been recorded on a wide variety of fruit, beverage, vegetable crops and ornamental plants (Cox, 1989). *Planococcus citri* was recorded on *Musa paradisiaca* L. from Ghana (Williams and Matile-Ferrero, 2000). In transmission test, Su (1998) successfully demonstrated transmission of Banana Streak Virus (BSV; genus *Badnavirus*) by *P. citri.*

2.4.3 Life cycle of citrus mealybugs

Life cycle (egg-to-egg-laying adult) duration ranges from 20 to 44 days. Citrus mealybug populations are generally composed of about equal numbers of males and females. It completes two to seven generations per year (Ortu *et al.,* 2002; Gutierrez *et al.,* 2008). The citrus mealybug female can produce about 600 eggs that are laid in groups in cottony structures called ovisacs. Generally, within two to 10 days, new mealybugs (crawlers) emerge and start to go out of the ovisac (Coffee Board Research Department, 1984; Ortu *et al.,* 2002).

Male citrus mealybugs have four nymphal stages instars and each larval stage is separated by a moult (Chong *et al.,* 2008; Vennila *et al.,* 2010; Raphael *et al*., 2015). Based on

laboratory studies on coffee leaves, the first nymphal stage lasts for seven to 14 days, the second, six to 16 days, the third, two to three days, and the fourth, one to six days (Coffee Board Research Department, 1984). Approximately four days into the second instar, a black tinge develops around the insect body. Two days later, the nymph starts spinning a cocoon around itself. This cocoon is continuously spun increasing in density until the winged adult mealybug is ready to emerge two moults later (Coffee Board Research Department, 1984; Fand *et al.*, 2014).

Female mealybugs have only three nymphal stages (Chong *et al.,* 2008; Vennila *et al.,* 2010; Raphael *et al*., 2015). The first nymphal stage lasts seven to 17 days, the second five to 13 days, and the third five to 14 days (Coffee Board Research Department, 1984; Fand *et al.,* 2014). Male mealybugs live for two to four days after the final larva moult and females live for about 88 days as adults and may start laying eggs 15 to 26 days into her adult life (Coffee Board Research Department, 1984; Fand *et al.,* 2014).

Raphael *et al*. 2015 observed that the duration of first instar males varied from seven to nine days and females from seven to 11 days. The second instar duration of males varied from six to nine days, whereas in females the variation was from four to 14 days. In studies of the biology and development of *P. citri*, Raphael *et al*. 2015 observed that the duration of the first instar was the longest and could last up to four days longer than the second instar.

2.4.4 Description of life stages of the citrus mealybug

The adult female is flat, oval in shape with a body length of 2.5 mm to 5 mm with 18 pairs of conical waxy radii arranged externally along the edges, 17 of which are equal and the anal one slightly longer (no more than the double length of the others). The body colour is

mainly brownish yellow, covered with a powdery wax through which a distinguished transversal line of segmentation appears (Ortu *et al.,* 2002; Raphael *et al*., 2015). The adult male has only a pair of wings with an elongated body nearly equal in length as the female. The male is reddish in colour and has two long waxy radii (Ortu *et al.,* 2002; Raphael *et al*., 2015).

The eggs from the female are oblong, yellow and are enmeshed in a dense, fluffy, white ovisac. The first instar larva called crawler is tiny, oval and yellow, with red eyes and distinct antennae. The second instar larva is bigger than the first instar, oval in shape and has six segmented antennae, the third-instar larva resembles the larger adult female, it has a yellowish body colour and seven segmented antennae (Ortu *et al.,* 2002; Raphael *et al*., 2015). Plate 2.1 is a tuber of *D. rotundata* var. Pona infested with the various life stages of *P. citri.*

Plate 2.1. A tuber of *D. rotundata* var. Pona infested with various stages of *P. citri*

2.4.5 Mode of reproduction of citrus mealybug

Planococcus citri reproduces sexually and there is a strong dimorphism between the sexes both in life history and behaviour (Laura *et al.,* 2010). While the sexes are indistinguishable as nymphs, males undergo a form of metamorphosis after the second instar and adult males are winged, while the females do not undergo metamorphosis and grow much larger than the males (Laura *et al.,* 2010). Additionally males do not feed after their second instar and adult males lack functional mouthparts, while females continue feeding until they die. This results in a large difference in lifespan between the sexes, with males only living up to three days after eclosion while females can live several weeks after becoming reproductively mature (Ross *et al.,* 2010). Adult female are almost completely sedentary and the highly mobile crawlers (first instar nymphs) are assumed to be the main agent of dispersal (Gullan and Kosztarab, 1997; Ross *et al.,* 2010).

2.4.6 Sex ratio of citrus mealybug

Sex allocation is an important reproductive decision that can have significant effects on an individual's fitness (West, 2009). Environmental factors acting on parents can affect sex allocation through parental condition. Trivers and Willard (1973) showed that, if there is variation in parental condition and if the fitness of one offspring sex is more strongly affected by their parent's condition, then parents should bias their offspring sex ratio towards the sex that either benefits most or suffers least from their condition.

Several environmental factors are known to affect both parental condition and offspring sex ratio. These factors include extreme temperature, drought, parental age and lack of

resources (Cockburn *et al.*, 2002; West, 2009). Alternatively, environmental factors acting upon the offspring themselves can alter sex allocation, as such effects might again influence the fitness return parents get from their offspring (Trivers and Willard, 1973). A variety of factors could have differential fitness effects on offspring. One particular factor that has been the focus of many sex allocation studies is the level of competition between kin, as competition between siblings can reduce the fitness return of offspring to their parents and parents are therefore expected to over produce the sex that suffers least from kin competition i.e. local resource competition theory (Charnov, 1982; West, 2009).

In *P. citri,* several factors experienced by females have been found to affect sex allocation. These include population density and food (Varndell and Godfray, 1996; Ross *et al.*, 2010), temperature (Nelson-Rees, 1960) and age (Nelson-Rees, 1960; Ross *et al.*, 2010). The observed effects are of particular interest for two reasons. First, mealybugs have an unusual genetic system, paternal genome elimination (PGE), whereby both sexes develop from fertilised eggs, but in males, the paternal genome is deactivated during development and lost from the germline during spermatogenesis (Brown and Nur, 1964; Nur, 1980). As a consequence, males have haploid gene expression and only transmit maternal genes (i.e., PGE is akin to true haplodiploidy in terms of transmission genetics and gene expression patterns (Brown and Nur, 1964; Nur, 1980).

Fox *et al.* (1990) observed correlation between host plant quality and sex ratio of emerging parasitoids as more female parasitoids emerged from hosts reared on highnitrogen plants than on low-nitrogen plants. On rapidly growing willow plants, the sex ratio of the willow sawfly *Euura lasiolepis* was female biased, while on slow-growing plants it was male biased (Craig *et al.*, 1992). Similar effects are also seen in other sawflies xylophagous leafhoppers (Brodbeck *et al.,* 1999) and aphids (Fujita and Mitsuhashi, 1995).

2.4.7 Factors affecting oviposition

Several factors such as the nutritional and biochemical properties of a substrate influence the oviposition of insects (Osuji, 1976). In majority of insects, total number of eggs laid is largely related to adult nutrition and amount of food ingested. The type of food on which an insect is bred influences the number of eggs laid (Noha and Shaaban, 2010). Presumably, better foods enable the larvae to grow bigger and yield adults with greater potential for egg production.

It has been shown that there is an optimum temperature at which maximum oviposition by an insect occurs (Howe and Currie, 1964). The number of eggs laid by adult female citrus mealybug is temperature dependent with females laying less than 100 eggs at temperatures above 30 ^{o}C, but laying over 400 eggs at 18 ^{o}C (Copland *et al.*, 1985). It has been demonstrated that relative humidity influences the number of eggs laid by insects. At 30 ^{0}C, *Callosobruchus analis* (F) laid its highest mean number of eggs (73.2) at 70 % relative humidity, *Zabrotes subfasciatus* (Boheman) and *Callosobruchus maculatus* (F) laid their highest mean number of eggs of 41.4 and 106.2 respectively at 100 % relative humidity (Howe and Currie, 1964). The number of eggs laid at 2 % relative humidity by *C. maculatus* was only half the number laid at 100 % relative humidity (Howe and Currie, 1964).

2.4.8 Effect of environmental factors on mealybug development

Temperature is the driving force for mealybug development, although development times and temperature thresholds differ among species. For instance, *Pseudococcus maritimus* (Ehrhorn) have two generations in California's interior valleys (Geiger and Daane 2001), whereas *Planococcus ficus* (Signoret) have seven generations in the same region

(Gutierrez *et al.,* 2008) but was reported to have only three generations per year in Italy (Ben-Dov 1994). Similarly, *P. citri* in Brazil has six generations per year in the south, but up to 11 per year in the northeast where grapes are produced all year round (Gutierrez *et al.*, 2008).

Noha and Shaaban (2010) found that host plants and temperatures greatly influenced the development of *P. citri*, the lowering of temperature increased the dimension of the mealy bug and lengthened the developmental period, they studied the biological parameters of the citrus mealybug, at three different constant temperatures (i.e. 18, 24 and 30 ^{o}C) on citrus, *C. aurantifolia* Swing. The mean durations of the first instar were 8.5 ± 0.53, 6.0 ± 0.67 and 3.4 ± 0.52 days at 18, 24 and 30 ^{o}C, respectively. Second instar lasted for 11.5 ± 0.71, 9.2 ± 0.92 and 5.2 ± 0.79 days, respectively. While third instar durations were 12.7 ± 0.82, 10.0 ± 0.67 and 8.2 ± 0.82 days, respectively. Incubation periods were 7.3 ± 0.67, 4.5 ± 0.53 and 2.3 ± 0.48 days, respectively. The generation time was 47.3 ± 2.11, 34.2 ± 1.95 and 21.4 ± 2.45 days, respectively. As a result the durations of the adult female longevity were 17.3 ± 0.57, 13.1 ± 0.66 and 11.7 ± 0.95 days, respectively. The results indicated that 30 ^{o}C was the most suitable temperature for the citrus mealybug, *P. citri* life, because it resulted in the highest oviposition (362.3 ± 4.95 eggs/female), the shortest incubation period (2.3 ± 0.48 days) and adult longevity (11.7 ± 0.95 days).

Noha and Shaaban (2010) also studied the biological parameters of the citrus mealybug, *P. citri* at three different constant temperatures (i.e. 18, 24 and 30 ^{o}C) on guava, *Vitis vinifera* L. the mean durations of the first instar were 10.6 ± 0.70, 8.3 ± 0.48 and 5.5 ± 0.53 days at 18, 24 and 30 ^{o}C, respectively. Second instar lasted for 14.6 ± 0.70, 11.7 ± 0.82 and 8.7 ± 0.67 days, respectively. While third instar durations were 15.1 ± 0.99, 12.6

± 0.52 and 9.4 ± 0.52 days, respectively. Incubation periods were 9.6 ± 0.70, 7.4 ± 0.52 and 4.5 ± 0.53 days, respectively. The generation time was 59.5 ± 2.55, 47.4 ± 1.76 and 32.6 ± 2.44 days, respectively. As a result the durations of the adult longevity were 14.7 ± 0.55, 10.7 ± 0.88 and 8.4 ± 0.21 days, respectively. The results indicated that 30 ^{o}C was the most suitable temperature for the *P. citri* life, because it resulted in the highest oviposition (373.3 ± 1.70 eggs/female), the shortest incubation period (4.5 ± 0.53 days) and adult longevity (8.4 ± 0.21days).

Noha and Shaaban (2010) again studied the biological parameters of the *P. citri* at three different constant temperatures (i.e.18, 24 and 30 ^{o}C) on grape**,** *Psidium guajava* L. the mean durations of the first instar were 12.2 ± 0.63, 10.1 ± 0.57 and 6.9 ± 0.74 days at 18, 24 and 30 ^{o}C, respectively. Second instar lasted for 15.9 ± 0.74, 13.2 ± 0.63 and 10.1 ± 0.57 days, respectively. While third instar durations were 16.8 ± 0.42, 14.9 ± 0.99 and 10.6 ± 0.52, respectively. Incubation periods were 11.1 ± 0.88, 8.7 ± 0.48 and 5.6 ± 0.52 days, respectively. The generation time was 67.1 ± 1.76, 55.6 ± 1.65 and 38.8 ± 1.56 days respectively. As a result the durations of the adult longevity were 23.8 ± 0.54, 19.1 ± 0.76 and 15.7± 0.76 days, respectively. The results indicated that 30 ^{o}C was the most adequate tested temperature for the citrus mealybug, *P. citri* life, because it resulted in the highest oviposition (379.8 ± 2.97 eggs / female), the shortest incubation period (5.6 ± 0.52 days) and adult longevity (15.7 ± 0.76 days). Noha and Shaaban (2010) found that the host plants and temperatures greatly influenced the development of *P. citri*. The lowering of the temperature increased the dimension of the mealybug and lengthened the developmental period. The results on citrus, guava and grape showed that the life cycle of the citrus mealy bug, *P. citri* at 30 ^{o}C were 21.4 ± 2.45, 32.6 ± 2.44 and 38.8 ± 1.56 days, respectively. These results indicated that *P. citri* prefers citrus followed by guava and grape.

Polat *et al.* (2008) observed that development, longevity and fecundity of *P. citri* varied according to the suitability of the host plant on which it was bred. Tania *et al.* (2010) in their studies on the influence of different plants substrates on development and reproduction of *Pseudococcus calceolariae* (Maskell) observed that the shortest developmental time, highest female fecundity and egg fertility were obtained on sprouted potatoes, than on lemons or butternut squash.

2.5.0 Management of citrus mealybugs

Mealybugs are difficult to control for several reasons; (i) They are tiny (less than 4 mm long) and move infrequently. Therefore, it is easy for them to go unnoticed, particularly at the onset of an infestation when the numbers are low. (ii) Mealybugs most often are attached to plants in places that make them difficult to detect and difficult to contact with insecticides. (iii) The waxy secretion that covers their bodies provides protection against water-soluble insecticides and (iv) Infestation usually comprises of overlapping generations and certain developmental stages are not susceptible to insecticides. Thus, repeated applications are required in order to treat the vulnerable stages within the population as they emerge (Saeed *et al.*, 2007; Dhawan *et al.*, 2008; Okigbo, 2008).

2.5.1 Monitoring as a control tool

Monitoring systems can improve pest detection making it possible to avoid over and under spraying. They therefore form the backbone of insect pest management (Brown and Pringle, 2006). One of the main reasons noted by Van Der Merwe (2000) for the recent increase in mealybug populations in apple and pear orchards is the absence of effective pest monitoring systems on farms. Swart (1977) also recommends the careful determination of the status of mealybug infestations in orchards to manage mealybugs

effectively and economically. Swart (1977) standardized a method of sampling and inspection of fruit before they enter pack houses. The method is still recommended to date (Barnes, 1992; Van Der Merwe, 2000).

There are no simple and effective methods to visually monitor vineyard mealybugs, and the process itself can be time-consuming and laborious. As exemplified for *Pseudococcus maritimus* (Ehrhorn), the accuracy of monitoring plant material depends on mealybug population density and the number of samples needed for an accurate count is often high because most mealybugs have a clumped distribution pattern (Geiger and Daane, 2001).

The appropriate sampling programs also vary throughout the season, depending largely on mealybug location as there are periods when much of the population is hidden under bark rather than exposed on leaves. Also, as species have different numbers of annual generations and preferred feeding locations throughout the season, there is not a single sampling procedure appropriate for all vineyard mealybugs. In most vineyards, signals of an infested vine can be used to aid the sampling program. First, ants are closely associated with mealybugs (Ripa and Rojas, 1990; Addison and Samways, 2000) and their presence can help select vines for further sampling. Second, honeydew on the leaves can also be a good signal; a large population hidden under the bark will excrete enough honeydew that the infested trunk region will have a darker, wet appearance (Daane *et al.,* 2011). Third, when some mealybug species numbers build, their feeding damage may cause leaves to turn yellow or brown and drop from the vine (Daane *et al.*, 2011).

A faster sampling method is the use of sticky traps baited with sex pheromone to lure in and trap adult winged males (Rotundo and Tremblay, 1975). These pheromones can be synthesized and used in the field (Bierl-Leonhardt *et al.*, 1981). Numerous sex pheromones

have recently been identified. A recent but general, monitoring system for pests on pome fruit was developed by Brown and Pringle (2006). This system is based on scouting, trapping, pre-thinning and pre-harvest damage assessments conducted in orchard subdivisions.

2.5.2 Physical control

Mealybugs can be controlled by brushing or picking them from affected plants but this is practicable when infestation is low. Pruning and destruction of affected plant parts are particularly useful at the initial stage of infestation. A steady stream of water at a reasonably high pressure can be sprayed on heavily infested plants to knock off the mealybugs. Once on the ground, the fallen ones will be available to ground predators (Gullan, 2000).

2.5.3 Improvement of soil fertility

Healthy plants are able to withstand mealybug attack and improvement of soil fertility can enhance biological control activity as shown in the cassava mealybug (Schulthess *et al*., 1997). It was observed that cassava grown in poor soils (pure sand and no mulch cover) had high mealybug infestations even after the release of the parasitic wasp, *Apoanagyrus lopezi* (De Santis)**.** In experiments conducted in Benin, the use of manure or other fertilizers resulted in a reduction in the cassava mealybug population (Schulthess *et al*., 1997). Improved plant nutrition resulted in the production of larger mealybugs, which in turn resulted in a higher proportion of female parasitic wasps with higher fertility levels. Mulch and fertilizer use enhanced the antibiotic properties of cassava against mealbug infestation (Schulthess *et al*., 1997).

2.5.4 Biological control

Generally, scale insects are often well controlled by natural enemies such as parasitic wasps, lady beetles, hover flies and lacewings, especially when predators and parasite activities are not disrupted by ants or applications of broad-spectrum insecticides such as carbaryl, malathion or pyrethroids (Gullan, 2000; Danne and Bentley, 2003).

There are excellent biological control agents for mealybugs, which attack above ground parts of plants. The most often used agent is the mealybug destroyer, *Cryptolaemus montrouzieri* Mulsant. The larvae of this lady beetle are very effective predators, especially when mealybug numbers are high. The lady beetle is most effective when there is a high population of mealybugs because of its high searching efficiency and they fly away from areas of low pest population density to high-density places and adults also feed on mealybugs. A parasitic wasp, *Leptomastix dactylopii* Howard, can be used in combination with the lady beetle (Okigbo, 2008).

2.5.5 Ants control to facilitate build-up of natural enemies

Effective control of seven genera of dominant ant species, *Oecophylla, Dolichoderus, Anoplolepis, Wasmannia, Azetca, Solenopsis* and *Formica* is a prerequisite for mealybug attacking above-ground plant parts since ants protect mealybug against natural enemies (Way and Khoo, 1992). Ueckermann (1998) found that the most effective means of ant control is to spray circularly the basal trunk of plants with chloropyrifos EC. This method of control is also environmentally friendly than chemical treatments on the soil. The purpose of this application method is to keep ants away from reaching the vines, but still allow them on the soil surface. Chloropyrifos EC is a registered pesticide against ants but for limited use only (Nel *et al.*, 1999). Chloropyrifos has also been tested at 41 ml/l in vineyards and gave reasonably good results (Ueckermann, 1998).

2.5.6 Prevention

One of the first methods of mealybug control is to use plants that are not infested with mealybugs for planting purposes. Commercial flower growers, sometimes, discard plants infested with mealybugs rather than to treat them with insecticide. Prevention is key in the management of mealybugs, because mealybug is very difficult to control once established. The prevention of the spread and establishment of mealybugs involves (i) inspecting newly procured plant materials, (ii) treating or removing infested plant hosts from premises and (iii) preventing water from infested areas to drain into clean areas, as crawlers can be transported in water (Hara *et al.*, 2001).

2.5.7 Cultural control

Management and control of weeds is one of the components in the management of mealybugs because various weeds may serve as hosts for mealybugs. A relationship has been found between the occurrence of weeds and mealybug problems in grape vines (Walton, 2000). Cover crops which are not hosts to mealybugs, may reduce population of the pest (Walton, 2000). Some plants reduce mealybug attack if they reduce ant population and if they help in the reduction of dusts especially, aerial parts attacting mealybugs (Walton, 2000). Weeds have to be controlled early in the season, since they act as access routes for ants and do not contribute much to the quality of the soil (Walton, 2000).

2.5.8 Hot water treatment

Hot water treatment, a type of physical control, is equally effective as submerging potted plants in insecticides. Submerging potted *Raphis* palm plants in water at 49 ^{0}C until the internal root ball temperature reaches 46 ^{0}C resulted in 100 per cent mortality of root mealybugs and did not significantly affect the potted palms (Hara *et al.*, 2001).

Drenching potted palms in hot water at 49 ^{0}C for 15 minutes not only controls mealybugs but also eliminates burrowing nematodes (Hara *et al.*, 2001).

Post harvest treatment of the flowers of red ginger, *Alpinia purpurata* (Vieill) K. Schum, in hot water at 49 ^{0}C for 12 to 15 minutes eliminated greater than 95 % of ants, banana aphids and mealybugs (Hara *et al.*, 1997; Hu *et al.*, 1996). Combination of hot water treatment with insecticides is used for quarantine security work to eliminate pests such as mealybugs, aphids, thrips, soft scales and ants (Hu *et al.*, 1996; Hara *et al.*, 1997). Conditioning infested flowers in hot air prior to hot-water immersion eliminated cotton aphids and ants and killed 99, 91 and 84 % of banana aphids, mealybug nymphs and adults, respectively (Hara e*t al*., 1997; Miller, 1999).

2.5.9 Chemical control

Historically, pesticides have been used a lot in vineyard mealybug control. Early programs involved the use of potassium cyanide, sodium cyanide, and sulfur fumigation, which gave way to chlorinated hydrocarbons such as Dichlorodiphenyltrichloroethane (i.e. DDT) and organophosphates (e.g. parathion) from the 1940s to the 1990s (Grimes and Cone, 1985). Most of these chlorinated hydrocarbons and organophosphates materials became less effective or were banned from use because of concerns on nontarget organisms (Flaherty *et al.*, 1982). Newer materials, with more novel modes of action, have also gained recognition, including botanicals, insect growth regulators, neonicotinoids, and biosynthesis inhibitors (Daane *et al.*, 2006; Sunitha *et al.*, 2009; Lo and Walker, 2010).

A major difference between the older and newer pesticides is the extend of coverage. For instance, a portion of the mealybug population is often under plant bark, and for some species, on vine roots. Many of the older foliar sprays did not effectively contact and kill

mealybugs in these more protected locations. Some of the more novel materials have systemic properties, applied either through the irrigation system or as a foliar spray (Srinivas *et al.*, 2007).

Another major difference is that the earlier pesticides were often broad spectrum and killed more than just the targeted mealybugs. Flaherty *et al.* (1982) found that extensive use of DDT and other synthetic insecticides used to control leafhoppers apparently disrupted natural control of grape mealybug *Pseudococcus maritimus* (Ehrhorn). Other researchers have since discussed the impact of broad spectrum insecticides on mealybug natural enemies (Mani and Thontadarya, 1988; Satyanarayana *et al.*, 1991; Walton and Pringle 2001; Mgocheki and Addison, 2009).

Aheer *et al.* (2009) indicated that the insecticides profenofos, methidathion, chlorpyrifos and methomyl were effective against citrus mealybug after seven days post- treatment. Lo *et al.* (2009) showed that a single application of profenofos was more effective than a single application of buprofezin (ApplaudTM). Mansour *et al.* (2010) showed that imidacloprid, methidathion and spirotetramat were effective against mealybug but they affected other predator insects. Chemical insecticides can have adverse effects on the environment and human health and can induce insecticide resistance in the target pest species and kill beneficial insects (Mansour *et al.*, 2010).

2.5.10 Soap spray

Insecticidal soaps work best on soft-bodied insects such as aphids, mealybugs, spider mites, thrips and whiteflies (Hata *et al.*, 1996). The addition of plant oils can increase the effectiveness of soap for waxy insects such as mealybugs. However, beneficial insects such as predatory mites, green lacewing larvae and small parasitic wasps (*Encarsia*,

Trichogramma and *Aphidius* wasps) can be killed with soap (Hata *et al.,* 1996).

Soaps have low mammalian toxicity but can be mildly irritating to the skin or eyes. Insecticidal soaps are biodegradable, do not persist in the environment and they do not contain any organic solvents. It is less likely that resistance to insecticidal soaps will develop as quickly as to the traditional insecticides. Resistance within the insects tends to develop more quickly with materials that have a very specific mode of action. Insecticidal soaps can be used in rotation with other insecticides such as malathion or nicotine, or systemic insecticides such as dimethoate or formathion with more specific modes of action to help slow the development of resistance (Hata *et al.,* 1996).

2.5.11 Oils

Oils such as vegetable oils (e.g. rape and neem oil) and mineral oils are useful for controlling mealybugs (Okigbo, 2008). Good spray coverage and good timing are important when using soapy solutions and oils but they must come into contact with the mealybugs to be effective. Crawlers are the easiest to kill, since they are more susceptible and are more exposed than eggs, older nymphs and adults. As they grow, the wax covering their bodies becomes thicker, rendering them more resistant to insecticides (Okigbo, 2008). Oil sprays suffocate the insects and can aid in controlling crawlers and eggs, while soap sprays cause the insects cell membranes to rupture effectively causing it to desiccate (Okigbo, 2008).

2.5.12 Mode of action of soaps, detergent and oils

The mode of action of soaps, detergents and oils remains uncertain, but may involve removal of insect cuticle wax, physical action, repellence or cell membrane disruption (Hodgson and Kuhr, 1990; Larew and Locke, 1990). Soaps have been reported to exhibit

insecticidal properties. Most of the research has been conducted on soft-bodied arthropods such as whiteflies (Butler *et al.,* 1993; Javed and Matthews, 2002), aphids (Pinnock *et al.*,1974; Fournier and Brodeur, 2000), scales (Riehl and Carman, 1953), mites (Osborne, 1984), thrips (Oetting and Latimer, 1995) and mealybugs (Lindquist, 1981). Oils extracted from plants have been extensively used in tropical countries for crop protection (Righi-Assia *et al.,* 2010). It is well known that oils, both mineral and vegetable can kill insects (Martin and Woodcock, 1983), but they depend upon physical contact and have no residual activity (Johnson, 1980).

2.6.0 Bioassay of chemicals

Bioassays are widely used in entomology to determine (i) development of resistance in insects to insecticides (ii) the amount of an insecticide residue in plant or animal tissue and (iii) screening of chemicals for insecticidal activity (Hoskin and Craig, 1962).

Duration of exposure is important in bioassay, therefore according to Hoskins and Craig (1962) exposure time should be carefully chosen so that the effect of the poison or stimulus is not influenced by factors such as natural mortality or physiological changes due to starvation.

2.6.1 Statistical procedures in bioassay

Except for rough approximation, the assumption of linearity over any considerable segment of the log dose response curve is untenable so Gaddum (1933) proposed a unit based on the standard deviation of this curve, known as a normal equivalent deviation (NED). A NED is the response increment brought about by increasing or decreasing the log-dose by one standard deviation taking the LD_{50} as the mid-point. Even more convenient than the NED are units known as probits. A probit is identical to NED except that zero NED is defined as five probit thus, eliminating negative values (Bliss, 1935).

When the logarithm of dosage is plotted against per centage kill in probit, a straight line is obtained and this is usually referred to as the log dosage probit line (ld-p) line. The slope of the regression line of probit on log-dose is a direct measure of the standard deviation (σ) of logarithms of individual effective doses (doses just effective on the individual responsive units). The true slope is equal to σ^{-1} when the data is plotted on these transformed co-ordinates or probits (Goldstein, 1964).

2.6.2 Mathematical procedure for the determination of the probit regression (ld-p) line

When experimental data on the relation between dose and mortality have been obtained, either a graphical or an arithmetical process is used to estimate the parameters. Both utilise the probit transformation. The graphical approach is rapid and sufficiently good for many purposes but when an accurate assessment of the precision of estimate is needed, a more detailed arithmetical analysis is necessary. The relation between the probit of the expected proportion of responses and the dose is given by the line and equation $Y=5+1/\sigma(x-\mu)$, where, x is the logarithm of the dose, μ the mean and σ the standard deviation. The equation $Y=5+1/\sigma(x-\mu)$ is commonly written as $Y=A+Bx$, where, A and B become the new parameters in place of μ and σ (Finney, 1971). The probits are then plotted against the logarithm of the dose and a straight line (best fit) is drawn to fit the probits. The line is a graphical approximation to the regression line of response probit on x, from the equation $Y=A+Bx$.

The log LD_{50} is estimated from the line as 'm', the dose at which Y=5. The slope of the line b is an estimate of $\sigma^{-1}$and is obtained as the increase in Y for a unit increase in x. These two estimates are then substituted for the parameters in the equation $Y=5+(x-\mu)$ to give the estimated relation between dose and response. The position on log dosage probit line

signifies the LD_{50}, while the slope represents the variability in susceptibility of the test population (Hoskins and Gordon, 1956). The slope of the ld-p line is a measure of the heterogeneity of an insect population towards a particular toxicant (Bliss, 1935). Flat slopes indicate great heterogeneity of a population to a particular toxicant, whereas a steep slope indicates a more homogenous population with reference to a particular toxicant and the population has a small variation in dosage over the response intervals.

An ld-p line with a low slope value is associated with relative resistance of the test insects to the toxicant. The flatter the ld-p line the greater the change in dosage needed to cause a chosen change in effect. The slope is therefore a measure of the average sensitivity of the test population.

2.6.3 Correction for mortality due to natural cause

In bioassay, some of the test mealybugs may die through natural causes even when they have not been exposed to any toxicant. The level of natural mortality may be estimated from mortalities occurring in the control treatment. If mortality in the control treatment is appreciable, this will affect the precision of the results. A correction is usually applied using the Abbott's formula (Abbott, 1925), $P_T = P_O - P_C \times 100/100 - P_C$, where P_T = % corrected mortality, P_o = observed % mortality and Pc = % mortality in the control.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1.0 Identification of mealybugs

For accurate identification of the mealybugs infesting yam (*Dioscorea* spp.) in storage in

Ghana, the mealybug samples were packed and shipped to Dr. Gillian W. Watson, a Senior Insect Biosystematist at Plant Pest Diagnostic Center, California, Department of Food and Agriculture, USA, for identification since there was no requisite technical know-how and expertise in Ghana to identify the mealybug to the species level.

The mealybugs on the yam tubers were probed repeatedly for them to retract their mouthparts before being picked up with a standard aspirator. A number of the mealybugs at various stages of development were picked up, placed in 80 % ethanol in vials, sealed and the samples placed in a dish of boiled water for 20 minutes to denature the enzymes and prevent the blackening of the specimens.

The specimens were kept in alcohol for five days before they were transferred into smaller vials containing small amount of fresh alcohol (5 ml). The samples were then packed and shipped.

3.2.0 Yam varieties used for the study

Four varieties of *D. rotundata* (Pona, Labreko, Dente and Muchumudu) and one of *D. alata* (Matches) were selected based on consumer preference for the studies. These varieties were randomly selected from improved yam barns in Ejura in Ejura-Sekyedumase District a predominately yam growing area in the Ashanti Region and were maintained in the screen house of the Faculty of Agriculture, KNUST, Kumasi.

3.3.0 Laboratory culture of *P. citri*

Planococcus citri adults were collected from yam barns and introduced onto tubers of *D. rotundata* var. Dokoba to ensure that mealybugs used for subsequent experiments developed from a common food source (standardization of mealybugs). Three tubers of *D. rotundata* var. Dokoba were placed in 30 x 30 x 60 cm wire mesh cage (Plate 3.1) with five

replications and the mealybugs allowed to oviposit for 72 h after which the mealybugs were removed from the tubers with a standard aspirator leaving the egg sacs

'*in situ'*.

The eggs were allowed to develop into adults and produced ovisacs. Fifty eggs were collected using a paintbrush (No.000) (American Painter 4000, Loew-Cornell Inc. Englewood Cliffs, NJ) and placed on the head region of each of the five test yam varieties (*D. rotundata* var. Pona, *D. rotundata* var. Labreko, *D. rotundata* var. Dente, *D. rotundata* var. Muchumudu and *D. alata* var. Matches) in 30 x 30 x 60 cm wire mesh cage with five replications. The eggs were allowed to develop on the test varieties into adults and the resulting eggs from them were used for the experiment. This was to ensure that any changes in behaviour associated with the change in host varieties were eliminated (Dobie, 1974).

Plate 3.1. 30 x 30 x 60 cm wire mesh cage for mealybug rearing.

3.4.0 Infestation of test varieties

The test yam tubers were infested with 24-hour old mealybug ovisacs. The ovisacs were teased open with blunt probes under a stereo microscope (x15) and 10 eggs were counted and refolded into the cottony ovisac and placed on the head region of each of the five test yam varieties (Pona, Labreko, Dente, Muchumudu and Matches) in the 30 x 30 x 60 cm wire mesh cage. Each 30 x 30 x 60 cm wire mesh cage had three tubers of each variety with five replication.

3.5.0 Determination of incubation period and per cent egg hatch of *P. citri*

Ten 24-hour old eggs wrapped in ovisacs were separately placed on the head region of three tubers of each yam variety in 30 x 30 x 60 cm wire mesh cage with five replications and observed after every 12 h for hatching and the mean incubation period and per cent egg hatched determined.

3.6.0 Determination of number of instars and the effect of yam variety on the developmental periods of *P. citri*

Ten 24-hour old eggs wrapped in ovisacs were separately placed on the head region of three tubers of each yam variety in 30 x 30 x 60 cm wire mesh cage with five replications. These eggs were allowed to hatch, the developmental period (duration of time from hatch to adult emergence) of the various instars of *P. citri* were observed daily, the presence of exuviae was used to identify each instar stage. Daily monitoring continued until maturity. On the death of an insect, mealybug of equal age was introduced from the test varieties used for reserve rearing.

3.7.0 Determination of longevity of adult *P. citri*

Longevity was determined for both sexes. For adult female longevity, 10 pre-ovipositing females from each yam variety were placed on the head region of three tubers of each yam variety (Pona, Labreko, Dente, Muchumudu and Matches) in the 30 x 30 x 60 cm wire mesh cage with five replications. The set-ups were observed until all the insects died and longevity recorded. A similar experiment was performed for the adult male mealybug. Longevity was taken as the period between adult emergence and death.

3.8.0 Mode of reproduction and total number of eggs laid by *P. citri*

The possibility of sexual and asexual reproduction of the mealybug was tested on each test yam variety. Forty mealybug females were collected immediately after moulting into adults to ensure their virginity. Twenty of the virgin females were isolated without males while the other 20 females were caged individually with males. Each female in the mating treatment was paired with three newly emerged adult males to increase the chances of mating. The pre-oviposition period defined as the duration between adult moult and first day of egg laying, were recorded for all the females.

The onset of oviposition was determined by the presence of an elongated white cottony egg sac extending beneath and behind the female. During the oviposition period, freshly laid eggs in ovisac were removed daily with paintbrush (No.000) and counted using stereo microscope (x15), until all the females died. The duration of oviposition was determined as the period between the start and the end of egg production. At the end of egg production, all female mealybugs that did not respond (no contractions of abdomen and movements of limbs) to probing by pin under a hand lens were considered dead. Other parameters such as the post-oviposition period and total number of eggs laid were also determined.

3.9.0 Determination of per cent survival of *P. citri*

Ten 24-hour old eggs wrapped in ovisacs were separately placed on the head region of three tubers of each yam variety (Pona, Labreko, Dente, Muchumudu and Matches) in 30 x 30 x 60 cm wire mesh cage with five replications. These eggs were allowed to hatch and the larvae allowed to develop into adults. The per cent survival of each instar was recorded by counting the number of individuals that had successfully moulted to the next instar using hand lens. The overall per cent survival from egg to adult female or male on each yam variety was based on the number of adult females (or males) per the total number of eggs.

3.10.0 Determination of *P. citri* offspring sex ratio on the yam varieties

The population for determining sex ratio was started with ten 24-hour old eggs wrapped in ovisacs separately placed on the head region of three tubers of each yam variety (Pona, Labreko, Dente, Muchumudu and Matches) in 30 x 30 x 60 cm wire mesh cage with five replications. The eggs were allowed to hatch and develop into adults. The adult males (a tiny two-winged gnat-like insect) and females (wingless eight-segmented antennae insect) emerging from the eggs laid on each yam variety by the adults were counted with the aid of a hand lens and the offspring sex ratio determined on all the varieties tested.

The proportion of females was used as a measure of sex ratio and was determined at the end of the experiment by dividing the number of adult females successfully emerged by the total number of surviving adults (progeny) on each yam variety (Godfray, 1994; Van Alphen and Jervis, 1996).

3.11.0 Susceptibility of the yam varieties to *P. citri* infestation

Susceptibility of the yam varieties to the mealybug infestation was assessed on the basis of growth index of the mealybugs using the formula Log n/Av (Howe, 1971). Where, n is the per cent adult survival and Av is the average developmental period.

3.12.0 Experimental design and data analyses

The experimental design used for the biology of *P. citri* was completely randomized design with five treatments (varieties) and five replications. The data were subjected to analysis of variance (ANOVA) using the statistical Package GenStat (GenStat, 2008) software.

3.13.0 Experimental site and condition

All the experiments were conducted in the Insect Laboratory of Faculty of Agriculture, KNUST, Kumasi in the Ashanti Region of Ghana at a temperature of 26-30 ^{0}C and relative humidity of 70-75 %.

3.14.0 Characteristics of chemicals used in the bioassay

3.14.1 Cypermethrin

Cypermethrin is a synthetic pyrethroid insecticide which acts as a stomach and contact insecticide; it acts as a fast-acting neurotoxin in insects. It is easily degraded on soil and plants and can be effective for weeks when applied to indoor inert surfaces. Exposure to sunlight, water and oxygen accelerates its decomposition (Wothing, 1987; Anonymous, 1995).

Its structure is based on pyrethrum, a natural insecticide which is contained in chrysanthemum flowers but it has a higher biological activity and is more stable than its natural model. The molecular formula is $C_{22}H_{19}Cl_2NO_3$ with a molecular weight of

416.32 IUPAC: (RS)-ý-cyano-3-phenoxybenzyl (1RS)-cis, trans-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate (Wothing, 1987; Anonymous, 1995).

3.14.2 Physical chemistry of Cypermethrin

Pure isomers of cypermethrin form colourless crystals. When mixed isomers are present, cypermethrin is a viscous semi-solid or a viscous yellow liquid. It has water solubility of 0.01 mg/l at 20 ^{0}C. The pure isomer has a melting point of 60-80 ^{0}C and vapour pressure of 5.1 x 10^{-7} nPa at 70 ^{0}C, partition coefficient of 6.602 and adsorption coefficient of 100,000.

Trade and other names include Ammo, Arrivo, Barricade, Basathrin, CCN52, Cymbush, Cymperator, Cynoff, Cypercopal, Cyperguard 25EC, Cyperhard Tech, Cyperkill, Cypermar, Demon, Flectron, Fligene CI, Folcord, Kafil Super, NRDC 149, Polytrin, PP 383, Ripcord, Siperin and Stockade (Wothing, 1987; Anonymous, 1995).

3.14.3 Formulation and uses of Cypermethrin

Technical cypermethrin is a mixture of eight different isomers, each of which may have its own chemical and biological properties. Cypermethrin is photo-stable. It is available as an emulsifiable concentrate or wettable powder (Wothing, 1987; Anonymous, 1995). Cypermethrin (a product of Hockley International Limited, United Kingdom) has wide uses in cotton, cereals, vegetables and fruit pest management and for food storage in public health and in animal husbandry (Wothing, 1987; Anonymous, 1995).

3.14.4 Imidacloprid

Imidacloprid is a neonicotinoid, which is a class of neuro-active insecticides modeled after nicotine. A patented chemical, imidacloprid is manufactured by Bayer Cropscience (part of Bayer AG) and sold under the trade names Kohinor, Admire, Advantage, Gaucho, Merit, Confidor, Hachikusan, Premise, Prothor and Winner. It is marketed for pest control, seed treatment, an insecticide spray, termite control, flea control and a systemic insecticide (Boyd and Boethel, 1998; Caroline, 2001).

Imidacloprid based insecticide formulations are available as dustable powder, granular, seed dressing (flowable slurry concentrate), soluble concentrate, suspension concentrate, and wettable powder (Boyd and Boethel, 1998; Caroline, 2001). Typical application rates

range from 0.056 - 0.140 kg/ha. These application rates are considerably lower than older traditionally used insecticides (Boyd and Boethel, 1998; Caroline, 2001).

3.14.5 Physical properties of Imidacloprid

Imidacloprid has a colourless crystal appearance with a weak characteristic odour with the chemical name 1-[(6-chloro-3-pyridinyl) methyl]-N-nitro-2-imidazolidinimine. It has a molecular weight of 255.7 and water solubility of 0.51 g/l at 200 ^{0}C. Its crystal form has a melting point of 136.4-143.8 ^{0}C (Boyd and Boethel, 1998; Caroline, 2001).

3.14.6 Uses of Imidacloprid

Imidacloprid (a product of Zhechem Ent. China) works by blocking the elements of the insect nervous system, which are more susceptible to the toxic effects of imidacloprid than those of homoitherms (Boyd and Boethel, 1998; Caroline, 2001). Imidacloprid has a wide range of uses; it is used to control sucking insects such as leaf and plant hoppers of rice, aphids, thrips and whitefly. It is also effective against termites and some species of biting insects, such as rice weevil and Colorado beetle and has no effect on nematodes or spider mites (Boyd and Boethel, 1998; Caroline, 2001).

It can be used as seed dressing, soil treatment and foliar treatment in different crops including rice, cotton, cereals, maize, sugar beet, potatoes, vegetables, citrus fruit, apples, pears and stone fruit. Worldwide, it is considered one of the insecticides used in the largest volume (Boyd and Boethel, 1998; Caroline, 2001).

3.14.7 Groundnut oil

Groundnut oil is derived from the seeds of groundnuts (*Arachis hypogaea* L.) it has a high smoke point relative to many other cooking oils. A 100 g groundnut oil contains

17.7 g of saturated fat, 48.3 g of monounsaturated fat and 33.4 g of polyunsaturated fat (Shewfelt and Young, 1977; Passera, 1981; Hariod, 1990). Its major component fatty acids are oleic acid (46.8 % as olein), linoleic acid (33.4 % as linolein) and palmitic acid (10.0 % as palmitin). The oil also contains some stearic acid, arachidic acid, arachidonic acid, behenic acid, lignoceric acid and other fatty acids (Shewfelt and Young, 1977; Passera, 1981; Hariod, 1990). Groundnut oil is the main ingredient in some earwax removing products. It is also used as a faecal softener and most commonly used when frying foods, particularly french fries and chicken. Groundnut oil can also be used to make soap in a process called saponification. The soap produced is soft and stable
(Shewfelt and Young, 1977; Passera, 1981; Hariod, 1990).

3.14.8 Soybean oil

Soybean oil (a product of Ghana Nuts Ltd. Techiman, Ghana) is extracted from the seeds of the soybean (*Glycine max* (L.) Merr.,). It is one of the most widely consumed cooking oils. A 100 g soybean oil has 16 g of saturated fat, 23 g of monounsaturated fat and 58 g of polyunsaturated fat (Poth, 2002). The major unsaturated fatty acids in soybean oil, triglycerides are 7–10 % alpha-linolenic acid, 51 % linoleic acid and 23 % oleic acid. It also contains saturated fatty acids, 4 % stearic acid and 10 % palmitic acid. Soybean oil is mostly used for frying, baking and as a condiment for salads. As a drying oil, processed soybean oil is also used as a base for printing inks (soy ink) and oil paints (Poth, 2002).

3.14.9 Sunlight^{TM} dishwashing detergent

Sunlight^{TM} dishwashing detergent is a product of Unilever Ghana Limited and contains among other ingredients 20-40 % anionic detergent and less than 15 % solubilisers with colourant and fragrance (Vagas, 2008).

3.14.10 Key soap

Key soap is a product of Unilever Ghana Limited and is obtained by treating vegetable oil with a strongly alkaline solution. It is naturally scented, anti-bacterial and mildly antiseptic soap and a very traditional type of soap that is recommended for acne and makes a great deodorant bar (Vagas, 2008).

3.15.0 Stages of *P. citri* used for chemical bioassay test

The most waxy, bigger and destructive stages of *P. citri* (third instar larvae and adult females) were used for the bioassay tests to determine susceptibility to the chemicals Cypermethrin, Imidacloprid, Soyabean oil emulsion, Groundnut oil emulsion, Sunlight™ detergent solution and Key soap solution.

Using hand lens, third-instar larvae were recognized readily by their yellowish body colour and seven segmented antennae. The adult female *P. citri* is reddish yellow in colour, has eight-segmented antennae, wingless, covered with white-cottony wax and has a fringe of elongated waxy filaments around the periphery of the body.

3.16.0 Preparation of chemical solutions

The required concentrations of the chemicals were prepared using the formula v = ck/p (Gruzdyev *et al.*, 1988), where v = volume (ml) of chemical in k, c = required concentration, k = required volume of solution in (ml) and p = per cent active ingredient in chemical formulation.

Distilled water was added to make up the required volume k. Preliminary tests were carried out for all the chemicals to determine the range of concentrations required for the actual tests. Four concentrations were used i.e. (0.01, 0.02, 0.03 and 0.04) % for

Cypermethrin, (0.01, 0.02, 0.03 and 0.04) % for Imidacloprid, (0.125, 0.25, 0.50 and 1.00) % for Groundnut oil, (0.125, 0.25, 0.50 and 1.00) % for Soyabean oil, (0.125, 0.25, 0.50 and 1.00) % for Sunlight™ detergent and (0.125, 0.25, 0.50 and 1.00) % for Key Soap.

3.17.0 Preparation of the vegetable oil emulsion

The groundnut and soybean oil emulsions were prepared by dissolving 0.125, 0.25, 0.50 and 1.00 % respectively of the oil in 0.50 % Sunlight™ detergent solution (Sunlight™ detergent acted as an emulsifier) and vigorously shaken**.**

3.18.0 Preparation of the Key soap solution

A bar of Keysoap was grated into flakes (10 g) and boiled whiles stirring until the soap dissolved in a litre of water and left to cool overnight from which the various Key soap solution concentrations were prepared.

3.19.0 Exposure of third instar larvae and adult female *P. citri* to chemicals

Bioassays were conducted using the topical application method (Eric and Gfeller, 2001). Ten third instar larvae bred on *D. rotundata* var. Pona were placed in a glass Petri dish (9-cm) lined with moistened filter paper (Whatman, Clifton, NJ) with five replications, 10 microlitre volume of the chemicals were applied topically on the dorsum of each third instar larva of *P. citri* using a micropipette. A similar procedure was used for the control in which distilled water was used instead of a chemical. Mortality counts were made at 12, 24 and 48 h with the aid of a stereo microscope (x15). A larva was considered dead when there was absence of natural movement even after tactile stimulation with a pin. The LD_{50} and LD_{90} of each chemical were calculated from the per cent insect mortality by Probit analysis (Finney, 1971). A similar experiment was done for the adult females of *P.*

citri. Abbort's formula was not applied to the data because of the absence of deaths in the control treatments.

3.20.0 Control of *P. citri* on *D. rotundata* var. Pona in improved barns

A trial was conducted on *D. rotundata* var. Pona stored in improved yam barns since it was found to be more susceptible to *P. citri* infestation during this study. Five infested *D. rotundata* var. Pona tubers with various stages of *P. citri* were randomly selected and replicated five times i.e. twenty five (25) tubers for each chemical treatment. The tubers were arranged horizontally on the shelves of the improved barn. Using a small hand-held sprayer the infested tubers were sprayed using the LD_{90} value obtained for the adult *P. citri* 12 h exposure period i.e. (0.05 % Cypermethrin, 0.07 % Imidacloprid, 1.0 % Groundnut oil emulsion, 1.2 % Soyabean oil emulsion, 1.9 % Sunlight detergent solution and 2.5 % Key soap solution) respectively, since these were the highest dose that killed 90 per cent of the adult *P. citri* after 12 h exposure during the laboratory bioassay. With the aid of magnifying glass, mortality counts of *P. citri* on the tubers were recorded at 12, 24, 48 and 72 h after spraying them with each chemical.

3.21.0 Weekly chemical control of *P. citri* on *D. rotundata* var. Pona in improved barns

To determine the effect of weekly application of the chemicals on *P. citri* mortality on *D. rotundata* var. Pona in improved barns. The tubers were sprayed again with 0.05 % Cypermethrin, 0.07 % Imidacloprid, 1.0 % Groundnut oil emulsion, 1.2 % Soyabean oil emulsion, 1.9 % Sunlight™ detergent solution and 2.5 % Key soap solution and water (control) using a small hand-held sprayer on the seventh, fourteenth and twenty first day after the initial 24 h exposure of *P. citri* infested yams to the chemicals. With the aid of

magnifying glass, mortality count of *P. citri* on the tubers was recorded 24 h after each weekly spray.

3.22.0 Statistical analysis of chemical bioassay and Chi-square test

The chemical bioassay data were analysed by probit analysis (Finney, 1971) and a test of significance (Chi-square test) was calculated to determine whether the probit regression lines satisfactorily represented the data using SPSS (SPSS, 2007) statistical software. Significant differences were inferred by non-overlapping of 95 % fiducial limits. Data on per cent mortality of *P. citri* during weekly chemical application were arcsine transformed and analysed with GenStat software (GenStat, 2008), where single factor analysis of variance was calculated and means compared with least significant difference (LSD) test at the 5 % probability level. Significant differences were inferred by $P < 0.05$ and non-significant differences by $P > 0.05$.

CHAPTER FOUR

4.0 RESULTS

4.1.0 Identification of the mealybugs on the stored yams

The mealybugs on the stored yams were identified morphologically as *Planococcus citri* of the Class Insecta, Order Hemiptera, Suborder Sternorrhyncha, Superfamily Coccoidea, Family Pseudococcidae, Subfamily Pseudococcinae, Genus *Planococcus* and Species *citri* by Dr. Gillian W. Watson, a Senior Insect Biosystematist at Plant Pest Diagnostic Center California, Department of Food and Agriculture, USA.

4.2.0 Life stages of *P. citri*

The citrus mealybug (a short-tailed mealybug) is a soft-bodied pseudococcid. The female has three nymphal stages called instars with each larval stage separated by a moult. The immature or nymph resembles adult female in appearance except that it is smaller. The male *P. citri* has four nymphal stages during its development. The male nymph is more elongated or narrower and often occurs in a loose cocoon, approximately four days into the second instar, a black tinge that is easily discernible develops around the insect body. Two days later, the nymph starts spinning a cocoon around itself. This cocoon is continuously spun increasing in density until the winged adult mealybug is ready to emerge two moults later.

4.2.1 Eggs of *P. citri*

The eggs are oblong in shape, pale yellow in colour and are in clumps of five to 20 inside egg sacs composed of white cottony-waxy filaments (Plate 4.1).

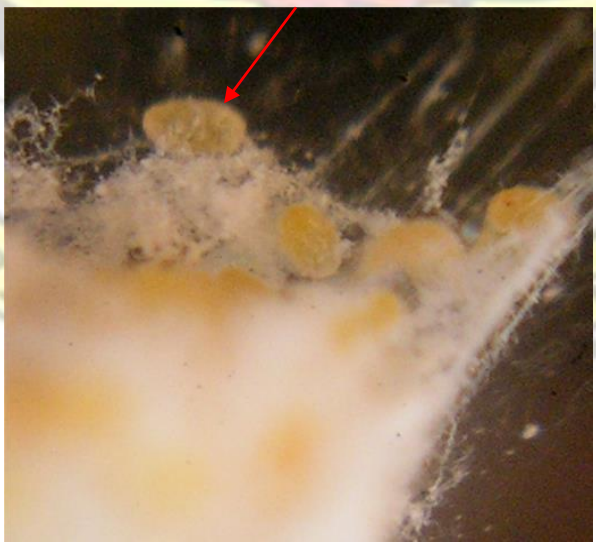

Plate 4.1. Egg of *P. citri*

4.2.2 First-instars of *P. citri*

The first-instars (crawlers) are very active, oval in shape, yellow in colour with six segmented antennae and lack waxy coating on the body and are often found congregated upon the egg sac (Plate 4. 2).

Plate 4.2. First-instar (crawler) of *P. citri*

4.2.3 Second-instar larva of *P. citri*

The second-instar larva is bigger than the first instar, oval in shape, yellow in colour and has six segmented antennae (Plate 4. 3).

Plate 4.3. Second-instar larva of *P. citri*

4.2.4 Third -instar larva of *P. citri*

The third-instar larva resembles the larger adult female and is recognized readily by its yellowish body colour, seven segmented antennae and its larger anal ring (Plate 4. 4).

Plate 4.4. Third-instar larva of *P. citri*

4.2.5 Adult female of *P. citri*

The adult *P. citri* female has an elongated oval body about 3 mm long and 1.5 mm in width, flat with 18 pairs of short-waxy filaments around the margin of the body 17 of which are equal and the anal one slightly longer (no more than double the length of the others). The body colour is mainly reddish yellow, covered with powdery wax through which distinguished transverse lines of segmentation appear. It is wingless and has eightsegmented antennae (Plate 4. 5).

Plate 4.5. Female adult *P. citri*

4.2.6 Adult male of *P. citri*

The adult *P. citri* male is a tiny two-winged gnat-like insect. It has an elongated body, reddish in colour and has two long filaments at the rear end (Plate 4. 6). It is a short-lived insect.

Plate 4.6. Adult Male *P. citri*

4.3.0 Effect of yam variety on incubation period of *P. citri*

The mean incubation periods of eggs of *P. citri* laid on the five yam varieties are presented in Table 4.1.

Table 4.1. Mean incubation periods of eggs of *P. citri* laid on tubers of five stored yam varieties

	Mean incubation period in days ± s.e	
Yam varieties	Male egg	Female egg
D. rotundata var. Pona	10.5 ± 0.3	10.8 ± 0.2

D. rotundata var. Labreko	10.7 ± 0.2	11.0 ± 0.2
D. rotundata var. Muchumudu	10.8 ± 0.3	11.1 ± 0.3
D. alata var. Matches	11.0 ± 0.2	11.2 ± 0.2
D. rotundata var. Dente	11.2 ± 0.2	11.4 ± 0.2
LSD (5 %)	0.7	0.6
CV (%)	5.0	4.7

The mean incubation periods of male eggs (eggs that developed into males) on *D. rotundata* var Pona, *D. rotundata* var. Labreko, *D. rotundata* var. Muchumudu, *D. alata* var. Matches and *D. rotundata* var. Dente ranged from 10.5 to 11.2 days (Table 4.1). The longest mean incubation period of male egg was recorded on *D. rotundata* var. Dente and the shortest on *D. rotundata* var. Pona. The differences between the mean incubation periods of *P. citri* male eggs on the yam varieties, were not significant (Table 4.1).

The mean incubation periods of female eggs (eggs that developed into females) on Pona, Labreko, Muchumudu, Matches and Dente ranged from 10.8 to 11.4 days (Table 4.1). The longest mean incubation period of female egg was recorded on Dente and the shortest on Pona. The differences between the mean incubation periods of *P. citri* female eggs on the yam varieties, were not significant.

4.4.0 Effect of yam variety on per cent egg hatch of *P. citri*

The mean per cent egg hatch of *P. citri* on each yam variety is shown in Table 4.2. The mean per cent egg hatch on Pona, Labreko, Muchumudu, Matches and Dente was between 95.3 % and 90.7 % (Table 4.2). The greatest egg hatch was observed on Pona and lowest on Matches (Table 4.2). The differences between the mean per cent egg hatch on the yam varieties, were not significant (Table 4.2).

Table 4.2. Mean per cent egg hatch of *P. citri* on tubers of five stored yam varieties

Yam varieties	Mean per cent *P. citri* egg hatch ± s.e
D. rotundata var. Pona	95.3 ± 2.5
D. rotundata var. Labreko	93.3 ± 3.2
D. rotundata var. Muchumudu	92.0 ± 2.7
D. alata var. Matches	90.7 ± 3.2
D. rotundata var. Dente	92.7 ± 1.9
LSD (5 %)	2.7
CV (%)	3.3

4.5.0 Developmental period of *P. citri* female bred on the different yam varieties

The mean developmental periods (in days) of *P. citri* female reared on each yam variety are shown in Table 4.3. Developmental period of *P. citri* female first instar on the yam varieties Pona, Labreko, Muchumudu, Matches and Dente ranged from 7.9 days to 12.8 days (Table 4.3).

Table 4.3. Mean developmental period of *P. citri* female bred on tubers of the yam varieties

Yam Varieties	DURATION OF LARVAL STAGES IN DAYS ± s.e			
	Instar I	Instar II	Instar III	Total developmental period
D. rotundata var. Pona	7.9 ± 0.2	6.9 ± 0.1	9.6 ± 0.2	24.4 ± 0.2
D. rotundata var. Labreko	9. 6 ± 0.2	7.3 ± 0.1	9.9 ± 0.1	26.7± 0.2

D. rotundata var. Muchumudu	11.1 ± 0.2	9.6± 0.2	11.2 ± 0.2	31.9 ± 0.5
D. alata var. Matches	11.2 ± 0.2	9.8 ± 0.1	11.6 ± 0.2	32.7 ± 0.3
D. rotundata var. Dente	12.8 ± 0.3	11.2 ± 0.1	12.9 ± 0.1	37.0 ± 0.5
LSD (5 %)	0.7	0.3	0.5	1.1
CV (%)	4.8	3.2	3.1	2.6

Developmental period of the first instar was longest on Dente (12.8 days) and the shortest was on Pona (7.9 days). The differences between the yam varieties were significant except between Muchumudu and Matches which did not differ (Table 4.3). The mean developmental period of *P. citri* second instar female on the yam varieties ranged from 6.9 to 11.2 days (Table 4.3). The longest mean developmental period of second instar female was recorded on Dente and the shortest on Pona. The differences between the mean developmental periods of second instar female on the yam varieties were significant except between Muchumudu (9.6 days) and Matches (9.8 days) which did not differ (Table 4.3).

The mean developmental period of *P. citri* third instar female on the yam varieties ranged from 9.6 to 12.9 days (Table 4.3). The longest mean development period of third instar female was recorded on Dente and the shortest on Pona. The differences between the mean third instar female duration on Dente and the other yam varieties were significant. The difference between the mean third instar female duration on Muchumudu and Matches was not significant but differed significantly from the rest of the varieties (Table 4.3). The difference between the mean third instar female duration on Pona and Labreko was not significant but differed significantly from the other varieties (Table 4.3). The total

P. citri female larval developmental period on the yam varieties followed the same trend as observed on the first and second *P. citri* female instar duration.

4.6.0 Effect of yam variety on mode of reproduction

It was observed that reproduction was sexual. A cohort of 20 virgin females isolated and caged without males did not lay eggs. The 20 virgin females that were paired with three newly emerged adult males to increase the chance of mating laid eggs in ovisacs (Table 4.4).

The mean number of eggs laid by *P. citri* on the yam varieties ranged from 257.0 to 497.0 eggs (Table 4.4). The largest mean number of eggs was laid on Pona and the smallest on Dente. The differences between the mean numbers of eggs laid on all the yam varieties were significant (Table 4.4).

Table 4.4. Mean number of eggs laid by virgin and non-virgin *P. citri* females bred on tubers of five yam varieties

Yam varieties	Mean number of eggs laid ± s.e	
	Virgin females without males	Virgin females with males
D. rotundata var. Pona	0.0	497.0 ± 5.5
D. rotundata var. Labreko	0.0	402.8 ± 5.4
D. rotundata var. Muchumudu	0.0	373.0 ± 7.1
D. alata var. Matches	0.0	292.0 ± 7.2

D. rotundata var. Dente	0.0	257.0 ± 6.8
LSD (5 %)	0.0	19.0
CV (%)	0.0	4.0

4.7.0 Effect of yam variety on oviposition and adult longevity of female *P. citri*

The effects of yam variety on the duration of pre-oviposition, oviposition, postoviposition and longevity of *P. citri* female are shown in Table 4.5. The mean preoviposition period on Pona, Labreko, Muchumudu, Matches and Dente ranged from 12.2 to 14.9 days (Table 4.5). The longest pre-oviposition period was recorded on Dente and the shortest on Pona.

The differences between the mean pre-oviposition period of *P. citri* female on Dente and the other yam varieties were significant. The difference between the mean pre-oviposition period on Muchumudu and Matches was not significant but differed significantly from the mean pre-oviposition period on Pona. The differences between the mean preoviposition period on Muchumudu and Labreko were not significant but differed significantly from the mean pre-oviposition period on Pona (Table 4.5).

Table 4.5. Mean pre-oviposition, oviposition, post-oviposition and adult longevity of female *P. citri* reared on tubers of five yam varieties

Yam Varieties	Duration of *P. citri* stages in days ± s.e.			
	Pre-oviposition Period	Oviposition Period	Post -oviposition	Mean adult longevity
D. rotundata var. Pona	12.2 ± 0.3	22.0 ± 0.2	3.9 ± 0.1	38.1 ± 0.3
D. rotundata var. Labreko	13.3 ± 0.3	20.2 ± 0.5	3.3 ± 0.1	36.8 ± 0.6

D. rotundata var. Muchumudu	13.6 ±0.2	18.4 ± 0.2	2.8 ± 0.1	34.9 ± 0.4
D. alata var. Matches	14.1 ± 0.2	17.9 ± 0.3	2.7 ± 0.1	34.6 ± 0.3
D. rotundata var. Dente	14.9 ± 0.3	16.1 ± 0.2	1.9 ± 0.1	32.9 ± 0.5
LSD (5 %)	0.7	0.8	0.2	1.3
CV (%)	4.0	3.2	4.1	2.8

The mean oviposition period on Pona, Labreko, Muchumudu, Matches and Dente ranged from 16.1 to 22.0 days (Table 4.5). The longest oviposition period was recorded on Pona and the shortest on Dente. The differences between the oviposition periods on the yam varieties were significant except between Muchumudu (18.4 days) and Matches (17.9 days) which did not differ (Table 4.5).

The mean post-oviposition period on Pona, Labreko, Muchumudu, Matches and Dente ranged from 1.9 to 3.9 days (Table 4.5). The longest post-oviposition period was recorded on Pona and the shortest on Dente. The differences between the post-oviposition periods on the yam varieties were significant except between Muchumudu and Matches which did not differ (Table 4.5).

The mean adult female longevity on Pona, Labreko, Muchumudu, Matches and Dente was between 32.9 and 38.1 days (Table 4.5). Pona recorded the longest mean adult female longevity and Dente the shortest. The differences between the mean adult female longevity on the yam varieties were significant except between Muchumudu and Matches which did not differ (Table 4.5).

4.8.0 Developmental period of *P. citri* male bred on tubers of five different yam varieties

The mean developmental periods (in days) of *P. citri* male reared on each yam variety are shown in Table 4.6. The developmental periods of the first instar male ranged from 7.6 to 11.5 days (Table 4.6). The developmental periods of the first instar male were longest on Dente (11.5 days) and shortest on Pona (7.6 days) (Table 4.6), the differences being significant except between Muchumudu and Matches which did not differ (Table 4.6).

Developmental periods of *P. citri* second instar male on Pona, Labreko, Muchumudu, Matches and Dente ranged from 8.7 to 12.5 days (Table 4.6). The periods of the second instar male was longest on Dente (12.5 days) and the shortest on Pona (8.7 days) (Table 4.6). The differences between the developmental periods of the second instar male *P. citri* on the yam varieties were significant except between Muchumudu and Matches which did not differ (Table 4.6).

The mean pre-pupa duration on Pona, Labreko, Muchumudu, Matches and Dente was between 2.3 and 3.3 days (Table 4.6). The longest pre-pupa duration was recorded on Dente and shortest on Pona (Table 4.6). The differences between the mean pre-pupa duration on Dente and the other yam varieties were significant. The difference between the mean pre-pupa duration on Muchumudu and Matches was not significant but differed significantly from the rest of the varieties. The difference between the mean pre-pupa duration on Pona and Labreko was not significant but differed significantly from the other varieties (Table 4.6).

The mean pupa duration on Pona, Labreko, Muchumudu, Matches and Dente was between 2.7 and 4.7 days (Table 4.6). The longest pupa duration was recorded on Dente and shortest

on Pona (Table 4.6). The differences between the mean pupa duration on Dente and the other yam varieties were significant. The difference between the mean pupa duration on Muchumudu and Matches was not significant but differed significantly from the rest of the varieties (Table 4.6). The difference between the mean pupa duration on Pona and Labreko was not significant but differed significantly from the other varieties (Table 4.6).

Table 4.6. Mean developmental period of *P. citri* male reared on tubers of five yam varieties

Yam varieties	Duration of *P. citri* Male Larval Stages In Days ± S.E.			
	Instar I	Instar II	Pre-pupa	Pupa
D. rotundata var. Pona	7.6 ± 0.2	8.7 ± 0.2	2.3 ± 0.1	2.7 ± 0.1
D. rotundata var. Labreko	8.0 ± 0.1	10.1 ± 0.4	2.4 ± 0.1	2.9 ± 0.1
D. rotundata var. Muchumudu	9.7 ± 0.2	11.6 ± 0.1	2.8 ± 0.1	3.9 ± 0.1
D. alata var. Matches	10.0 ± 0.2	11.8 ± 0.1	2.9 ± 0.1	4.0 ± 0.1
D. rotundata var. Dente	11.5 ± 0.1	12.5 ± 0.2	3.3 ± 0.1	4.7 ± 0.1
LSD (5 %)	0.4	0.5	0.2	0.2
CV (%)	3.5	3.4	4.3	3.5

4.9.0 Effect of yam variety on total larval developmental period of male *P. citri*

The mean total larval developmental periods of *P. citri* males reared on tubers of five yam varieties are shown in Figure 4.1. The mean total larval developmental period of *P. citri* males on Pona, Labreko, Muchumudu, Matches and Dente ranged from 21.3 to 32.0 days (Figure 4.1).

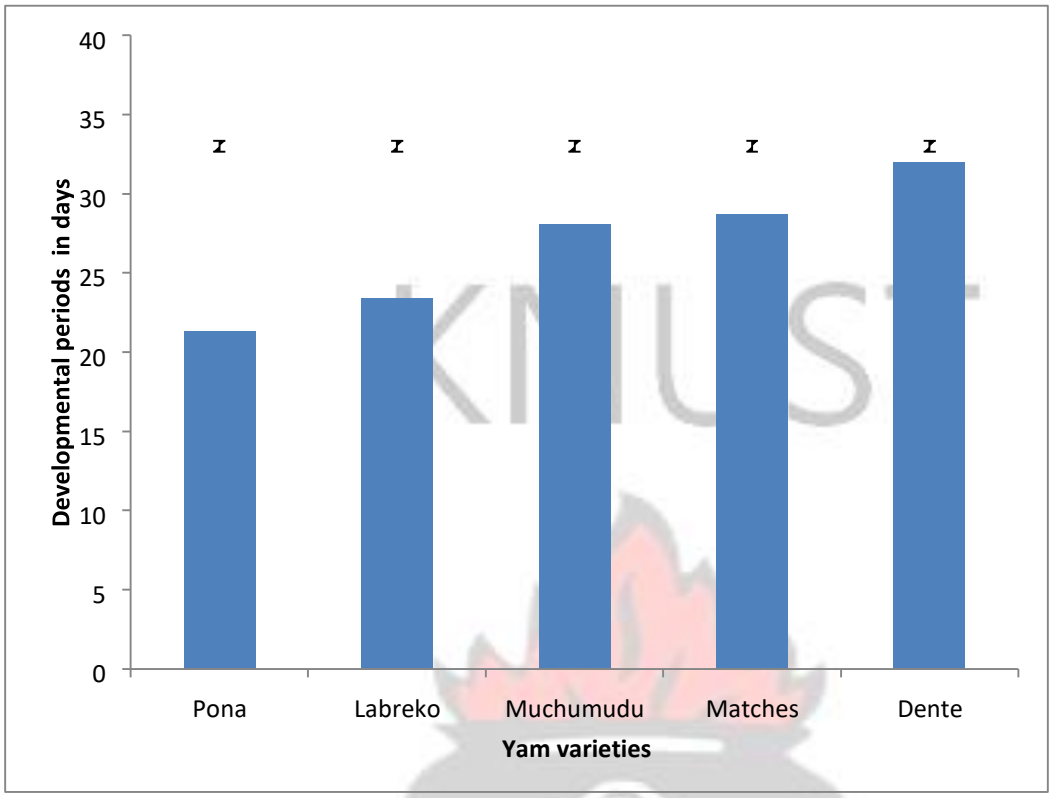

Figure 4.1 Mean total larval developmental periods of *P. citri* males reared on tubers of five yam varieties. *Bars on the graphs represent LSD values*

The mean male larval developmental period was longest on Dente (32.0 days) and the shortest on Pona (21.3 days) the differences between the mean total male larval developmental periods of *P. citri* on the yam varieties were significant except between Muchumudu (28.0 days) and Matches (28.7 days) which did not differ from each other (Figure 4.1).

4.10.0 Effect of yam variety on longevity of male adult *P. citri*

The mean longevity of adult *P. citri* males reared on tubers of five yam varieties are shown in Figure 4.2.

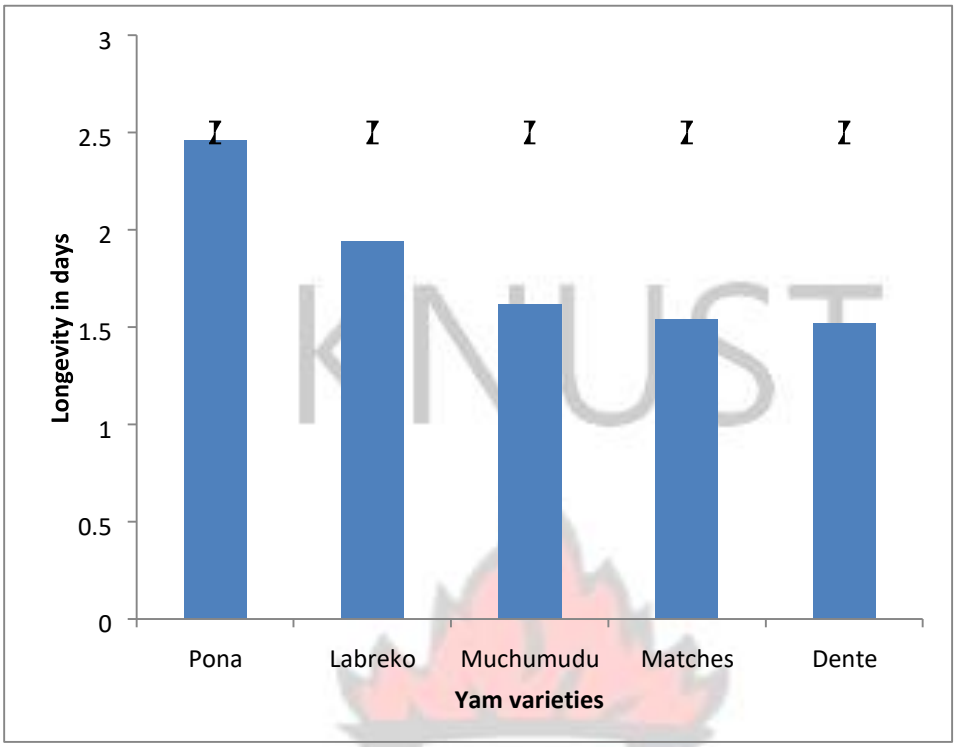

Figure 4.2 Mean longevity of adult *P. citri* males reared on tubers of five yam varieties

The mean longevity of adult *P. citri* males on Pona, Labreko, Muchumudu, Matches and Dente ranged from 1.52 to 2.46 days (Figure 4.2). The longest mean longevity was recorded on Pona and shortest on Dente. The differences between the mean longevity of *P. citri* adult male on Muchumudu, Matches and Dente were not significant but differed significantly from the mean longevity of *P. citri* adult male on Pona and Labreko which also differed significantly from each other (Figure 4.2).

4.11.0 Effect of yam variety on per cent survival of *P. citri* female

The mean per cent survival of *P. citri* females reared on tubers of five yam varieties are shown in Figure 4.3.

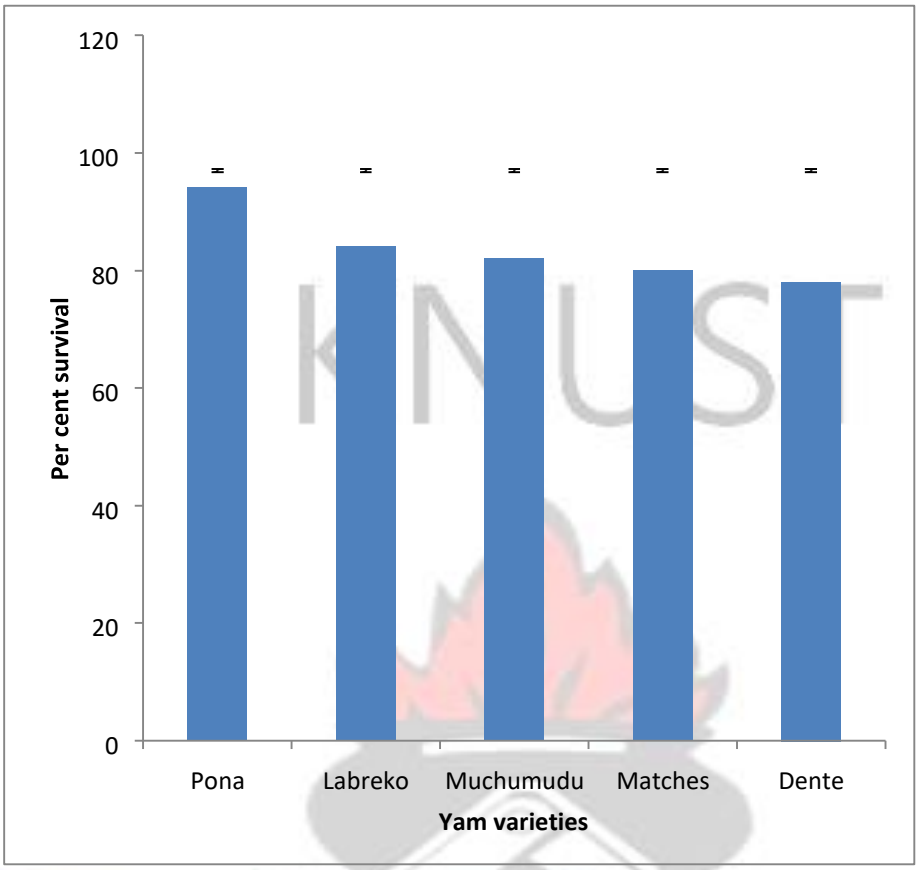

Figure 4.3 Mean per cent survival of *P. citri* females reared on tubers of five yam varieties

The mean per cent survival of female *P. citri* reared on Pona, Labreko, Muchumudu, Matches and Dente was between 78.0 % and 94.0 % (Figure 4.3). The highest mean per cent survival was observed on Pona and lowest on Dente. The differences between the mean per cent survival of *P. citri* female reared on all the yam varieties were significant (Figure 4.3).

4.12.0 Effect of yam variety on per cent survival of *P. citri* male

The mean per cent survival of *P. citri* males reared on tubers of five yam varieties are shown in Figure 4.4.

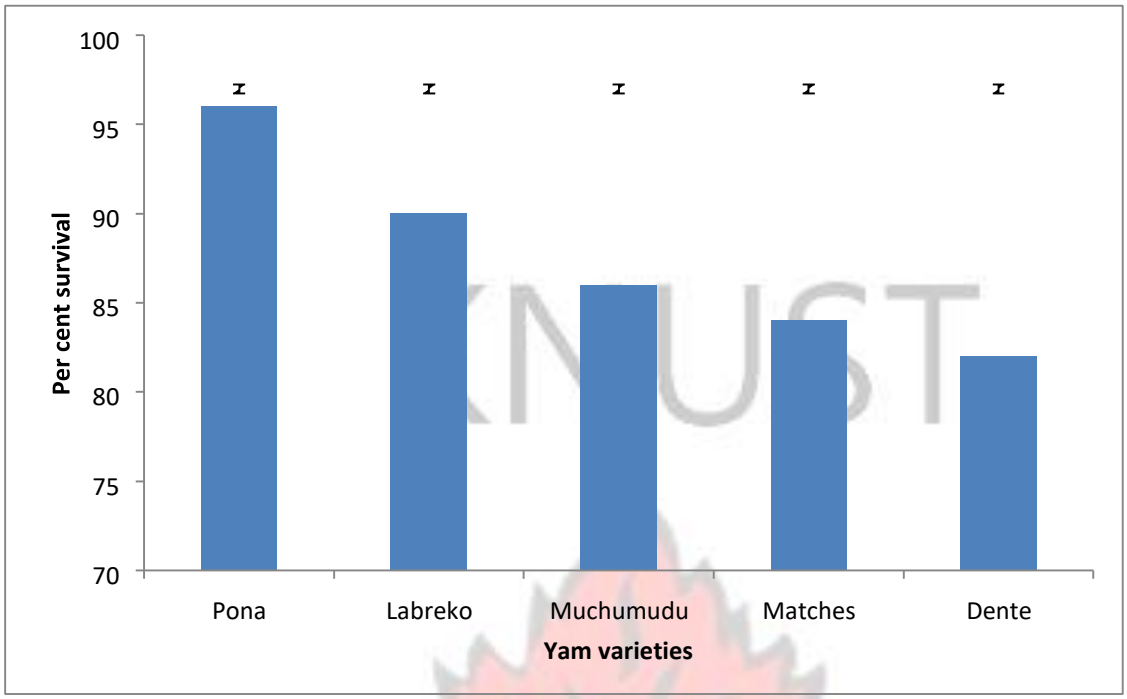

Figure 4.4 Mean per cent survival of *P. citri* males reared on tubers of five yam varieties

The mean per cent survival of *P. citri* males reared on Pona, Labreko, Muchumudu, Matches and Dente was between 82 % and 96 % (Figure 4.4). The highest mean per cent survival was observed on Pona and lowest on Dente. The differences between the mean per cent survival of *P. citri* male reared on all the yam varieties were significant (Figure 4.4).

4.13.0 Effect of yam variety on *P. citri* offspring sex ratio

The mean per cent offspring sex ratios are presented in Figure 4.5. The sex ratio expressed in terms of the proportion of females, favoured females on all the five yam varieties. The mean per cent offspring sex ratio of *P. citri* reared on Pona, Labreko, Muchumudu, Matches and Dente ranged from 52.6 to 59.2 % (Figure 4.5).

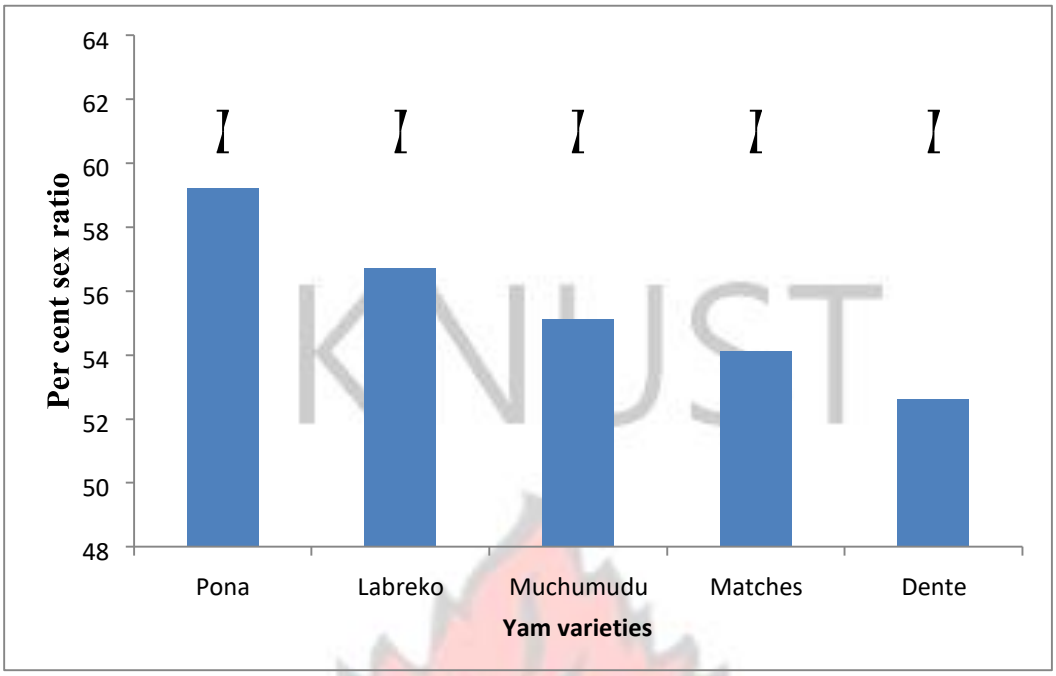

Figure 4.5. Offspring sex ratio of *P. citri* reared on tubers of five yam varieties

The highest mean per cent offspring sex ratio was recorded on Pona and the lowest on Dente. The differences between the mean per cent offspring sex ratio of *P. citri* reared on the yam varieties were significant except between Muchumudu and Matches which did not differ from each other (Figure 4.5).

4.14.0 Susceptibility of the yam varieties to *P. citri* infestation

4.14.1 Effect of yam variety on the growth index of *P. citri*

The growth index expressed as logarithm of per cent adult survival over average developmental period of *P. citri* bred on the five yam varieties are presented in Table 4.7. The mean growth index of female *P. citri* reared on Pona, Labreko, Muchumudu, Matches and Dente ranged from 0.05 to 0.08 (Table 4.7). The highest mean female growth index was recorded on Pona and the lowest on Dente which means Pona was the most susceptible yam variety to *P. citri* female infestation. The differences between the mean growth indices of *P. citri* female bred on the yam varieties were significant except between Muchumudu and Matches which did not differ from each other (Table 4.7).

Again, Pona recorded the highest *P. citri* male growth index of 0.09 followed by Labreko with 0.08. Dente recorded the lowest growth index of 0.06 (Table 4.7). The differences between the mean growth indices of *P. citri* male bred on the yam varieties were significant except between Muchumudu and Matches which did not differ (Table 4.7).

This indicates that Pona was the most susceptible yam variety to *P. citri* male infestation.

Table: 4.7. Growth indices of *P. citri* bred on the various yam varieties

Yam varieties	Mean growth index ± s.e.	
	Female	Iale
D. rotundata var. Pona	0.08 ± 0.003	0.09 ± 0.003
D. rotundata var. Labreko	0.07 ± 0.004	0.08 ± 0.005
D. rotundata var. Muchumudu	0.06 ± 0.006	0.07 ± 0.003
D. alata var. Matches	0.06 ± 0.003	0.07 ± 0.004
D. rotundata var. Dente	0.05 ± 0.003	0.06 ± 0.003
LSD (5 %)	0.0021	0.)038
CV (%)	2.5	3.9

4.15.0 Chemicals bioassay test

4.15.1 LD_{50} for the third instar larvae and adult *P. citri* exposed for 12, 24 and 48 h to the six chemicals

The LD_{50} of the third instar and adult *P. citri* at 12, 24 and 48 h exposure periods are shown in Figure 4.6. The results indicate that for both stages of the pest, LD_{50} decreased with exposure period. Thus lower dosages are required to kill fifty per cent of the larvae or the adults at longer exposure periods. However for the same exposure period, the lethal dosage for the third instar was smaller than the lethal dosage of the adult insect.

4.15.2 LD90 for third instar larvae and adult *P. citri* exposed for 12, 24 and 48 h to the six chemicals

The LD_{90} of the third instar and adult *P. citri* at 12, 24 and 48 h exposure periods are shown in Figure 4.7. For both stages of the pest, LD_{90} decreased with exposure period. Thus lower dosages are required to kill fifty per cent of the larvae or the adults at longer exposure periods. However, for the same exposure period, the lethal dosage for the third instar was smaller than the lethal dosage of the adult insect.

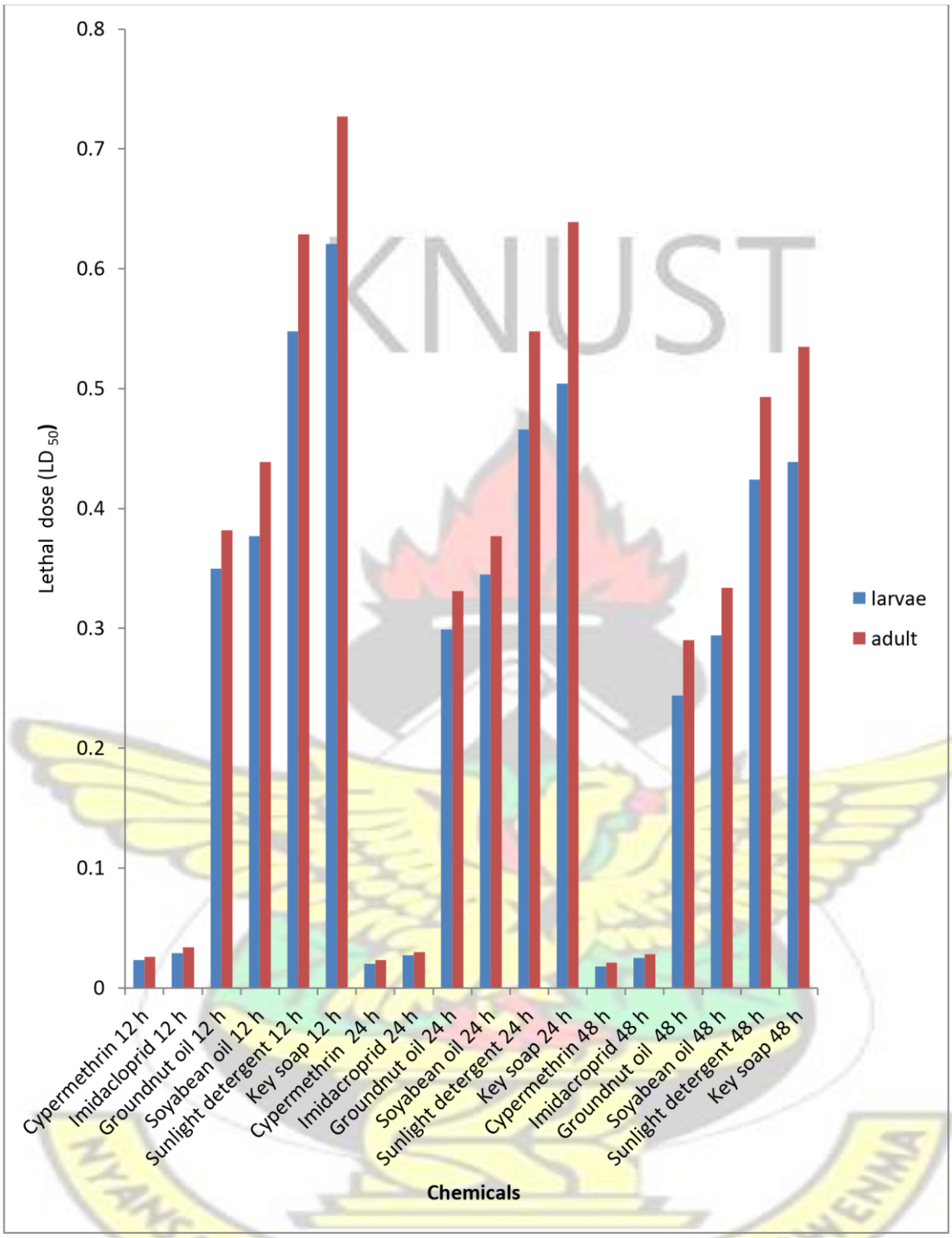

Figure 4.6. LD_{50} of the third instar larvae and adult *P. citri* exposed for 12, 24 and 48 h to the six chemicals

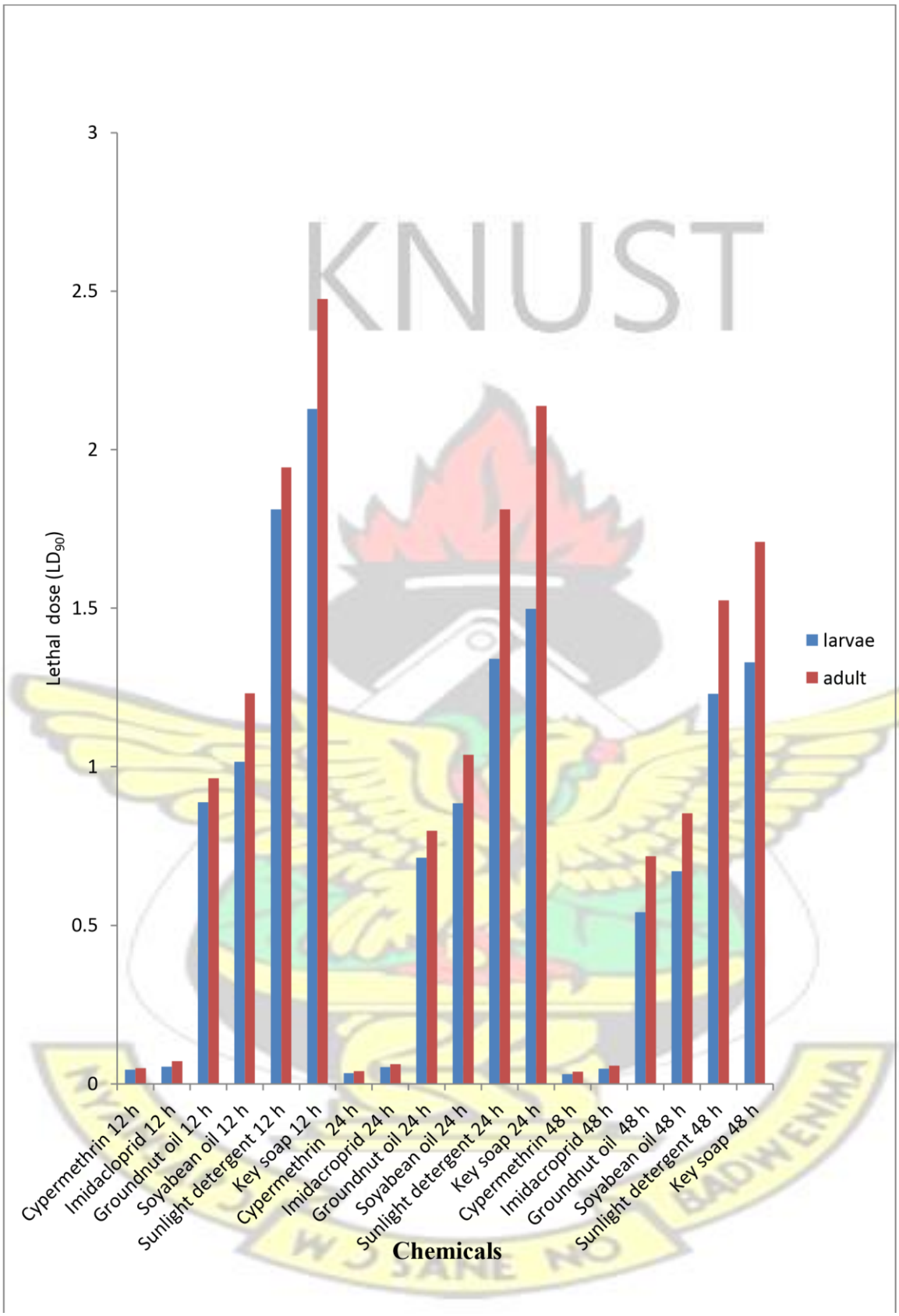

Figure 4.7. LD_{90} for the third instar larvae and adult *P. citri* exposed for 12, 24 and 48 h to the six chemicals

4.16.0 Slope values of log dosage - probit (ld-p) line of third instar and adult *P. citri*

The slope values of log dosage - probit line of the third instar larvae and adult *P. citri* exposed to the six chemicals at 12, 24 and 48 h exposure periods are shown in Table 4.8. The ld-p line slope values of third instar *P. citri* larvae over time using Cypermethrin, Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion, Sunlight detergent solution and Key soap solution at 12 h exposure period ranged from 2.496 to 4.879 (Table 4.8). The highest ld-p line slope value was recorded by Cypermethrin and the lowest by Key soap. The slope values of Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion and Sunlight detergent solution lied between those of Cypermethrin and Key soap solution (Table 4.8).

Table 4.8. Slope values of log dosage-probit line of third instar larvae and adult *P. citri* exposed to the six chemicals at 12, 24 and 48 h

Chemicals	ld-p line slope of third instar *P. citri* larvae over time (h)			ld-p line slope of adult *P. citri* over time (h)		
	12	24	48	12	24	48
Cypermethrin	4.879	5.553	5.834	4.639	4.886	5.355
Imidacloprid	4.400	4.749	4.775	3.925	4.016	4.210
Groundnut oil emulsion	3.268	3.393	3.714	3.192	3.251	3.349
Soyabean oil emulsion	2.975	3.134	3.585	2.864	2.913	3.144
Sunlight detergent solution	2.568	2.772	2.893	2.468	2.610	2.619
Key soap solution	2.496	2.665	2.711	2.408	2.442	2.542

The ld-p line slope values of third instar *P. citri* over time using Cypermethrin, Imidacloprid, Groundnut oil, Soyabean oil, Sunlight liquid detergent and Key soap solution

at 24 h exposure period ranged from 2.665 to 5.553 (Table 4.8). The highest ldp line slope value was recorded by Cypermethrin and the lowest by Key soap solution.

The slope values of Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion and Sunlight detergent solution lied between that of Cypermethrin and Key soap solution (Table 4.8).

The ld-p line slope values of third instar *P. citri* over time using Cypermethrin, Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion, Sunlight detergent solution and Key soap solution at 48 h exposure period ranged from 2.711 to 5.834 (Table 4.8). The highest ld-p line slope value was recorded by Cypermethrin and the lowest by Key soap solution. The slope values of Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion and Sunlight detergent solution lied between those of Cypermethrin and Key soap solution (Table 4.8).

The ld-p line slope values of adult *P. citri* over time using Cypermethrin, Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion, Sunlight liquid detergent and Key soap solution at 12 h exposure period ranged from 2.408 to 4.639 (Table 4.8). Cypermethrin had the highest ld-p line slope value and Key soap had the lowest ld-p line slope value. The slope values of Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion and Sunlight detergent solution lied between that of Cypermethrin and Key soap solution (Table 4.8).

The ld-p line slope values of adult *P. citri* over time using Cypermethrin, Imidacloprid, Groundnut oil, Soyabean oil, Sunlight detergent solution and Key soap solution at 24 h exposure period ranged from 2.442 to 4.886 (Table 4.8). Cypermethrin had the highest ld-p line slope value and Key soap had the lowest ld-p line slope value. The slope values of

Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion and Sunlight detergent solution lied between that of Cypermethrin and Key soap solution (Table 4.8).

The ld-p line slope values of adult *P. citri* over time using Cypermethrin, Imidacloprid, Groundnut oil, Soyabean oil, Sunlight detergent solution and Key soap solution at 48 h exposure period ranged from 2.542 to 5.355 (Table 4.8). Cypermethrin recorded the highest ld-p line slope value and Key soap recorded the lowest ld-p line slope value. The slope values of Imidacloprid, Groundnut oil emulsion, Soyabean oil emulsion and Sunlight detergent solution lied between those of Cypermethrin and Key soap solution (Table 4.8).

Generally, the steepness of the slope of the ld-p lines decreased for both the third instar larvae and adult *P. citri* for all the six chemicals tested. The decreasing order was as follows: Cypermethrin > Imidacloprid > Groundnut oil emulsion > Soyabean oil emulsion > Sunlight detergent solution > Key soap solution (Table 4.8). The slope values of the ld-p lines of the six chemicals at 12, 24 and 48 h exposure periods for the third instar *P. citri* were higher than the corresponding slope values of the ld-p lines of the six chemicals at 12, 24 and 48 h exposure periods for the adult *P. citri.*

4.17.0 Fiducial limits of third instar *P. citri* larvae LD_{50} values

Cypermethrin had the smallest third instar larvae LD_{50} at 12, 24 and 48 h exposure periods followed by Imidacloprid (Table 4.9).

Table 4.9. LD_{50} values and fiducial limits for the third instar larvae of *P. citri* exposed for 12, 24 and 48 h to the six chemicals by topical application

	Exposure period for third instar larvae of *P. citri* (h)		
	12	24	48
Chemicals			

	LD_{50}	95 % Fiducial limit	LD_{50}	95 % Fiducial limit	LD_{50}	95 % Fiducial limit
Cypermethrin	0.023	0.021-0.026a	0.020	0.018-0.022a	0.018	0.016-0.020a
Imidacloprid	0.030	0.027-0.033b	0.027	0.024-0.030b	0.025	0.023-0.028b
Groundnut oil emulsion	0.350	0.299-0.409c	0.299	0.256-0.346c	0.244	0.210-0.281c
Soyabean oil emulsion	0.377	0.321-0.444c	0.345	0.295-0.404c	0.294	0.254-0.339c
Sunlight detergent solution	0.548	0.454-0.687d	0.466	0.411-0.560d	0.424	0.358-0.508d
Key soap solution	0.621	0.509-0.802d	0.504	0.424-0.614d	0.439	0.369-0.530d

Significant difference is inferred by non-overlapping of 95 % fiducial limits

There were significance differences between the two insecticides. The two insecticides were significantly different from the other chemicals tested (Table 4.9).

The differences between the LD_{50} of Groundnut oil emulsion and that of Soyabean oil emulsion at 12, 24 and 48 h exposure periods were not significant even though the LD_{50} of Groundnut oil emulsion was smaller than that of Soyabean oil emulsion. However, these were significantly different from LD_{50} of Sunlight detergent solution and Key soap solution at 12, 24 and 48 h exposure periods (Table 4.9). Again, the differences between LD_{50} of Sunlight detergent solution and Key soap solution at 12, 24 and 48 h exposure periods were not significant even though the LD_{50} of Sunlight detergent solution was smaller than that of Key soap solution (Table 4.9).

4.17.1 Fiducial limits of adult *P. citri* LD_{50} values

Cypermethrin recorded the smallest values for all the exposure periods and were significantly different from the rest of the chemicals tested (Table 4.10). Imidacloprid

which was next to Cypermethrin was also significantly different from the remaining chemicals for all the exposure periods (Table 4.10).

Table 4.10. LD_{50} values and fiducial limits for the adult *P. citri* exposed for 12, 24 and 48 h to the six chemicals by topical application

Chemicals	Exposure period for adult *P. citri* (h)					
	12		24		48	
	LD_{50}	95 % Fiducial limit	LD_{50}	95 % Fiducial limit	LD_{50}	95 % Fiducial limit
Cypermethrin	0.026	0.023-0.029a	0.023	0.020-0.025a	0.021	0.018-0.023a
Imidacloprid	0.034	0.030-0.040b	0.030	0.027-0.034b	0.028	0.025-0.032b
Groundnut oil emulsion	0.382	0.327-0.447c	0.331	0.284-0.384c	0.290	0.247-0.337c
Soyabean oil emulsion	0.439	0.372-0.524c	0.377	0.320-0.446c	0.334	0.285-0.390c
Sunlight detergent solution	0.629	0.529-0.795d	0.548	0.454-0.687d	0.493	0.413-0.602d
Key soap solution	0.727	0.589-0.971d	0.639	0.525-0.825d	0.535	0.446-0.665d

Significant difference is inferred by non-overlapping of 95 % fiducial limits

The differences between the LD_{50} of Groundnut oil emulsion and that of Soyabean oil emulsion at 12, 24 and 48 h exposure periods for adult *P. citri* were not significant even though the LD_{50} of Groundnut oil emulsion was smaller than that of Soyabean oil emulsion. However, these were significantly different from LD_{50} of Sunlight detergent solution and Key soap solution at 12, 24 and 48 h exposure periods (Table 4.10). Again, the differences between LD_{50} of Sunlight detergent solution and Key soap solution at 12, 24 and 48 h exposure periods for adult *P. citri* were not significant even though the LD_{50} of Sunlight detergent solution was smaller than that of Key soap solution (Table 4.10).

4.17.2 Fiducial limits of third instar LD_{90} values

Cypermethrin had the smallest third instar LD_{90} at 12 h exposure period but was not significantly different from the LD_{90} of Imidacloprid at 12 h exposure period (Table 4.11). Even though the difference between Cypermethrin and Imidacloprid was not significant, the LD_{90} of Cypermethrin and Imidacloprid at 12 h was significantly different from those of the rest of the chemicals (Table 4.11). However, the differences between the LD_{90} of Groundnut oil, Soyabean oil, Sunlight detergent and Key soap solution at 12 h exposure were not significant, even though Groundnut oil had the smallest LD_{90} and Key soap the highest (Table 4.11).

Table 4.11. LD_{90} values and fiducial limits for the third instar larvae of *P. citri* exposed for 12, 24 and 48 h to the six chemicals by topical application

	Exposure period for third instar larvae of *P. citri* (h)					
	12		24		48	
Chemicals	LD_{90}	95% Fiducial limit	LD_{90}	95 % Fiducial limit	LD_{90}	95 % Fiducial limit
Cypermethrin	0.043	0.037-0.053a	0.034	0.031-0.040a	0.030	0.027-0.035a
Imidacloprid	0.055	0.046-0.074a	0.053	0.044-0.070b	0.047	0.040-0.059b
Groundnut oil emulsion	0.888	0.712-1.229b	0.713	0.583-0.955c	0.541	0.449-0.707c
Soyabean oil emulsion	1.016	0.799-1.455b	0.885	0.708-1.228c	0.670	0.552-0.886c
Sunlight detergent solution	1.811	1.276-3.232b	1.340	1.013-2.071c	1.230	0.936-1.874d
Key soap solution	2.128	1.447-4.109b	1.498	1.109-2.412c	1.329	0.994-2.093d

Significant difference is inferred by non-overlapping of 95 % fiducial limits

After 24 hours of exposure, Cypermethrin still had the smallest third instar LD_{90} and was significantly different from the LD_{90} of the remaining chemicals. The LD_{90} of Imidacloprid at 24 h exposure period was also significantly different ($P<0.05$) from the rest of the

chemicals (Table 4.11). The differences between the LD_{90} of Groundnut oil, Soyabean oil, Sunlight detergent and Key soap solution at 24 h exposure were not significant even though Groundnut oil had the smallest LD_{90} and Key soap the highest. It can also be seen from Table 4.11 that Cypermethrin had the smallest third instar LD_{90} after 48 h exposure which was significantly different from the rest of the chemicals tested, followed by Imidacloprid which was also significantly different from the remaining chemicals at 48 h exposure period (Table 4.11).

The differences between the LD_{90} of Groundnut oil and that of Soyabean oil at 48 h exposure period were not significant even though the LD_{90} of Groundnut oil was smaller than that of soyabean oil emulsion (Table 4.11). The LD_{90s} of Groundnut oil and Soyabean oil emulsion were significantly different from LD_{90} of Sunlight detergent and Key soap solution. The difference between the LD_{90} of Sunlight detergent and that of the Key soap solution was not significant even though the LD_{90} of Sunlight detergent solution was smaller than that of Key soap solution at 48 h exposure period (Table 4.11).

4.17.3 Fiducial limits of adult *P. citri* LD_{90} values

Cypermethrin had the smallest adult *P. citri* LD_{90} at 12, 24 and 48 h exposure periods and all were significantly different from the LD_{90} of the rest of the chemicals (Table 4.12). The LD_{90s} of Imidacloprid at 12, 24 and 48 h exposure periods respectively were also significantly different from the LD_{90} of the remaining chemicals at all the exposure periods ((Table 4.12)..

The differences between the LD_{90} of Groundnut oil, Soyabean oil, Sunlight detergent solution and Key soap solution at 12 and 24 h exposure were not significant even though

Groundnut oil emulsion had the smallest LD_{90} and Key soap the highest (Table 4.12).. The differences between the LD_{90} of Groundnut oil, Soyabean oil emulsion and Sunlight detergent solution at 48 h exposure were also not significantly different. However, LD_{90} of Groundnut oil and Soyabean oil emulsion were significantly different from the LD_{90} of Key soap solution at 48 h exposure even though the LD_{90} of Key soap solution and Sunlight detergent solution at 48 h exposure were not significantly different from each other (Table 4.12).

Table 4.12. LD_{90} values and fiducial limits for the adult *P. citri* exposed for 12, 24 and 48 h to the six chemicals by topical application

Chemicals	Exposure period for adult *P. citri* (h)					
	12		24		48	
	LD_{90}	95 % Fiducial limit	LD_{90}	95 % Fiducial limit	LD_{90}	95 % Fiducial limit
Cypermethrin	0.049	0.042 - 0.063a	0.039	0.035 - 0.047a	0.038	0.033 - 0.046a
Imidacloprid	0.071	0.076 - 0.114b	0.062	0.050 - 0.091b	0.057	0.047 - 0.079b
Groundnut oil emulsion	0.963	0.770 - 1.340c	0.798	0.649 - 1.077c	0.718	0.582 - 0.973c
Soyabean oil emulsion	1.231	0.945 - 1.843c	1.038	0.811 - 1.504c	0.853	0.683 - 1.182c
Sunlight detergent solution	1.944	1.372 - 3.481c	1.811	1.276 - 3.232c	1.525	1.118 - 2.505cd
Key soap solution	2.475	1.634 - 5.123c	2.138	1.459 - 4.096c	1.709	1.224 - 2.949d

Significant difference is inferred by non-overlapping of 95 % fiducial limits.

4.18.0 Efficacy of the chemicals against *P. citri* on infested yams in improved barns

The laboratory LD_{90} of the adult *P. citri* after 12 h exposure was used for this assay. The analyses of variance of the arcsine (angular) transformed data on effect of the chemicals on per cent mortality of *P. citri* on stored Pona after 12, 24, 48 and 72 h exposure indicated

that per cent mortality was significantly influenced by the chemicals. Mean per cent mortalities caused by each chemical are shown in Table 4.13.

Table 4.13. Mean per cent mortalities of *P. citri* on stored yams after 12, 24, 48 and 72 h exposure to six chemicals using adult 12 h LD_{90} of respective chemicals

Chemicals	LD_{90}	Mean per cent mortality ± s.e after periods of exposure (h)			
		12	24	48	72
Cypermethrin	0.049	67.4 ± 2.94	76.2 ± 1.56	82.8 ± 1.16	87.0 ± 1.92
Imidacloprid	0.071	59.2 ± 1.39	67.6 ± 3.23	75.2 ± 1.28	78.6 ± 1.08
Groundnut oil	0.963	52.2 ± 1.80	58.0 ± 1.82	70.2 ± 1.80	73.4 ± 1.50
Soyabean oil	1.231	42.8 ± 1.39	53.2 ± 1.69	66.0 ± 2.28	69.2 ± 1.50
Sunlight™	1.944	38.0 ± 0.63	45.0 ± 1.52	54.2 ± 1.53	58.2 ± 1.43
Key soap	2.475	34.0 ± 1.82	40.2 ± 1.02	50.2 ± 1.59	54.0 ± 1.34
Control (water)		0.8 ± 0.37	1.0 ± 0.32	1.4 ± 0.25	1.6 ± 0.25
LSD (5 %)		3.43	3.49	2.83	2.83
CV (%)		6.90	6.30	4.50	4.40

Cypermethrin (0.049 %) sprayed on stored Pona caused 67.4 %, 76.2 %, 82.8 % and 87.0 % mortalities at 12, 24, 48 and 72 h exposure respectively, which were significantly different ($P<0.05$) from mortalities caused by the other chemicals (Table 4.13).

Imidacloprid (0.071 %) sprayed on stored Pona caused 59.2 %, 67.6 % , 75.2 % and 78.6 % mortalities at 12, 24, 48 and 72 h exposure periods respectively, which were also significantly different ($P<0.05$) from mortalities caused by the remaining chemicals (Table 4.13).

Mortality caused by Groundnut oil emulsion (0.963 %) at 12 h exposure period was significantly different from mortality caused by the rest of the chemicals. Mortality caused

by Soyabean oil emulsion (1.231 %) did not differ significantly from mortality caused by Sunlight detergent (1.944 %) but differed significantly from mortality caused by Key Soap (2.475) after 12 h of exposure. The difference between mortalities caused by Sunlight detergent and Key soap solution at 12 h exposure period was not significant (Table 4.13).

Groundnut oil emulsion (0.963 %) sprayed on stored Pona caused 58.0 % and 70.2 % mortalities at 24 and 48 h exposure periods respectively and were not significantly different from mortalities caused by Soyabean oil emulsion (1.231 %) at 24 and 48 h exposure. Even though the mortality caused by Groundnut oil emulsion was higher than the mortality caused by Soyabean oil emulsion these were significantly different from mortality caused by Sunlight detergent solution and Key Soap solution at 24 and 48 h exposure periods respectively (Table 4.13). Mortalities caused by Groundnut oil emulsion (0.963 %) and Soyabean oil emulsion (1.231 %) at 72 h exposure period were significantly different from each other and also significantly different from the rest of the chemicals (Table 4.13).

Mortalities caused by Sunlight detergent solution (1.948 %) at 24, 48 and 72 h exposure periods were not significantly different from mortality caused by Key soap (2.475 %) at 24, 48 and 72 h exposure period even though the mortality caused by Sunlight detergent solution was higher than the mortality caused by Key Soap solution (Table 4.13).

4.18.1 Effect of multiple application of chemicals on *P. citri*

The chemicals were applied at 0, 7, 14 and 21 days and data taken 24 h after each application. Mean per cent mortalities caused by each chemical are shown in Table 4.14.

Table 4.14. Mean per cent mortalities of *P. citri* on stored *D. rotundata* var. Pona after one, seven, 14 and 21 days applications of the six chemicals

		Mean per cent mortality ± s.e of *P. citri* in days			
Chemicals	LD_{90} (%)	0	7	14	21
Cypermethrin	0.049	76.2 ± 1.56	99.0 ± 0.32	100 ± 0.0	0.00
Imidacloprid	0.071	67.6 ± 3.23	98.0 ± 0.71	100 ± 0.0	0.00
Groundnut oil	0.963	58.0 ± 1.82	97.8 ± 0.20	100 ± 0.0	0.00
Soyabean oil	1.231	53.2 ± 1.69	89.4 ± 1.03	99.6 ± 0.24	100 ± 0.0
Sunlight™	1.944	45.0 ± 1.52	80.8 ± 0.80	93.2 ± 1.28	100 ± 0.0
Key soap	2.475	40.2 ± 1.02	78.8 ±0.58	92.0 ± 0.95	100 ± 0.0
Control (water)		1.0 ± 0.32	0.40 ± 0.24	1.0 ± 0.32	1.0 ± 0.32
LSD (5 %)		3.49	4.10	2.92	2.20
CV (%)		6.3	3.4	3.1	2.0

Table 4.14 shows that a second application of Cypermethrin, Imidacloprid and Groundnut oil emulsion to stored yams caused over 95 per cent mortality which was significantly higher than mortality caused by Soyabean oil, Sunlight detergent and Key soap solution, which killed less than 90 % of the mealybugs. However, after a fourth application, Soyabean oil emulsion, Sunlight detergent and Key soap solution also killed all the *P. citri* on the tubers, even as the third application killed more than 90 % (Table 4.14).

4.19.0 A Test of Significance (Chi- Square Test)

A test of significance was calculated to determine whether the probit regression lines satisfactorily represented the data. Table 4.15 shows the calculated X^2 values for the six chemicals.

Table 4.15. Calculated X^2 values for the six chemicals

Chemicals	X^2 for the stages of *P. citri* development over time (h)					
	P. citri	rd instar vae		Adult *P. citri*		
	12	24	48	12	24	48
Cypermethrin	0.795	0.907	0.717	0.779	0.961	0.640
Imidacloprid	0.518	0.632	8.715	0.543	0.805	0.775
Groundnut oil emulsion	0.607	0.693	0.626	0.863	0.605	0.631
Soyabean oil emulsion	0.528	0.984	0.927	0.960	0.720	0.447
Sunlight detergent	0.842	0.615	0.805	0.759	0.842	0.832
Key soap solution	0.791	0.791	0.961	0.892	0.977	0.875

The X^2 values obtained for all the chemicals were smaller than the critical values of 6.63 and 3.84 at 1 % and 5 % levels of significance. Therefore, it can be concluded that all the results obtained were within the limits of random variation and that the observed results are in very close agreement with the expected results. Therefore, the probit regression lines are satisfactory representations of the results.

CHAPTER FIVE

5.0 DISCUSSION

5.1.0 Influence of nature of food on insect development

From the results, it was clear that oviposition period, developmental period, longevity, total eggs laid, per cent survival, offspring sex ratio and growth index of *P. citri* followed almost a set pattern, with Pona being the preferred variety while Dente was the least preferred.

The nutrient compositions of the five yam varieties (*D. rotundata* var. Pona, *D. rotundata* var. Labreko, *D. rotundata* var. Muchumudu, *D. rotundata* var. Dente and *D. alata* var. Matches) although, not determined in this experiment would be different from each other. Several researchers have shown that the nutrient composition of white and water yams differ (Bradbury and Holloway, 1988; Osagie, 1992; Muzac-Tucker *et al.,* 1993; Opara, 1999). These differences in the nutrient composition might have influenced the performance of the mealybugs on the five yam varieties to varying degrees. Painter (1968) observed that the developmental period of an insect is influenced by the nature of food on which it is reared and generally, development is faster on favourable ones, according to Polat *et al.* (2008), development, longevity and fecundity of *P. citri* vary according to the suitability of the host plant on which it is bred. Noha and Shaaban (2010) also observed that host plants and temperatures greatly influenced the development of *P. citri*.

Several researchers have reported variations in biological parameters such as oviposition period, developmental period and longevity of insects on different hosts. Persad and Khan (2007) and Woets and van Lenteren (1976) attributed differences in whitefly populations on different host plants to a combination of the effect of the host plant on lifespan and

developmental rate of the insect. Sakurai *et al.* (1991) reported that the quality of food and environmental factors such as temperature and humidity play an important role in the different aspects of the biology of coccinellid beetles**.** Metspalu *et al.* (2003) also observed that the amount and composition of nutrients, particularly vitamins contained in plants, considerably influence the development of caterpillars of large white butterfly (*Pieris brassicae* L.).

According to Hogendorp *et al.* (2006) leaf nitrogen concentration affected citrus mealybug life history parameters, with those mealybugs fed on plants containing the highest leaf nitrogen having the shortest developmental times.Working with *P. citri*, Yang and Sadof (1997) reported that growth and development of the mealybug differed substantially when fed on red, yellow or green leafed *S. scutellarioides* (L.) Codd. Indeed, there must be variations in the nutrient composition of the various yam varieties, accounting for the differences observed in the different parameters measured.

5.2.0 Life stages of *P. citri*

Studies on the development of *P. citri* showed that the female *P. citri* has three nymphal stages and the male has four nymphal stages, a result not different from what was reported by Gullan (2000) and Noha and Shaaban, (2010). The fourth nymphal stage exhibited by the male citrus mealybug is the pupal stage where physiological development and changes enable its transformation into a winged adult capable of flying in search of females for mating purpose (McKenzie, 1967).

5.3.0 Effect of yam variety on incubation period of *P. citri*

Results of the present study showed that the incubation period of *P. citri* eggs placed on the five yam varieties ranged from 10 to 11 days at a temperature range of 26 – 30 ^{o}C in

the laboratory and the differences between the various yam varieties were not significant. Studies in India by the Research Department of the Coffee Board (1984) showed that the incubation period of studies of *P. citri* reared on coffee leaves in the laboratory ranged from 2 to 10 days. Fasulo and Brooks (2005) also reported the incubation period for *P. citri* was dependent on temperature and spanned 6 to ten days often times to several weeks. The lack of differences between the mean incubation periods of *P. citri* eggs on the yam varieties could be due to the similar temperature and relative humidity in the laboratory (Fasulo and Brooks, 2005; Goldasteh *et al.,* 2009; Noha and Shaaban, 2010).

5.4.0 Effect of yam variety on per cent egg hatch of *P. citri*

The per cent egg hatchability of *P. citri* recorded in this study are similar to those reported by Polat *et al.* (2008), even though their experiments were carried out on *Sygonium podophyllum* Schott, *Nerium oleander* L., *Kalanchoe blossfeldiana* Poelln. and *Schefflera arbicola* (Hayata) Merr., plants not related to yams. Their experiments were however conducted under a constant temperature of 28 ± 1 ^{o}C and varying relative humidity of 65 ± 10 %. This similiarity in per cent egg hatchability could be due to the similar temperature and relative humidity under which the experiments were conducted.

5.5.0 Effect of yam variety on developmental period of *P. citri*

5.5.1 Effect of yam variety on female larval duration and adult female longevity

Goldasteh *et al.* (2009) observed that the mean duration of first, second and third female instars of *P. citri* bred on red variegated coleus, *Solenostemon scutellarioides* (L.) Codd in the laboratory lasted about 6.4, 8.7 and 9.5 days respectively, with the mean female larval period (1st instar to 3rd instar) being almost 25 days. These durations are similar to those obtained on Pona but shorter than the results obtained on the other varieties.

However, Cloyd (1999) reported 33.7 days as the female larval period of *P. citri* on red variegated coleus, which is comparable to the mean female larval period on Muchumudu (31.9 days) and Matches (32.7 days) but shorter than that on Dente (37.0 days).

In this experiment, the female larval developmental periods on the five test varieties were in the increasing order from Pona through Labreko, Muchumudu, and Matches to Dente. These differences in the female larval duration might be due to differences in the nutrient contents of the yam varieties (Painter, 1968; Noha and Shaaban, 2010).

5.5.2 Effect of yam variety on male larval duration and adult male longevity

Studies on coffee leaves (Coffee Board Research Department, 1984) in the laboratory indicated that the male first nymphal stage lasted an average of 9.9 days, the second nymphal stage averaged 8.7 days, the male third nymphal stage (pre-pupa) lasted for about 2.5 days, while the male fourth nymphal stage (pupa) lasted for 3 days. These results are similar to the results of the present study. The observed similarities in duration of the pre-pupa and pupa stages of *P. citri* could be because the pre-pupa and pupa stages do not feed (Laura *et al.*, 2010).

The study by Goldasteh *et al*. (2009) on coleus and on coffee leaves (Coffee Board Research Department, 1984), showed that adult male mealybugs lived for a maximum 4 days after the final nymphal moult, a report comparable to what was observed on all the yam varieties. The observed similarity in mean male longevity of *P. citri* could be because the adult males do not feed and are therefore, short-lived (Laura *et al.*, 2010).

5.6.0 Effect of yam variety on mode of reproduction

It was observed that reproduction of *P. citri* was sexual which is in conformity with the report by Laura *et al.* (2010) which said that *P. citri* reproduction was strictly sexual.

5.7.0 Effect of yam variety on mean number of eggs laid by *P. citri*

There were significant differences between the mean number of eggs laid by female *P. citri* bred on the various yam varieties. This study revealed that the preference for oviposition decreased in the order Pona > Labreko > Muchumudu > Matches > Dente.

It is not clear why the number of eggs deposited on the different yam tubers should differ since the size of the tubers were not limiting. Probably the mother mealy bugs may have probed and determined the quality of food in the tubers hence determine the number of eggs to deposit per ovisac. Similar differences in total number of eggs laid by *P. citri* were reported by Noha and Shaaban (2010) who studied *P. citri* on citrus, guava and grape.

The differential preference for the various yam varieties by *P. citri* for oviposition could be due to the varying fibrous texture and nutrient differences between the various yam varieties. Messina (1984) and Wasserman (1981) found that oviposition preference of *C. maculatus* was strongly influenced by the testa texture of cowpea, *Vigna unguiculata* (L) Walp. variety on which it is bred; the greater the fibrous texture the higher the rate of oviposition. In another study on cassava, Anthony and Lisbeth (1994) observed that oviposition by *Crytomenmus bergi* Froeschner was inversely proportional to the cyanide content of the cassava variety.

5.8.0 Effect of yam variety on pre-oviposition, oviposition and post-ovipostion periods of *P. citri*

The pre-oviposition period of *P. citri* reared on the five yam varieties ranged from 12.2 to 14.9 days. Durations similar to those observed in this study were reported by Goldasteh *et al.* (2009), while durations shorter than those observed in this study were reported by Polat

et al. (2008). The pre-oviposition period of *P. citri* reared on the five yam varieties was longer than those determined by Polat *et al.* (2008). This difference in preoviposition periods could be due to differences in the chemical composition of the different host plants **(**Noha and Shaaban, 2010). The oviposition period of *P. citri* reared on the five yam varieties ranged from 16 to 22 days which is similar to the findings of Goldasteh *et al.* (2009), who reported a range of 13 to 23 days.

5.9.0 Effect of yam variety on per cent survival of female *P. citri*

Among the five yam varieties studied, per cent survival of female *P. citri* followed the following trend, Pona > Labreko > Muchumudu > Matches > Dente. Polat *et al.* (2008) observed differences in per cent survival of *P. citri* bred on *K. blossfelddiana, S. podophyllum, N. oleander* and *S. arbicola*. The differences in per cent survival of *P. citri* on the various yam varieties could be due to differences in the nutrient compositions of the five yam varieties.

5.10.0 Effect of yam variety on offspring sex ratio

The sex ratio expressed in terms of the proportion of females was female-biased on all the five yam varieties, similar to that reported by Polat *et al.* (2008) and Goldasteh *et al.* (2009). Mealybugs vary their sex ratio depending on the availability of food and also on the level of crowding. When food is not limiting more females are produced, but when food quality and quantity become limiting more males are produced as they lack functional mouthparts and are also alates and fly away (Charnov, 1982; West, 2009).

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5.11.0 Growth index of *P. citri*

The proportion of young larvae reaching the adult stage was assessed and the time required to do so. The growth index of *P. citri* on the various yam varieties followed the descending

order of Pona > Labreko > Muchumudu > Matches > Dente. Bhattacharya *et al.* (1976) attributed low growth index to poor dietary quality and presence of some growth inhibitors in the substrate. The various yam varieties most likely had different nutrient quality and composition which reflected in the varying growth indices.

5.12.0 Bioassay test on *P. citri*

5.12.1 The log dosage-probit (ld-p) line, LD_{50} and LD_{90} of third instar and adult *P. citri* exposed to the various chemicals

Among all the six chemicals tested the mealybugs were most susceptible to Cypermethrin and least susceptible to Key soap solution and the LD_{50} and LD_{90} of third instar and adult *P. citri* decreased with time for all the chemicals tested. All the chemicals tested affected juveniles of the mealybug at concentrations smaller than for adults, as has been observed for other hemipterans (Curkovic *et al.*, 2007). This is probably related to the smaller size of nymphs and the presence of less amount of surface wax on nymphs than adults which make them more susceptible to the chemicals. This observation corroborates the findings of Larry *et al.* (2002) who reported that methyl bromide caused the highest mortality in Pink Hibiscus mealybug, *Macronellicoccus hirsutus* (Green) in the egg stage followed by instars and adults. Curkovic *et al.* (2007) also reported the greater susceptibility of longtailed mealybug, *Pseudococcus longispinus* (Targioni-Tozzetti) nymphs than adults.

The concentration of a chemical required to give a particular kill increases with older and bigger instars, the rate depending on the species and the chemical (Busvine, 1971). The results of this study agree with this in which the concentration of chemical required to kill the adult *P. citri* was higher than the third instar larva. Mulla (1961) observed an increase in the concentration of the organophosphate insecticides to kill *Culex pipiens fatigans* L. larvae from the first to fourth instar, while Klinger (1969) also reported that the increase in

concentration of organophosphate insecticides to kill old instars was due to an increase in culticular thickness, often associated with a sclerotised exocuticle and sometimes with a reduction in the number of pore canals.

The adult *P. citri* was bigger than the third instar. It appears that the bigger size of the adult *P. citri* with increased body wax cover might have also contributed to the lower sensitivity to the chemicals tested. Robertson and Falther (1966) also showed that size was a contributing factor to the mortality of the larvae of silkworm in which third instar stage was killed faster than the fourth instar.

5.13.0 Application of chemicals to infested yams in improved barns

Two applications of Cypermethrin, Imidacloprid and Groundnut oil were needed to cause about 95 % of *P. citri* on infested Pona yams in improved storage barns. But two more applications of Soyabean oil, Sunlight detergent and Key soap were needed to achieve similar results.

Cypermethrin is widely used in the control of various agricultural pests belonging to the orders Lepidoptera, Coleoptera, Diptera, and Hemiptera (Liu *et al.,* 1998; Cox, 1996). Likewise, Imidacloprid is known for its efficacy against several insects including the Pseudococcidae, Psyllidae, the Thysanoptera and many Coleoptera (Kaaken *et al.,* 1996; Martin and Workman, 1999; Boulahia *et al.,* 1996; Mistra *et al.,* 2003).

The insecticidal ability of dilute Soyabean oil emulsions have been reported for the whitefly *Bemisia tabaci* Grenadius and mites (Puri *et al.,* 1991; Butler *et al.,* 1993; Pless *et al.,* 1995; Broza *et al.,* 1988). Other oils, for example, coconut, groundnut and safflower, deterred oviposition and caused high mortality in cowpea bruchids (Messina and Renwick, 1983);

cottonseed, maize and groundnut oils have caused both adult mortality and reduction in the progeny in granary weevil (Qi and Burkholder, 1981).

Fenigstein *et al.* (2001) reported that groundnut oil was more effective than soyabean oil on sweet potato whitefly. This finding is similar to the results of this study in which groundnut oil emulsion gave a better control of *P. citri* than the soyabean oil emulsion. Cockfield (1992) found that groundnut oil appears nearly as effective as pirimiphosmethyl for control of *C. maculatus* in cowpea grain in The Gambia. Butler and Henneberry (1990) showed that soybean, maize (Zea *mays* L.) sunflower (*Helianthus annuus* L.), safflower (*Carthamus tinctorius* L.), groundnut (*A. hypogaea*) and cotton (*Gossypium hirsutum* L.) seed oils provided between 97 and 99 % control of the twospotted spider mites (*Tetranychus urticae* Koch) on several vegetable crops.

In this study, 1.9 % Sunlight detergent solution and 2.5 % Key soap solution caused 93.2 % and 92 % mortality of *P. citri* on the stored *D. rotundata* var. Pona respectively. The pesticidal ability of the detergents may be due to blockage of the spiracles which will cause the pests to suffocate and or that the detergents cause a reduction in the superficial tension of the insect (Curkovic *et al.*, 2007). In other research elsewhere, Wayne *et al.* (1999) observed consistent control of a mixed aphid population of *Myzus persicae* (Sulz), *Aphis citricola* van der Goot and *A. fabae* Scop. using Ivory detergent solution in the range of 1 to 2 per cent concentration. Lindquist (1981) used soaps to control citrus mealybug, *P. citri*, while David *et al.* (2009) reported that the populations of *Polyphagotarsonemus latus* Banks on chillies could be suppressed with soft soap sprays.

According to Curkovic *et al.* (2007) both nymphs and adults of the long-tailed mealybug, *Pseudococcus longispinus* (Targioni-Tozzetti) were highly susceptible to the SU 120 and Tecsa Fruta detergents tested at varying levels.

Abbasi *et al.* (1984) reported that a splash of commercially available soaps in water would kill American cockroaches at a 1-2 % solution. Szumlas (2002) reported that Dawn Ultra®dishwashing liquid concentrations higher than 1 % achieved 95 % or greater knockdown or mortality of German cockroaches. The results from this study are in close agreement with Szumlas (2002) and Abbasi *et al.* (1984) in which 1.95 % sunlight dishwashing detergent and 2.48 % key soap solution achieved 90 % control of *P. citri* fourteen days after application on infested stored yams**.**

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATIONS

6.1.0 CONCLUSION

The results obtained from the study indicated that there are three instar stages of the female *P. citri* and four instar stages of the male during development, and that the adult female lived longer than the adult male.

Development, longevity and the total number of eggs laid by *P. citri* were influenced by the yam variety. The order of preference of *P. citri* for development, longevity and oviposition was *D. rotundata* var. Pona followed by *D. rotundata* var. Labreko, *D. rotundata* var. Muchumudu then *D. alata* var. Matches and *D. rotundata var.* Dente. Per cent survival of female *P. citri* on the yam varieties followed the same trend as the order of preference.

Sex ratio favoured females on all the five yam varieties and that the population was female-biased. The maximum female sex ratio occurred on *D. rotundata* var. Pona and the minimum on *D. rotundata* var. Dente with *D. rotundata* var. Labreko, *D. rotundata* var. Muchumudu and *D. alata* var. Matches in between.

D. rotundata var. Dente was less susceptible to the mealybug infestation and therefore should be the preferred yam variety for long-term storage in areas where the mealybug is endemic whereas *D. rotundata* var. Pona was the most susceptible and should not be stored for long but rather consumed early unless protective measures are taken against *P. citri* infestation.

The third instar stage of *P. citri* was more susceptible than the adult stage to all the tested chemicals.

Cypermethrin was the most effective of the six chemicals tested on *P. citri* followed by Imidacloprid. Groundnut oil emulsion was better than soyabean oil emulsion followed by Sunlight detergent solution which was better than Key soap solution.

For rapid control of *P. citri* infestation of yam setts in storage, cypermethrin should be the preferred chemical and should be applied at the concentration of 0.049 %. For control of *P. citri* infestation of ware yam, any of the following chemicals, groundnut oil emulsion (about 1 %), soyabean oil emulsion (about 1.2 %), sunlight dishwashing detergent (about 2 %) and key soap solution (about 2.5 %) concentrations could be used**.**

6.2.0 RECOMMENDATIONS

1. The laboratory LD_{90} of the adult *P. citri* after 12 h exposure, which was used to control the mealybugs on yams in improved barns, should be tested on large scale on farmers' fields in both the traditional and improved yam barns for authentication and adoption.

2. Residue analysis should be conducted on yam tubers treated with cypermethrin and imidacloprid.

3. The results from this research showed that soyabean oil emulsion and groundnut oil emulsion were effective in controlling *P. citri*. Further research should be conducted into the possibility of using other vegetable oils to control *P. citri* infestation.

4. The results from this study also showed that Sunlight dishwashing detergent and Key soap solution were effective in controlling *P. citri* infestation. Further research should be conducted into the possibility of using other household soaps and detergents to control *P. citri* infestation.

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