EFFECTS OF DIEFERENT BEAN VARIETIES ON OVIPOSITION BEHAVIOUR AND FECUNDITY OF *Acanthoscelides obtectus* SAY (COLEOPTERA: BRUCHIDAE).

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DECLARATION

This thesis is my original work and has not been presented for a degree in any university.

.... 30th November, 1995.

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This thesis has been submitted for examination with our approval as university supervisors.

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DEDICATION

To Jemosop and Jemuge

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ABSTRACT

The interactions between the bean bruchid *Acanthoscelides obtectus* and six bean varieties (Red haricot, Mwitemania, Rose coco, Nyayo, Canadian wonder and Mwezi moja) under cultivation in Kenya were studied under laboratory conditions. Experiments were conducted to investigate: (1) The effects of bean varieties on oviposition site preference by the beetles. (2) The effects of bean varieties on fecundity (3) The effects of delayed bean provision on oviposition (4) The susceptibility of bean varieties to attack by the beetles using a susceptibility index.

Similarly, bean seed sizes, testae thicknesses and hardness of seeds of the various bean varieties were determined. Later on they were used to find out whether their variations could be used to explain the differences in preference, fecundity and susceptibilities. The experimental room was maintained at temperatures of 27 ± 1^0c and 70 ± 5 % relative humidity.

The results indicated that Mwezi moja was the most preferred variety as oviposition site, whereas Mwitemania was the least preferred. Number of eggs laid by the beetles were significantly different between varieties (X^2 = 32.1, d.f = 5, $P < 0.05$). Beetles laid the highest number (54.6 ± 4.7) of eggs in Mwezi moja
and the least (35.4 ± 5.2) in Mwitemania. Egg laying

duration (days) was not significantly different between varieties ($F = 0.62$, d.f = (5, 54), $P > 0.05$). Beans induced oviposition in the majority of females. When provision of bean varieties was delayed for 0 - 16 days, beetles laid the highest number (15) of eggs on Mwezi moja and the least (7) on Mwitemania. Delayed bean provision reduced fecundity and prolonged the egg laying duration.

Number of progeny of *A. obtectus* emerging were significantly different between bean varieties ($X^2 = 77.0$, d.f = 5, $P < 0.05$). The highest number (490.4 ± 40.1) of progeny emerged from Mwezi moja and the least (299.7 ± 39.9) from Mwitemania. Duration taken by 50 % of progeny to emerge was significantly different between varieties ($F = 6.55$, d.f = (5, 54), $P < 0.05$). Beetles took the shortest period (36.6 ± 0.1 days) to emerge from Mwezi moja and the longest period (39.2 ± 0.5 days) from Mwitemania. Susceptibility index was significantly different between bean varieties ($F = 3.33$, d.f = (5, 54), $P < 0.05$). Mwezi moja was the most susceptible variety to attack by *A. obtectus* with a susceptibility index of 5.81 ± 0.4 and Mwitemania was the least susceptible with a susceptibility index of 3.36 ± 0.6.

Seed size and thickness of testa affected fecundity of the beetles. The differences in preference of bean varieties by the beetles was attributed to seed size. Physical characteristics of seeds of the bean varieties played a subordinate role on susceptibility in most of the bean varieties.

CHAPTER ONE

1.10 INTRODUCTION

Most diets in developing countries are deficient in prote in. Daily protein consumption in developing countries per person is 56 grams compared to 90 grams in economically developed countries. It has been recommended that for good health each person needs at least 70 grams of protein per day (Lowry, 1976). Some of the relatively cheap food crops that can help alleviate this protein crisis are the grain legumes of which the common bean (*Phaseolus vulgaris* L). has been reported to play a vital role in most developing countries (Borlang, 1975; Kaiser, 1976).

The common bean is one of the main grain legumes grown in the tropics and is grown extensively in East Africa and Asia. Singh and Van Emden (1979) estimated the world hectarage under beans as 2.4 million. As was noted above, beans as a source of protein can play an important role in the predominantly cereal diets in tropical countries such as Kenya because of their high food value (Table 1). The protein content of beans has been reported to be in the range of 16 - 33% depending on bean variety and environmental conditions under which they are grown (Bressani, 1975).

Nutritionally beans are 2 - 3 times richer in protein content than cereal grains (Parkin and Bills,1955), and are therefore often incorporated into livestock feed; and this has improved livestock production and inland fish production (F.A.O. Report, 1977). Furthermore, beans require little additional nitrogenous fertilizers for average growth because they have the capacity to provide their own nitrogenous compounds through bacterial nitrogen fixation in their root nodules. The bacteria convert nitrogen gas from the air into nitrogenous compounds which are utilized by beans for growth and development. In Kenya, the common bean is the major grain legume in terms of production and consumption (Khamala, 1978). It provides the cheapest source of protein, being about four times cheaper than animal protein. It is cultivated in various parts of the country along with other crops (Table 2). Mostly it is intercropped with other crops especially maize.

However, there are a number of factors that limit production of beans which include plant diseases, unreliable rainfall, high temperatures, inadequate plant nutrients in the soil and pests. Pests cause damage to the beans in the field as well as in the store. In the store, damage is caused by bruchids, of which the common bean bruchid *Acanthoscelides obtectus* Say causes considerable damage due to the feeding behaviour of their larvae. Once attacked by the bruchids the quantity and quality of the stored bean seeds are greatly reduced. The extent of economic losses due to insect attacks on stored beans are substantial in various countries. Thus, in Mexico and other

Central American countries economic losses have been estimated at 35 per cent and in Brazil as 13 per cent (Centro Internacional de Agricultura Tropical, 1986). In Kenya, it has been reported that 50 - 75 per cent of bean farmers experience crop loss to insects despite using various insect control methods (Schonherr *et al.*, 1976).

A. obtectus has been controlled using insecticides such as lindane, gamma B.H.C. and malathion. However,the control of stored product insects using malathion and lindane has been reported to have failed in several countries due to emergence of resistant strains (Champ and Dyte, 1977). Insect pests resistant to common insecticides are rapidly increasing (Hoskins and Gordon 1965; Parkin, 1965; Jacobson, 1975; Rowlands, 1976; Horber, 1979; Maxwell and Jennings, 1980). Moreover, insecticide residues in stored products could be more hazardous than in field crops. This is because in the store, insecticides are not exposed to weathering processes which reduce the hazardous effects of insecticides in field crops. Therefore, there is need to develop more effective and safe control measures for *A. obtectus*. One of these seem to be the breeding and selection of new varieties of beans that are resistant or less susceptible to attack by *A. obtectus* (Hussey, *et al.*, 1969; Kaiser, 1976; Horber, 1979; Vose and Blix, 1984). However, little has been done about it. Plant resistance as a method for insect pest control offers many advantages. In some cases it is the only method that is effective and practical (Horber, 1972). For crops grown in developing countries, perhaps the most attractive feature of using pest resistant varieties of crops including beans is that virtually no skill in pest control or cash investment is required at the grower level (Maxwell and Jennings, 1980).

Furthermore, a major advantage of using resistant varieties is the induction of a constant level of suppression on pest population growth. The number of pests which attack resistant varieties usually decline over time making control with insecticides easier. Resistant varieties are also highly compatible with biological control since both methods usually do not greatly affect natural enemies of the pest species (Adkisson and Dyck, 1980). Breeding and selection of plant varieties resistant or less susceptible to insect attack is based on insect-host plant relationships. Therefore before embarking on the breeding and selection programmes, it is of vital importance to know the characteristics of the plant and more so the behavioural and physiological characteristics of the pest.

Many varieties of beans are cultivated in Kenya and in fact some are grown in localized areas. Moreover, some of the varieties were recently bred and selected by researchers to suit various growing zones of the country. Furthermore, some selected varieties have been bred for their high yield and short growing season. However, although various control methods have been used to control *A. obtectus* in Kenya, it is still a major pest of stored beans.

In this study, it was considered important to study the behavioural relationship between *A. obtectus* and bean varieties under cultivation in Kenya. In particular, it was considered of scientific interest to establish how bean varietal differences affect the oviposition behaviour of the bruchids. It was hoped that certain bean varietal characteristics, physical or otherwise which significantly interfere with the bruchids oviposition behaviour would be useful to plant breeders in breeding and selecting varieties that are reasonably resistant to attack and damage by the beetles.

1.20 LITERATURE REVIEW

The common bean *Phaseolus vulgaris* has been domesticated for many years and records of its domestication date up to 700 years ago. Its origin has been attributed to Mexico (Kaplan, 1965; Westpal, 1977); and archaeological records show that varietal characteristics have remained remarkably stable for a long time (Kaplan, 1965).

P. vulgaris is a polymorphic species which includes several cultivated varieties. It has thus been known by many names depending on the locality where it is cultivated.This polymorphic characteristic is dependent on physical characters such as growth habit, flower colour, size and colour of pods and seeds.It is a twining or erect annual herb with fibrous roots (Smartt, 1976). It has been cultivated in Central and South America for many centuries and is known to have reached Europe by the 16th century and was probably spread to the coastal parts of Africa by the portuguese explorers. It became established as a food crop in Africa before the colonial era (Leakey, 1969). Beans grow well in most soil types from light sands to heavy clays (Purseglove, 1968). They also do well in areas of medium rainfall from the tropics to the temperate regions. However, excessive rain causes flower drop and increases the incidence of disease. Dry weather is required for harvesting dry shelled beans.

Beans supply edible immature pods and seeds, ripe seeds and some times leaves are used as vegetables. They are known for their high protein content as a source of food. Thus, Purseglove (1968) reported the chemical composition of beans to be 85.2% water, 6.1% protein, 0.2% fat, 6.3% carbohydrates, 1.4% fibre and 0.8% ash. This prompted Max Milner (1975) to recommend *P. vulgaris* for urgent and immediate attention by all concerned agencies and institutions as possessing the potential for significant contribution to the diets of people in major ecological zones of the developing regions.

Apart from agronomic factors, insect pests also greatly reduce bean yields. Beans are attacked by many species of insect pests, both before and after harvest. Field infestation may be contained by the plants through compensatory effects but damage in the store is often terminal. Some of the major field pests that infest beans are the bean fly (*Ophiomyia phaseoli*) and the black bean aphid (*Aphis fabae*)(Centro Internacional de Agricultura Tropical, 1986). While in the store, the bean seeds are further damaged by coleopterans from the family Bruchidae that results in qualitative and quantitative losses. The main bruchid species that cause severe damage are *Acanthoscelides obtectus* Say and *Zabrotes subfaciatus* Boheman (Centro Internacional de Agricultura Tropical, 1986). However, of these two, *A. obtectus* has been shown to be the major pest of stored dry beans (strong *et al.*, 1968; Schonherr and Mbogua, 1976;

Lugando, 1978; Nyiira, 1978) and it has been reported to destroy beans in various parts of the world (Davies, 1959; McFarlane, 1969; Caswel, 1973; Hill, 1975). Although the bean bruchid is reported to have originated from central and South America, it is now cosmopolitan in distribution (Munro, 1966; Southgate, 1978). It attacks beans starting from the field upto the store. Both species occur in Kenya, but *A. obtectus* is more widespread than the latter (McFarlane, 1969; Caswel, 1973).

The general life cycle of *A. obtectus* has been studied by Hereford (1935)), Howe and Currie (1964) and Centro Internacional de Agricultura Tropical (1986). These authors showed that females lay eggs on green bean pods or through cracks in the drying pods. The eggs may also be laid among beans in the store. Eggs are white in colour and ovoid in shape (plate 1). Fecundity per female depends on among other factors host plant, humidity and temperature. Hill (1975) found that a female bruchid laid 40-60 eggs in her life time at 28°C and 70% relative humidity. The eggs hatch into first instar larvae after 5 days.

Larvae are white in colour with a brown head, strong mandibles and rudimentary legs (plate 2). After wondering for a while on the surface of the bean, the larvae bore into them leaving behind a hole that cannot be detected easily. While inside the seed, the larvae feed and moult into the last instar in about 3-5 weeks. Feeding and pupal cells become visible externally on the seed as windows". The larva reduces the thickness of the window by first eating away the under surfaces of the testa and secondly by eating more around the circumference of this "window" (Hill, 1975). Before pupation, the larva lies with the mandibles facing the "window" and the prepupal stages are passed through in this position. Pupae (plate 3) develop into mature adults in 5 - 6 days. The adult may remain within the cell for several days before pushing out the window with the head and legs. When adults emerge, they leave the characteristic round "window" on the seed coat, typical of bruchid damage.

Duration of the life cycle varies depending on temperature, relative humidity, and host plant species among other factors (Hill, 1975; Howe and Currie, 1964; Strong *et al.*, 1968). On average, the life cycle takes about five weeks and six to twelve generations can develop in one year (Davies, 1959). Normally adults do not feed but they may consume water or nectar (Centro Internacional de Agricultura tropical, 1986). Adults (plate 4) live for about twelve days before they die.

It is difficult to distinguish between the females and males in this species for their colour and size are almost the same. This difficulty can be overcome by observing the last abdominal segment (Figure 1). In males, the pygidium is vertical and partially seen from the dorsal surface while the female pygidium is oblique and in full view from above (Halstead, 1963; Centro Internacional de Agricultura Tropical, 1986).

Breeding experiments have also been carried out to determine optimal conditions for the development of *A. obtectus*. The optimum temperature and relative humidity for rapid development of the beetles in haricot beans was found by Howe and Currie (1964) to be 30°C and 70 % respectivily. They found that the developmental period was relatively longer for females than males. Moreover, females were heavier than males and the sex ratio was close to 1:1. The optimal and minimal conditions for population increase for *A. obtectus* were therefore subsquently given by Howe (1965) as 27°C - 31°C at minimum relative humidity of 60 %.

The range of host plants attacked by *A. obtectus* have also been investigated by several researchers. Southgate (1978) reported that *A. obtectus* showed host specificity to *P. vulgaris* and *Vicia fabae* as indigenous host species and only bred on *Voandzeia subtterranea* (Bambra groundnut) as an introduced host species. Nevertheless, Olubayo (1980), studying the development of *A. obtectus* on beans, cow pea and pigeon pea seed diets found that there were significant differences in the development of the bruchid in the three pulses. She found that beans and cow peas were better food media for *A. obtectus* than pigeon peas. However, little has been done to compare the development of the bean bruchid in different bean varieties.

Control of the bean bruchid has been carried out using various methods. Davies (1959), McFarlane (1969) and Wearing (1970) recommended dusting with 0.04% gamma B.H.C. dust at 226.8% per 90 kg of beans for the control of the beetles. A mixture of beans with 170.1 g - 226.8 g of several of the dusts namely: colloidal silica, colloidal aluminium, pentasilicate, finely ground diatomite or kaolin and 0.5% technical D.D.T. mixed with diatomite or kaolin can protect the beans from bruchid damage (Parkin and Bills, 1955). Southgate (1978) recommended mixing of malathion with the seed and frequent fumigation of the store with methyl bromide as good control. Gamma B.H.C dust or pyrethrins can also control the bruchid (Hill, 1975). However, lindane and malathion remain the insecticides of choice on grounds of efficacy, availability and cost where there is no resistance (Rowlands, 1976). Vegetable oils have also been shown to offer some protection to stored beans against bruchid attack (Schoonhoven, 1978; Ngaah, 1986). Ngaah (1986) found that castor oil, clove oil, maize germ oil, almond oil, olive oil and mustard oil provided effective control but clove oil gave the best results. The mechanism of control of *A. obtectus* by oils has been attributed to the interference of normal respiration resulting in suffocation (Schoonhoven, 1978). Genetic control of the bean bruchid has also been attempted. For instance, Jermy (1972) reported that

A. obtectus was a good candidate for control by the sterile male technique at least in temperate countries.

Biological control studies on *A. obtectus* have shown promising results. Southgate (1978) reported that *Anisopteromalus calandrae* and *Dinarmus basili* (Hymenoptera: Pteromilidae) parasitised eggs, larval and pupal stages of *A. obtectus*. It has also been reported that mites of *Pyenotes ventriculus* complex can destroy bruchid cultures if they are present and escape detection when material is brought into the laboratory (Hereford, 1935; Southgate, 1978).

A new control method for the bean bruchid that is currently under investigation is the breeding and selection of bean varieties that are resistant or less susceptible to infestation by A. obtectus. Horber (1979) described the mechanisms of host-plant resistance to attack by insect pests; and placed the mechanisms into three categories thus: (i) Non preference; whereby insect pests show negative reaction or avoid totally plants with unsuitable characteristics during search for food, oviposition site or shelter. (ii) Antibiosis; which includes all adverse effects exerted by plants on the insect's biology especially survival, development and reproduction. (iii) Tolerance; which includes all plant responses resulting in the ability to withstand infestation and to support insect populations that would severely damage susceptible plants.

Therefore, plant resistance to insects can be described in terms of insect - host plant interactions. The interactions may be divided into two principle aspects: host selection by the insect and resistance to the insect by the plant (Beck, 1965). Dahms (1972) identified possible criteria used to evaluate plant resistance to insect pests. Some of them are: (i) Determination of the number of adult insects or larvae attracted to a cultivar when given a free choice. (ii) Observation of the comparative effects of forced feeding on plants or cultivars by measuring the length of insect life cycle, mortality, reproductive rates, or moulting.(iii) Determination of the number of surviving insects and progeny produced.

Therefore analyses of the causes of plant resistance must include consideration of behaviour and physiological characteristics of the insect (Beck, 1965). Among insects that lay their eggs on or near plants utilized by the progeny, the first point in the insect host-plant relationship at which the plant may show resistance is resistance to oviposition. Oviposition involves a series of behavioural events. The first component of oviposition behaviour is that of recognition and orientation to the host plant. Deposition of eggs follows and the insect finally departs from the oviposition site. Different plant characteristics may influence the initiation and completion of each of these events. Characteristics that tend to prevent oviposition may do so either by failing to provide the appropriate releasing stimuli for one or more of the behavioural components; or by providing stimuli that inhibit that behavioural release (Beck, 1965).

Feeding behaviour also plays a vital role in insect - host plant interactions. The steps for feeding behaviour include: host plant recognition and orientation, initiation of feeding (biting and piercing), maintenance of feeding and cessation of feeding followed by dispersal. Therefore any plant characteristic that prevents initiation, continuation or completion of each of these events would prevent the insect from utilizing the plant (Beck and Schoonhoven, 1980).

The relationship between bruchids and their host plants has been investigated by various authors. Janzen and Juster (1976) reported that neotropical bruchids are food specific, with the great majority of them feeding on only one species of plant in a given habitat. Bruchids that attack stored seeds show preference for certain species. However, it has been reported that varieties of a plant may show differences in their susceptibility to bruchid attack compared with other species. Infact, oviposition may be affected within a sample of a single variety by apparently small differences of surface smoothness, by plumness of the seed coat and perhaps by the size of and hardness of the seed (Howe and Currie, 1964). The testa may also be so thick that the newly hatched larvae die before they reach the cotyledon. Some larvae may also fail to escape from the seed after pupation.

P. vulgaris has been shown to resist attack by bruchids. Applebaum *et al.* (1970) isolated a heteropolysaccharide and starch from *P. vulgaris* and incorporated them into artificial beans. They found that the fractions, decreased the rate of larval development and the number of eggs laid by *Callosobruchus chinensis*. From these findings,they concluded that the heteropolysaccharide fraction as well as the starch were part of a complex of natural components of *P. vulgaris* seeds that make them resistant to attack by *C. chinensis*.

It has also been found that black bean phytohemagglutinin killed larvae of *Callosobruchus maculatus* when incorporated in the normal diet (Janzen and Juster, 1976). Because *C. maculatus* normally feeds on phytohemagglutinin-free cow pea (*Vigna unguilata*) they concluded that phytohemagglutinins protect bean seeds from attack by insect predators.

Studies on interactions between *P. vulgaris* and *A.obtectus* show that beans regulatethe reproductive activity of *A. obtectus* females. In the absence of beans, female *A. obtectus* do not lay eggs but retain 30 to 40 oocytes in their lateral oviducts (Huignard, 1974). Presence of beans induces oviposition in the majority of females. Furthermore, Jermy (1974) reported that, in *A. obtectus* signals coming from beans are picked by sensory organs and they probably regulate the reproductive function through the nervous system and haemolymph.

Various species of *Phaseolus* possess resistance to attack by *A. obtectus*. Centro Internacional de Agricultura Tropical (1980) and Schoonhoven *et. al*. (1983) found that the effects of this resistance consisted of increased mortality, low adult weight and reduced oviposition. Dobie *et. al*. (1990) screened four species of Phaseolus namely: *Phaseolus vulgaris* L., *Phaseolus lunatus* L., *phasoelus coccineus* L. and *Phaseolus acutifolius* Gray for resistance to attack by *A. obtectus*. They found that two varieties of *P. vulgaris* were resistant. In one, resistance was associated with a lectin - like protein (LLP) and in the other resistance was linked to the presence of a novel protein similar but not the same as LLP. They also found that five varieties of *P. lunatus* and six of *P. acutifolius* were resistant to attack by *A. obtectus* and the potential is recognized for transferring resistant genes to *P. vulgaris* via interspecific crosses.

Studies have also been carried out to compare inherent resistance or susceptibility of crops to weevil attack during storage. Such comparative studies were done on maize varieties by Wheatley (1973) and Dobie(1974), and on sorghum by Davey(1965). Wheatley(1973) and Dobie(1974) have given the parameters used for calculating susceptibility indices of different maize varieties when artificially infested with

Sitophilus zeamais Motsch. The same parameters were used on cow pea varieties when artificially infested by *C. maculatus* by Osuji (1986). Essentially, the susceptibility index of a crop with respect to a pest species may be given by the relationship

$$\textbf{SUSCEPTIBILITY INDEX} = \frac{\text{Log e Y}}{\text{T}}$$

Where Y is the number of progeny of the insect and T the duration in days taken by 50 % of progeny to emerge (Dobie, 1974; Osuji, 1976)

Differences in susceptibility to attack by *A. obtectus* among bean varieties has been reported in Kenya (Lukando, 1978, Rheenem et al. 1983). Lukando (1978) attributed the differences in susceptibility to the thickness of testa, presence of oxalate crystals in the testa and alkaloids.

In this study, it was considered necessary to investigate the interactions between *A. obtectus* and six Kenyan bean varieties and to quantify and document the extent of the differences in susceptibility to attack bythe same pest.

1.30 OBJECTIVES

The objectives of this study were as follows:

1. To investigate the effects of bean varieties on oviposition behaviour of *A. obtectus*.
2. To investigate the effects of bean varieties on fecundity of *A. obtectus*.

3. To investigate the effects of delayed bean provision on oviposition of *A. obtectus*
4. To assess the susceptibility of the various bean varieties to attack by *A. obtectus*.

5. To investigate the physical factors that are likely to affectsusceptibilityof the bean varietiesto attack by *A. obtectus*.

CHAPTER TWO

2.10 MATERIALS AND METHODS

2.11 GENERAL MATERIALS AND METHODS

Six bean varieties were used for the study namely: red haricot (plate 5), mwitemania (plate 6), rose coco (plate 7), nyayo (plate 8), canadian wonder (plate 9) and mwezi moja (plate 10). These were obtained from the local market. The varieties were identified with the help of the Horticultural Research Centre, Kenya. Since bean varieties are known by many names depending on the locality in which they are grown, the varieties used in this study were given code names commonly used by plant breeders in Kenya and their physical appearances described (Table 3).

Bean seeds were pre-conditioned to the experimental conditions for one week before being used. Before the start of the experiments, it was necessary to determine moisture contents of the seeds, to obtain proper experimental conditions and to obtain enough bruchids. Methods dealing with specific experiments are described under specific headings.

Determination of the moisture content of the bean seeds was considered important because moisture content affects growth and development of bruchids that feed on stored seeds. For average growth and development, most of them prefer seeds with about 7 - 12 % moisture content (Munro, 1966).

Moisture contents of the seeds were determined using the oven method (Hall, 1970). Fifty grams of bean seed of each variety was ground using an electric grinding machine at constant speed for 3 minutes. The ground powder was then placed in watch glasses. Each variety had six replicates. The ground powder was weighed and dried in an oven at 60°C for 48 hours. The watch glasses and contents were removed, cooled in a dessicator and re-weighed. The difference in weight was calculated as the percentage moisture content. The percentage moisture content of the seeds ranged between 8.85 % and 10.24 %. Temperature of the experimental room was maintained at 27 ± 1°C using a heater and a lamp (150 watts). Relative humidity was maintained at 70 ± 5 % using a fan and a water bath.

Samples of *A. obtectus* used were obtained from National Agricultural Laboratories Nairobi, Kenya. Strong's (1968) method was used to rear *A. obtectus* .The beetles were reared in a mixture of the six varieties to ensure that they did not get conditioned to one variety. 750 g of the mixture and 200 unsexed one day old beetles were placed in four 1000ml plastic containers and kept in the experimental room. Beetles were left for two weeks after which they were removed by sieving, leaving larvae to develop. Four such containers were established at intervals of one month to maintain a supply of young beetles for the experiments.

For the purpose of the experiments, plastic containers

(250, 500 and 1000 ml) and pill boxes were used. These were obtained from commercial suppliers. Small holes were made on the lids of the containers to allow for free circulation of air.

EXPERIMENT 1: INVESTIGATION OF THE EFFECTS OF BEAN VARIETIES ON OVIPOSITION SITE PREFERENCE BY *A.obtectus*.

Adults of *A. obtectus* do not usually feed. It is only the larval stage that feeds. Adults choose suitable media that would be used as food by the larvae and that would ensure that development is successful to the adult stage. In these experiments, effect of different bean varieties on oviposition site preference by *A. obtectus* was investigated.

In part one of the experiment 30 g of each bean variety were placed in pill boxes. Each variety had two replicates. A petri - dish (radius 4.2 cm and height 1 cm) was placed at the centre of a glass cage. The glass cage was to prevent the bruchids from escaping. The pill boxes were placed at distances 0, 4 and 8 cm from the petri-dish. The pill boxes were arranged in such a way that each variety occupied each of the distances (Figure 2a, b and c). 300 unsexed one day old beetles and damaged bean seeds were placed in the petri-dish. The petri - dish was to act as a source of infestation for the bean varieties. Beetles could enter or leave the petri-dishes at will. The cage was placed in the experimental room. After 24 hours, the number of beetles which had settled in each pill box were counted. This procedure was repeated six times to yield seven replicates for each distance. The experimental design had three sets of arrangements as shown in figures 2a, b and c. This was to ensure that the varieties faced all possible directions to avoid effects of direction and light.

In the scond part of the experiment, *A. obtectus* was provided with a mixture of all six bean varieties. Eighty seeds of each variety were placed in 250 ml plastic containers. Twenty female and ten male (one day old) beetles were introduced into the containers. The experiments had ten replicates. The containers were left in the experimental room until all adults of the next generation emerged. The number of emergence holes on each seed of bean variety were counted.

2.13 EXPERIMENT 2: INVESTIGATION OF THE EFFECTS OF BEAN VARIETIES ON FECUNDITY OF *A. obtectus*

Beans are the most preferred oviposition site for *A. obtectus* (Szentesi, 1974). This experiment was set up to investigate whether the number of eggs laid by A. obtectus is affected by different bean varieties.

For each variety, 20 g of seed were placed in a pill box. One female and one male (one day old) beetles were introduced into the pill boxes. Each bean variety had ten replicates. Number of eggs laid by the beetles were counted after 24 hours since the introduction of adults. Counting continued until all beetles had died. Similarly, the egg laying duration of each female was also noted.

2.14 EXPERIMENT 3: INVESTIGATION OF THE EFFECTS OF DELAYED BEAN (FOOD) PROVISION ON OVIPOSITION OF *A. obtectus*

It has been found that beans regulate the reproductive activity in *A. obtectus* and that presence of beans induce oviposition in the majority of females (Huignard, 1974).

An experiment was set up to find out the effects of bean varieties on egg laying behaviour in *A. obtectus* when provision of bean varieties was delayed. Four females and two males (one day old beetles) were placed in a pill box which was replicated 122 times. Thirty grams of seeds from each variety were introduced into two pill boxes successively after 0, 1, 2, 4, 6, 8, 10, 12, 14 and 16 days. The number of eggs laid on each variety were counted at intervals of 2 days until all the beetles died. For the control, 14 of the pill boxes with beetles in them were left without beans up to the end of the experiment. The number of eggs laid by the beetles in these pill boxes without beans were also counted.

2.15 EXPERIMENT 4: ASSESSMENT OF THE SUSCEPTIBILITY OF BEAN VARIETIES TO ATTACK BY *A. obtectus*

This experiment was set up to test whether there is a difference in susceptibility of different varieties of beans when artificially infested with *A. obtectus*. For each variety, 250 g of bean seed were mixed with 20 females and 10 males (one day old adult beetles) in 500 ml plastic containers. Each variety had ten replicates.

Containers were kept in the experimental room. After 30 days, emergence of progeny was checked. When the first progeny emerged, they were counted and discarded. The process of counting continued until emergence of progeny stopped. The number of progeny and duration in days taken by 50% of the progeny to emerge was used to calculate an index of susceptibility for each variety.

2.160 EXPERIMENT 5: INVESTIGATION OF THE PHYSICAL FACTORS THAT ARE LIKELY TO AFFECT SUSCEPTIBILITY OF BEAN VARIETIES TO ATTACK BY *A. obtectus*

The experiments were set up to investigate some of the physical factors that are likely to affect susceptibility of bean varieties to attack by *A. obtectus*. The relationship between susceptibility and hardness of seed, thickness of testa and size of seed were considered.

2.161. HARDNESS OF SEED

Dobie's (1974) method was used to determine seed hardness. Hundred grams of each bean variety were ground using an electric grinding machine at constant speed for 3 minutes. Five grams of powdered seed from each variety was weighed and placed in dry beakers. The beakers were then sealed with aluminium foil to avoid absorption of moisture. The procedure was repeated nine times for each variety to yield ten replicates. The powdered seed was then sieved with a sieve of aperture size of 500 microns. Fractions that were retained and those that passed through the sieve aperture were weighed separately.

2.162. THICKNESS OF TESTA

Testae of bean varieties were pealed out using a scalpel. The thickness of the pieces of testae pealed out were measured using a micrometer screw gauge. Each variety had 10 replicates.

2.163. SIZE OF SEED

Surface area was used as a measure of seed size. Ten seeds from each variety were picked at random. The seeds were soaked in water to facilitate removal of testa. Then the testa of each seed was pealed out and dried. The testae were then spread on a graph paper and the surface area estimated by counting the number of squares the testae occupied on the graph paper.

At the end of these experiments, the relationship between hardness of seed, thickness of testa and size of seed and susceptibility of these varieties to attack by the beetles was tested statistically using correlation analysis

(Sokal, 1973).

Plate 1. Eggs of *Acanthoscelides obtectus* (X 100)

Plate 2. Larvae of *Acanthoscelides obtectus* (X 10)

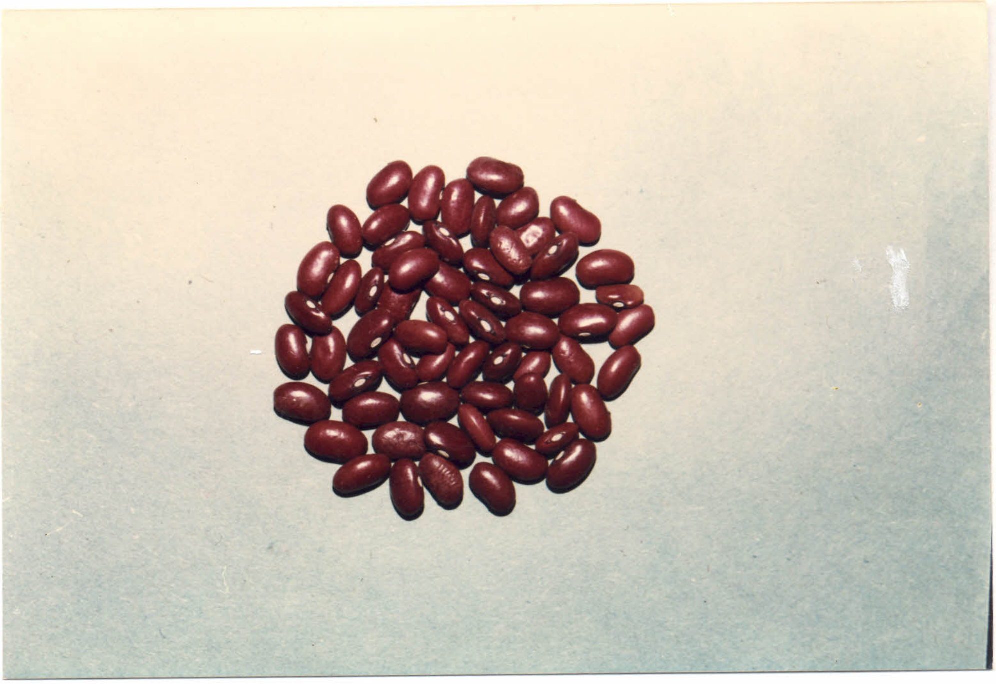
Plate 3. Pupae of *Acanthoscelides obtectus* (X 10)

Plate 4. Adult *Acanthoscelides obtectus*
(X 4)

* Plate 5. Red haricot (GLP 585)

* Plate 6. Mwitemania (GLP 92)

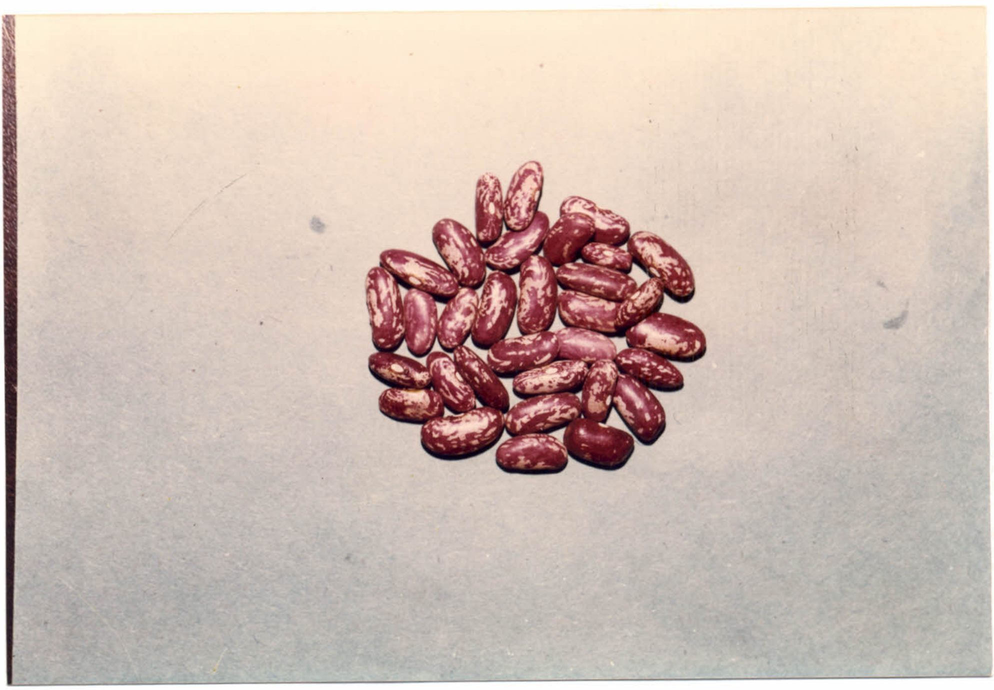
* Photographs were taken on 4th July, 1992



* Plate 7. Rose coco

* Plate 8. Nyayo (GLP 2)

* Photographs were taken on 4th July, 1992



* Plate 9. Canadian wonder (GLP 24)

* Plate 10. Mwezi moja (GLP 1004)

* Photographs were taken on 4th July, 1992

CHAPTER THREE

3.10 RESULTS

3.11 EFFECTS OF BEAN VARIETIES ON OVIPOSITION SITE PREFERENCE BY *A. OBTECTUS*

Table 4 shows mean percentage distribution of *A. obtectus* on bean varieties. Red haricot had the lowest percentage (11.40 ± 1.1 %) and mwezi moja the highest (23.33 ± 2.6 %). Percentage distribution was significantly different between varieties (F = 8.13, d.f = (5, 36), $P < 0.05$). Canadian wonder was different from red haricot and mwitemania. Mwezi moja was also different from all varieties except canadian wonder (Tukey multiple range test).

Figure 3 shows the percentage distribution of *A. obtectus* on bean varieties at distances 0, 4, and 8 cm from source of infestation. At 0 and 4 cm from source of infestation, red haricot had the lowest percentage and mwezi moja the highest (figure 3). While at 8 cm, nyayo had the lowest percentage (3.20 %) and mwezi moja the highest (5.80 %). Percentage distribution decreased from 0 to 8 cm in all varieties except red haricot and rose coco. Table 5 shows the mean number of emergence holes of *A. obtectus* on bean seeds of the different bean varieties. Number of emergence holes were significantly different between varieties (Kruskal wallis test: x^2 = 31.5, d.f = 5, $P < 0.05$). Canadian wonder had the highest number (66.4 ±

10.4) of emergence holes and mwitemania the lowest (33.1 ± 6.1).

Figure 4 shows number of seeds of bean varieties with 1, 2, 3 and more than 4 emergence holes of *A. obtectus* per seed. The number of seeds with these number of emergence holes per seed were significantly different between varieties (x^2 = 11.47, d.f = 5, P < 0.05). Maximum number of emergence holes per seed was 10 in red haricot, 6 in mwitemania, 8 in rose coco, 14 in nyayo, 11 in canadian wonder and 20 in mwezi moja.

3.12 EFFECTS OF BEAN VARIETIES ON FECUNDITY OF *A. OBTECTUS*

Figure 5 shows mean number of eggs laid by *A. obtectus* per day on different bean varieties. Average number of eggs laid per day was highest on mwezi moja and lowest on mwitemania (table 6). Number of eggs laid per day on different bean varieties was significantly different between varieties (Kruskal wallis test: x^2, 36.9, d.f = 5, P < 0.05).

Figure 6 shows mean cumulative number of eggs laid by *A. obtectus* on different bean varieties. Highest number of eggs laid was on mwezi moja and the lowest on mwitemania (table 6). Number of eggs laid were significantly different between varieties (Kruskal wallis test: x^2 = 32.1, d.f = 5, P < 0.05). Maximum number of eggs laid was 70 in red haricot, 63 on mwitemania, 57 on rose coco, 83 on nyayo, 67 on canadian wonder and 60 on mwezi moja repectively.

Egg laying duration (days) was 7.1 ± 0.3 on red haricot, 7.6 ± 0.4 on mwitemania, 7.4 ± 0.4 on rose coco, 7.9 ± 0.3 on nyayo, 7.6 ± 0.4 on canadian wonder and 8.0 ± 0.4 on mwezi moja. Egg laying duration was shortest on red haricot and the longest on mwezi moja. Egg laying duration was not significantly different between varieties ($F = 0.62$, d.f =(5, 54), $P > 0.05$).

3.13 EFFECTS OF DELAYED BEAN (FOOD) PROVISION ON OVIPOSITION OF *A. OBTECTUS*

Figure 7 shows mean number of eggs laid by female *A. obtectus* per day on different bean varieties when the provision of beans was delayed. Generally, the number of eggs laid decreased with the delay in bean provision. Mean number of eggs laid by a female per day without beans (control) was 0.6 ± 0.4 eggs.

Figure 8 shows mean number of eggs laid per day when bean provision was delayed for between 0 and 16 days. Number of eggs laid were significantly different between varieties ($x^2 = 77.7$, d.f = 5, $P < 0.05$). Average number of eggs laid when bean provision was delayed for sixteen days was 7.0 on red haricot, 6.5 on mwitemania, 7.9 on rose coco, 7.7 on nyayo, 9.5 on canadian wonder and 15.1 on mwezi moja.

Figure 9 shows the egg laying duration (days) when bean provision was delayed for between 0 - 5, 6 - 10, and 11 - 16 days. Egg laying duration was prolonged and was significantly different between varieties (x^2 = 49.3, d.f = 5, $P < 0.05$).

3.14 ASSESSMENT OF THE SUSCEPTIBILITY OF BEAN VARIETIES TO ATTACK BY *A. OBTECTUS*

Figure 10 shows mean number of progeny of *A. obtectus* emerged from different bean varieties per day after emergence of the first progeny. First progeny emerged around the 31st day after introduction of adult beetles into bean seeds. Peak emergence of progeny was on the 37th day in red haricot, 38th day in mwitemania, 37th day in rose coco, 39th day in nyayo, 37th day in canadian wonder and the 35th day in mwezi moja (Figure 10).).

Figure 11 shows mean cumulative number of progeny of *A. obtectus* emerged from bean varieties and table 8 shows the mean total number of adults that emerged from each variety. Mwezi moja had the highest number of progeny (490.4 ± 40.1) emerging from it and mwitemania the least (299.7 ± 37.9). Total number of adults emerged were significantly different between varieties (Kruskal wallis test: x^2 = 77.0 d.f = 5, $P < 0.05$).

Table 8 shows the duration (days) taken by 50% of the progeny of the beetles to emerge from the different bean varieties. Duration was longest (39.2 ± 0.54 days) in red haricot and shortest (36.6 ± 0.19) in mwezi moja. Duration was significantly different between varieties (F = 6.55, d.f

= (5, 54), $P < 0.05$). Mwezi moja was different from all varieties except from rose coco. Red haricot was different from rose coco (Tukey multiple range test).

Calculated susceptibility indices of the bean varieties are shown in table 8. Mwezi moja had the highest (5.81 ± 0.48) susceptibility index and mwitemania the lowest (3.36 ± 0.42). Susceptibility indices were significantly different between varieties ($F = 3.33$, d.f = (5, 54), $P < 0.05$). Mwezi moja was different from mwitemania (Tukey multiple range test).

3.150 PHYSICAL FACTORS THAT ARE LIKELY TO AFFECT SUSCEPTIBILITY OF BEAN VARIETIES TO ATTACK BY *A. OBTECTUS*

3.151 HARDNESS OF SEED

Bean seed particle sizes of more than 500 microns were used as a measure of seed hardness. It was convenient to weigh these particle sizes as compared to those less than 500 microns because they were not sticking on containers and weighing apparatus.

Table 9 shows mean weight (g) of particle sizes more than 500 microns. Canadian wonder had the highest weight (1.91 ± 0.04 g) and mwezi moja had the lowest (1.18 ± 0.03 g). The weight was significantly different between varieties ($F = 30.6$, d.f =(5, 54), $P < 0.05$). Canadian wonder and mwezi moja were different from all varieties (Tukey multiple range test). This meant that canadian wonder had the hardest seeds and mwezi moja the softest.

3.152 THICKNESS OF TESTAE

Table 9 shows mean thickness of testae (mm) of the different bean varieties. Mwitemania had the thickest (0.120 ± 0.040 mm) testa whereas rose coco had the thinnest (0.090 ± 0.002 mm) testa. Thickness in testa was significantly different between varieties ($F = 15.18$, $d.f = (5, 78)$, $P < 0.05$). Canadian wonder and mwitemania were different from all varieties (Tukey multiple range test).

3.153 SIZE OF SEED

Surface area of seeds was used as a measure of seed size. Table 9 shows mean surface areas of seeds of the different bean varieties . Mwezi moja had the largest surface area (278 ± 2 mm^2) and red haricot the smallest (127 ± 1 mm^2). Surface area was significantly different between varieties ($F = 339.7$, $d.f = (5, 54)$, $P < 0.05$). All varieties were different from each other (Tukey multiple range test).

Statistcal analysis of the results using correlation analysis showed that susceptibility was not significantly correlated with hardness of seed, seed size and thickness of testa in all varieties except canadian wonder ($0.524 > r > 0.001$, $d.f = 8$, $P > 0.05$). However, susceptibility was significantly correlated with seed size in canadian wonder ($r = 0.720$, $d.f = 8$, $P < 0.05$). Combined effect of hardness of seed, seed size and thickness of testa was not significantly correlated with susceptibility in all varieties except canadian wonder (0.678 > r > 0.333, d.f = 6, P > 0.05). In canadian wonder, the combined effect of seed size, seed hardness and thickness of testa were significantly correlated with susceptibility (r = 0.750, d.f = 6, P < 0.05).

Table 1. Calories and some nutrients in grain legumes compared with other food types (figures relate to edible protein).

	H_2O	Ca/100g	% Protein	% Fat	% CHO	Ca mg/100g	Iron mg/100g	Thiamine mg/100g	Riboglavine mg/100g	Nicotinic acid mg/100g
Rice (milled white)	13	360	6.7	0.7	78.9	10	0.9	0.08	0.03	1.6
Wheat (low extraction)	12	370	10.9	1.1	75.5	16	10	0.13	0.04	1.1
Maize (90-96% extraction)	12	360	9.3	4.0	73.5	6	1.8	0.35	0.09	1.3
Cassava (flour)	14	338	1.5	0.6	81.5	12	1.0	-	-	1.0
Beans (P. *Vulgaricus*)	11	341	22.1	1.7	61.4	137	6.7	0.54	0.18	2.1
Lentil (*L. esculenta*)	11	346	24.4	1.6	60.8	56	6.1	0.50	0.21	1.8
Cowpea (*V. siensis*)	11	342	23.4	1.8	60.8	76	6.7	0.92	9.18	1.9
Groundnut (*A. hypogea*)	5	346	25.6	4.3	23.4	52	1.9	0.84	0.12	16.0
Soyabean (*G. max*)	8	335	38.0	18.0	31.3	208	6.5	1.03	0.30	2.1
Beef (lean)	67	198	19.0	13.0	-	11	2.3	0.07	0.17	4.0
Fish (White sea)	82	75	16.4	0.5	-	25	0.7	0.05	0.08	2.2
Egg (hen)	74	163	12.4	11.7	0.9	50	2.5	0.10	0.30	0.1
Milk (skimmed)	4	360	36.0	1.0	51.0	123	0.9	0.35	1.80	1.0
Pigeon pea (C. *cajan*)	11	343	20.9	1.7	62.9	129	5.8	0.50	0.14	2.3

Source - Legumes in human nutrition. FAO Nutritional Studies. 1964 No.19

Table 2. Estimated area under some grain legumes in Kenya for the 1976/77 crop year (X 1000 hectares).

Province	Beans	Pigeon Peas	Cow Peas	Grams	Total
Eastern	211.2	72.1	90.3	36.4	410.0
Central	97.9	0.4	1.9	3.1	103.3
Nyanza	60.0	-	-	1.2	61.2
Western	85.0	-	-	0.2	85.2
Rift Valley	40.0	0.1	-	0.1	40.2
Coast	5.6	0.9	16.9	11.9	35.1
TOTAL	499.7	73.5	108.9	52.9	735.0

Source: Ministry of Agriculture, Provincial Annual Reports (unpublished)

Table 3. Description of seeds of bean varieties used for the study.

Variety	code name	colour	* surface area (mm^2)
Red haricot	GLP 585	maroon with shiny testa.	127 ± 1
Mwitemania	GLP 92	cream with brown spots.	172 ± 2
Rose coco	-	brown with dark brown spots.	195 ± 3
Nyayo	GLP 2	maroon with cream spots.	221 ± 3
Canadian wonder	GLP 24	dark brown with a shiny testa.	259 ± 3
Mwezi moja	GLP 1004	purple with cream spots.	287 ± 2

* $\bar{X}$ ± S.E (and is also referred to thereafter in other parts of the text)

Table 4. Mean percentage number of *A. obtectus settled* on bean varieties when provided with a random choice of bean varieties.

VARIETY	MEAN % NUMBER OF ADULTS SETTLED ON BEAN VARIETIES (x ± S.E)
Red haricot	11.4 ± 1.1*
Mwitemania	14.0 ± 1.3[ab]
Rose coco	15.5 ± 0.9
Nyayo	16.2 ± 1.2
Canadian wonder	19.6 ± 1.8
Mwezi moja	23.3 ± 2.6*[b]

**, [aa], [bb] denotes pairs that were significantly different, F = 8.13, d.f = (5, 36), $P < 0.05$

(Appendix 1)

Table 5. Mean number of emergence holes of *A. obtectus* on seeds of bean varieties after the beetles were provided with a mixture of all the varieties.

VARIETY	NUMBER OF EMERGENCE HOLES($x \pm$ S.E)
Mwitemania	$33.1 \pm 6.1^{*}$
Red haricot	50.3 ± 8.5
Mwezi moja	52.0 ± 6.6
Rose coco	52.6 ± 8.5
Nyayo	63.4 ± 9.4
Canadian wonder	$66.4 \pm 10.4^{*}$

** denotes a pair that was significantly different (Kruskal wallis test, $X^2 = 31.5$, d.f $= 5$, $P < 0.05$)

Table 6. Average number of eggs laid by *A. obtectus* per day and mean number of eggs laid in life time on bean varieties.

VARIETY	AVERAGE NO. OF LAID/DAY	NO. OF EGGS LAID EGGS LAID IN LIFE TIME
Mwitemania	4.0	35.4 ± 5.2*
Nyayo	4.6	42.3 ± 5.3
Red haricot	4.9	44.5 ± 3.8
Canadian wonder	5.0	45.1 ± 4.7
Rose coco	5.1	46.1 ± 2.9
Mwezi moja	6.0	54.6 ± 4.7*

** denotes a pair that was significantly different (Kruskal wallis test, $X^2 = 69.9$, d.f = 5, $P < 0.05$)

Table 7. Mean number of progeny of *A. obtectus* that emerged from bean varieties, duration (days) taken by 50 % of progeny to emerge and susceptibility indices of beanvarieties

VARIETY	NO. OF PROGENY (Y)	DURATION (T)	SUSCEPTIBILITY INDEX $\frac{(\text{Log e Y})}{T}$
V1	299.7 ± 37.9[*]	38.6 ± 0.17	3.36 ± 0.42[c]
V2	395.0 ± 47.8	39.2 ± 0.54	4.38 ± 0.54
V3	429.0 ± 40.1	38.8 ± 0.44	4.40 ± 0.38
V4	430.8 ± 34.5	37.4 ± 0.36[a]	4.99 ± 0.40
V5	436.5 ± 38.1	38.2 ± 0.30	4.95 ± 0.43
V6	490.4 ± 40.1[*]	36.6 ± 0.19[b]	5.81 ± 0.48[c]

V1 = MWITEMANIA V2 = RED HARICOT

V3 = NYAYO V4 = ROSE COCO

V5 = CANADIAN WONDER V6 = MWEZI MOJA

[**] denotes a pair that was significantly different (Kruskal wallis test, $X^2 = 77$, d.f = 5, $P < 0.05$)

[a] was significantly diferrent from others except [b] ($F = 6.55$, d.f (5,54), $P < 0.05$))

[cc] denotes a pair that was significantly different, $F = 3.33$, d.f =(5,54), $P < 0.05$ (Appendix 2)

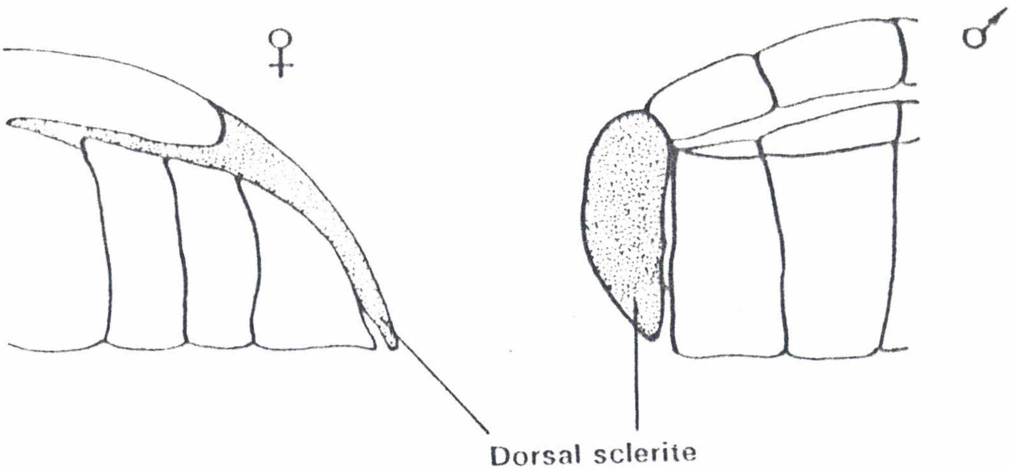
Table 8. Mean weight (g) of ground seed particles more than 500 microns, thickness of testae (mm) and surface areas (mm^2) of seeds of bean varieties.

VARIETY	WEIGHT(g)	THICKNESS(mm)[a]	SURFACE AREA (mm^2)[b]
Mwezi moja	1.18 ± 0.03*	0.100 ± 0.002	278 ± 2
Red haricot	1.47 ± 0.02	0.102 ± 0.002	127 ± 1
Mwitemania	1.52 ± 0.40	0.120 ± 0.040	172 ± 2
Rose coco	1.56 ± 0.06	0.090 ± 0.002	195 ± 3
Nyayo	**1.62 ± 0.02**	**0.100 ± 0.002**	**221 ± 3**
Canadian wonder	1.91 ± 0.04*	0.115 ± 0.002	259 ± 3

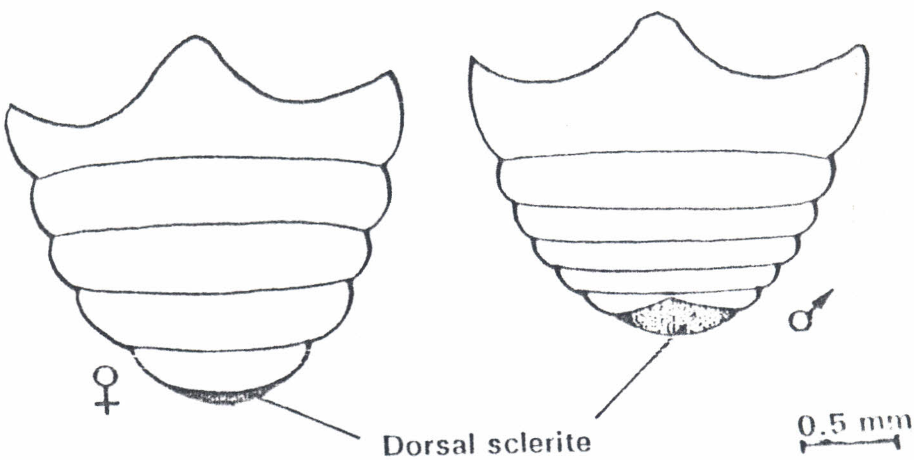
* denotes means that were significantly different from others, $F = 30.6$, d.f = (5, 54), $P < 0.05$

[aa] denotes a pair that was significantly different from others , $F = 15.2$, d.f = (5, 78), $P < 0.05$ (Appendix 3)

[b] all means were significantly different from each other, $F = 339.7$, d.f = (5, 54), $P < 0.05$



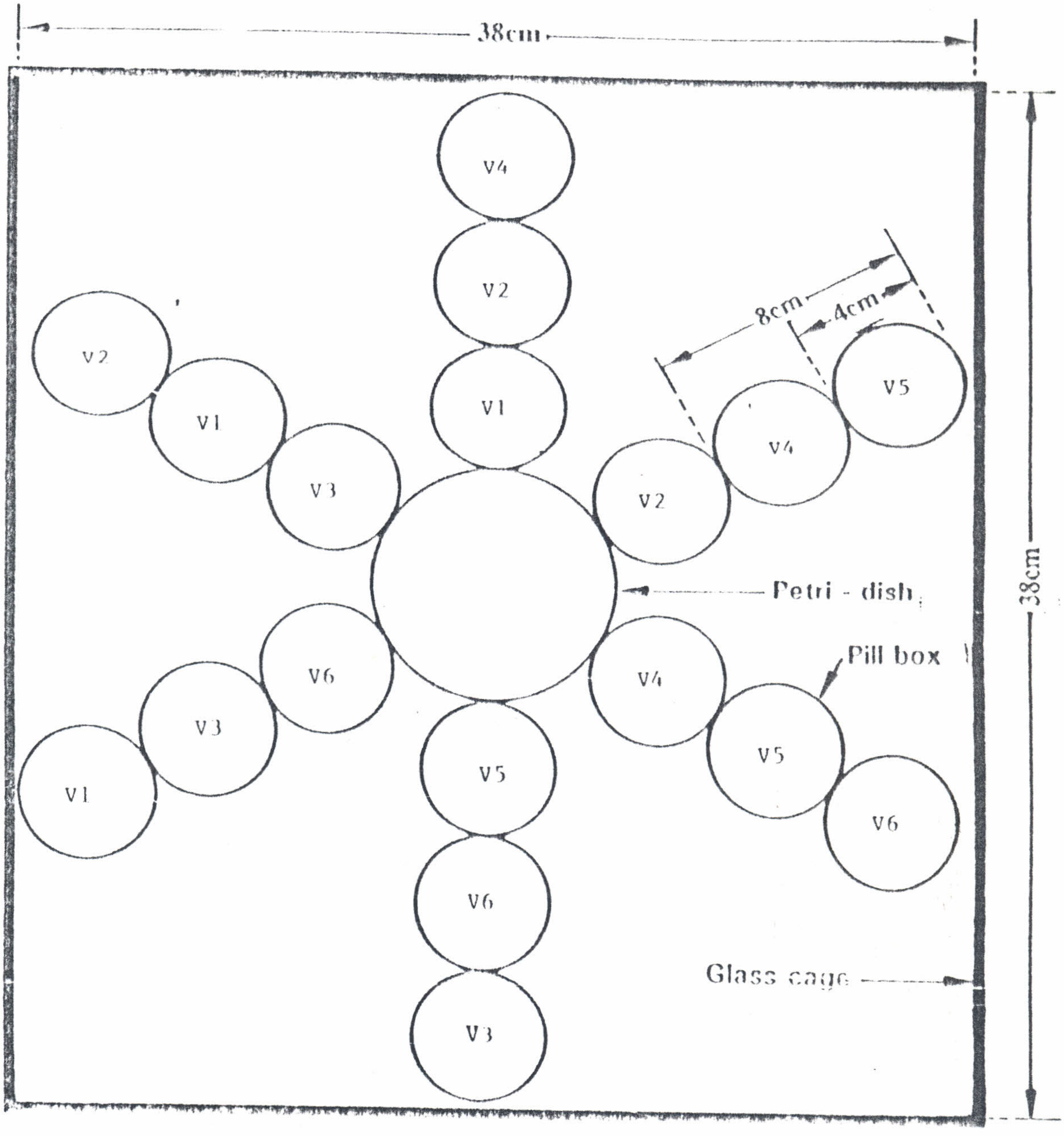
a) Laterial view (after Halstead, 1963)



b) Dorsal view (after Centro Internacional de Agricultura tropical, 1986)

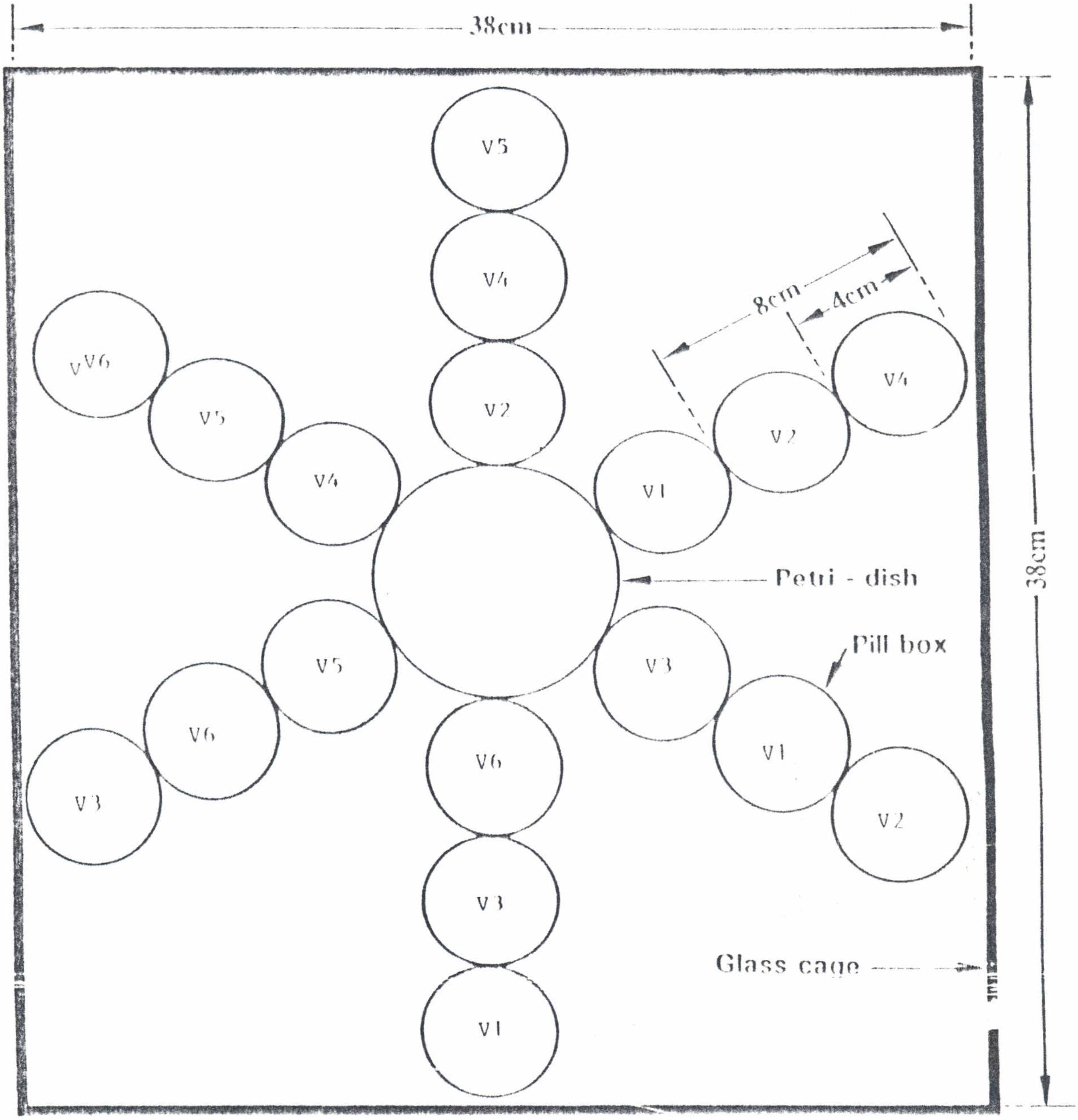
Fig. 1. Lateral and dorsal views of the last abdominal segments of *A. obtectus*.

Fig. 2. The experimental design to investigate bean varietal preference by *A. obtectus*. The petri-dish was the source of infestation. 18 Pill boxes containing 6 varieties of beans were arranged as shown.



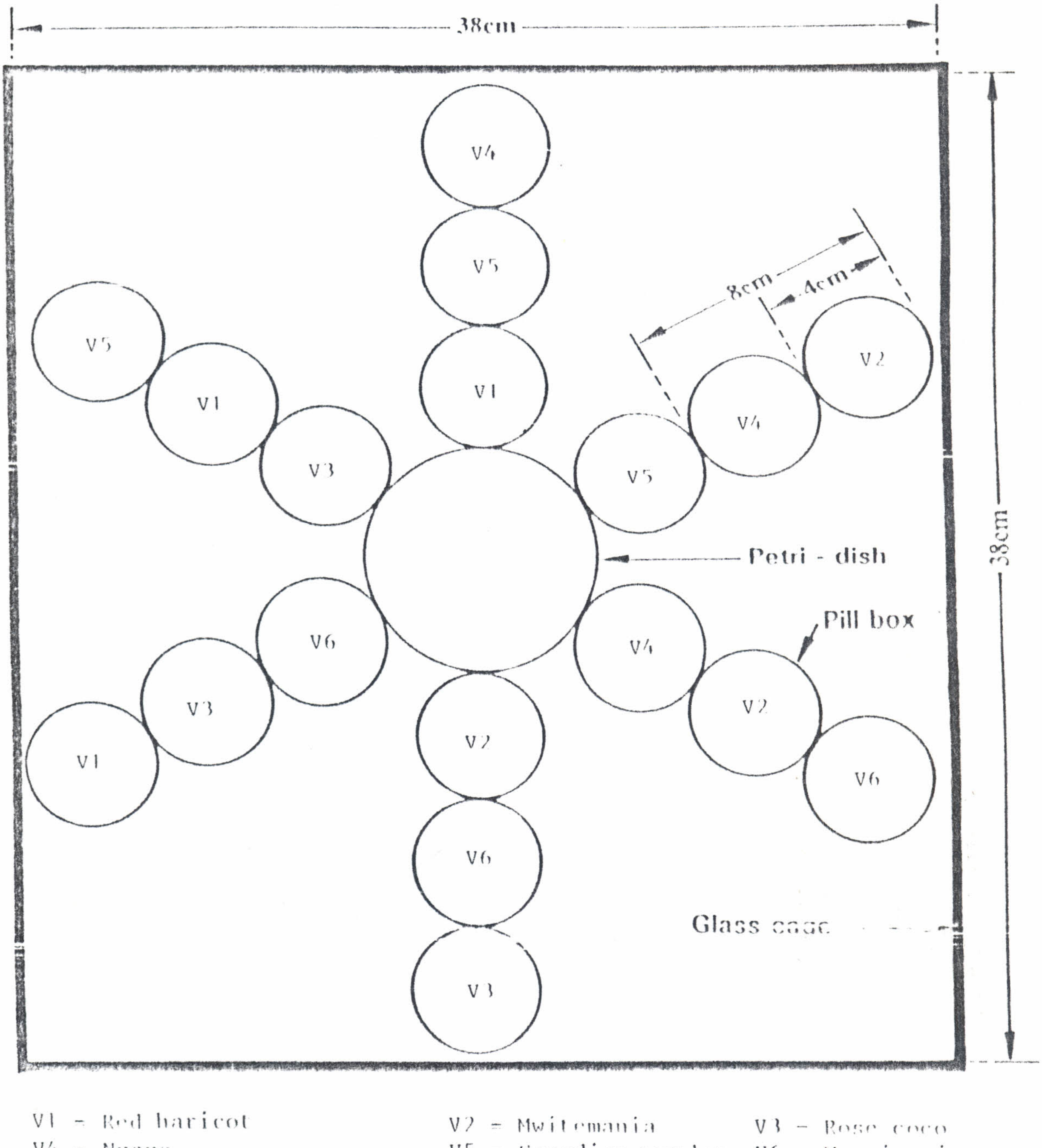
V1 = Red haricot
V4 = Nyayo
V2 = Mwitemania
V5 = Canadian wonder
V3 = Rose coco
V6 = Mwezi moja

2a. First set of arrangement



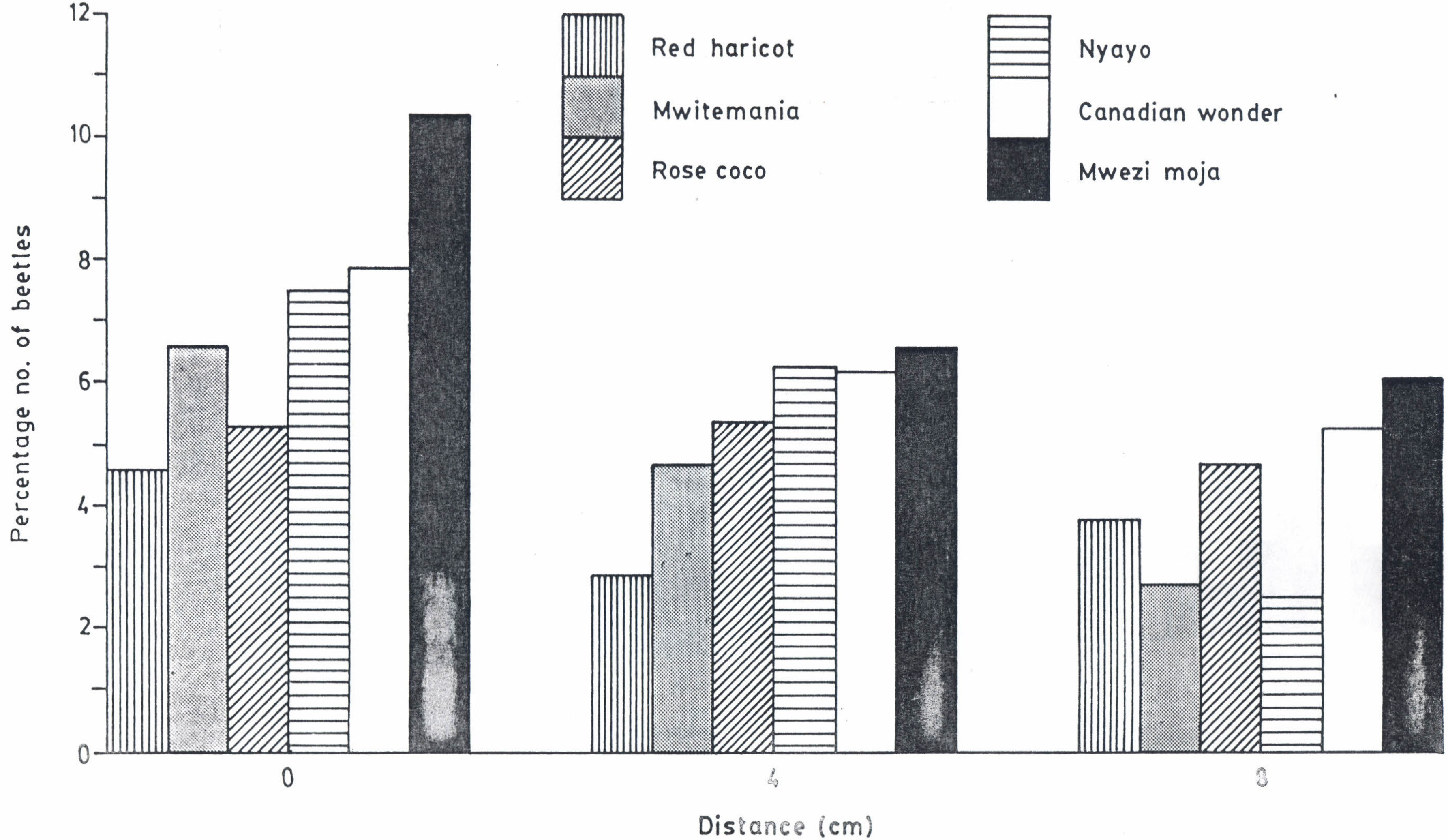
V1 - Red haricot V2 = Mwitemania V3 - Rose coco
V4 - Nyayo V5 = Canadian wonder V6 - Mwezi moja

Fig. 2b. Second set of arrangement



V1 - Red haricot V2 = Mwitemania V3 - Rose coco
V4 - Nyayo V5 = Canadian wonder V6 - Mwezi moja

Fig. 2c. Third set of arrangement


12
10
8
6
4
2
0
Percentage no. of beetles
Red haricot
Mwitemania
Rose coco
Nyayo
Canadian wonder
Mwezi moja
0
4
8
Distance (cm)

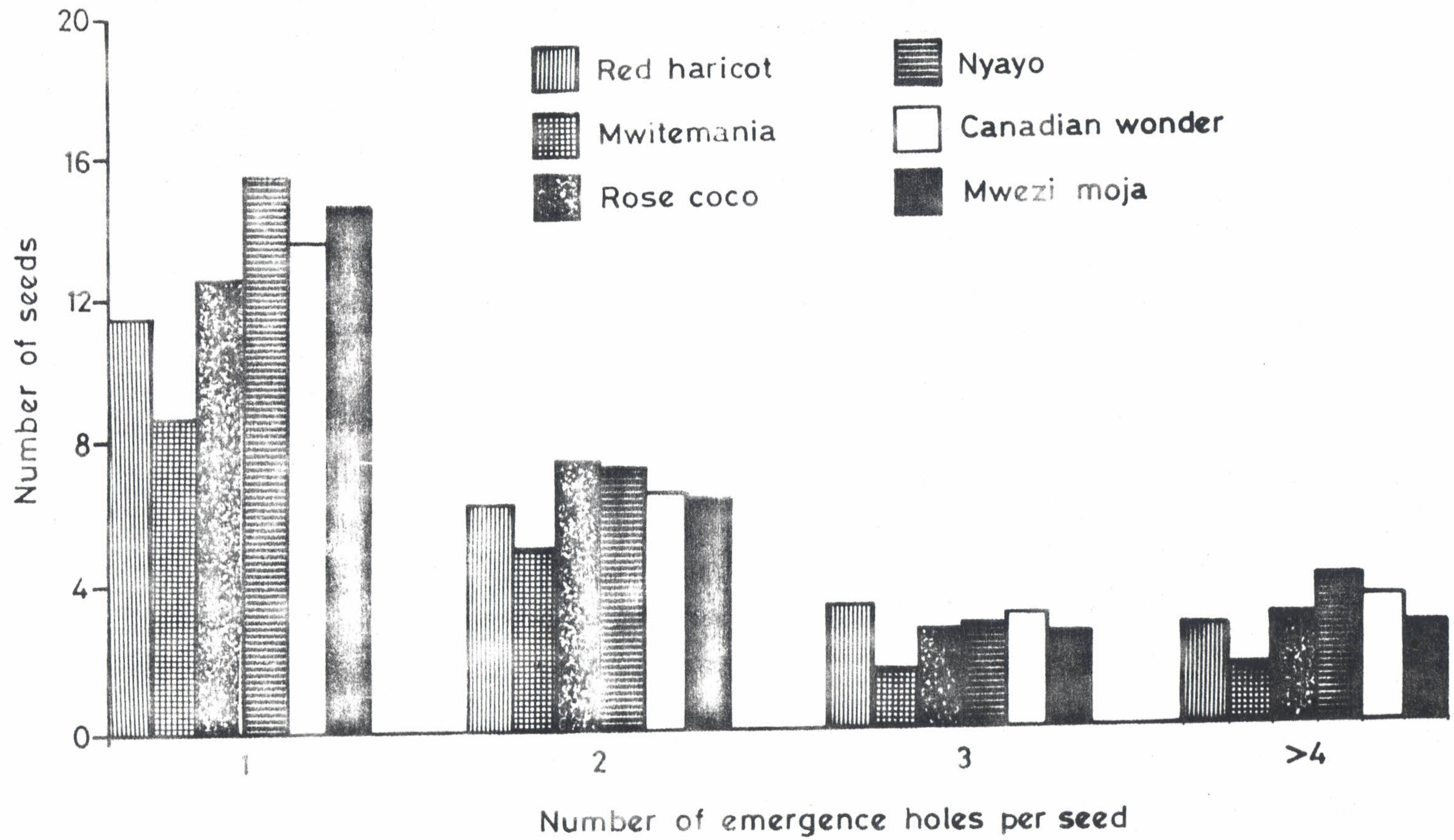


Fig. 4. Mean number of seeds of bean varieties with 1, 2, 3, and > 4 emergence holes of A. obtectus per seed after a mixture of varieties was artificially infested with the bettles.

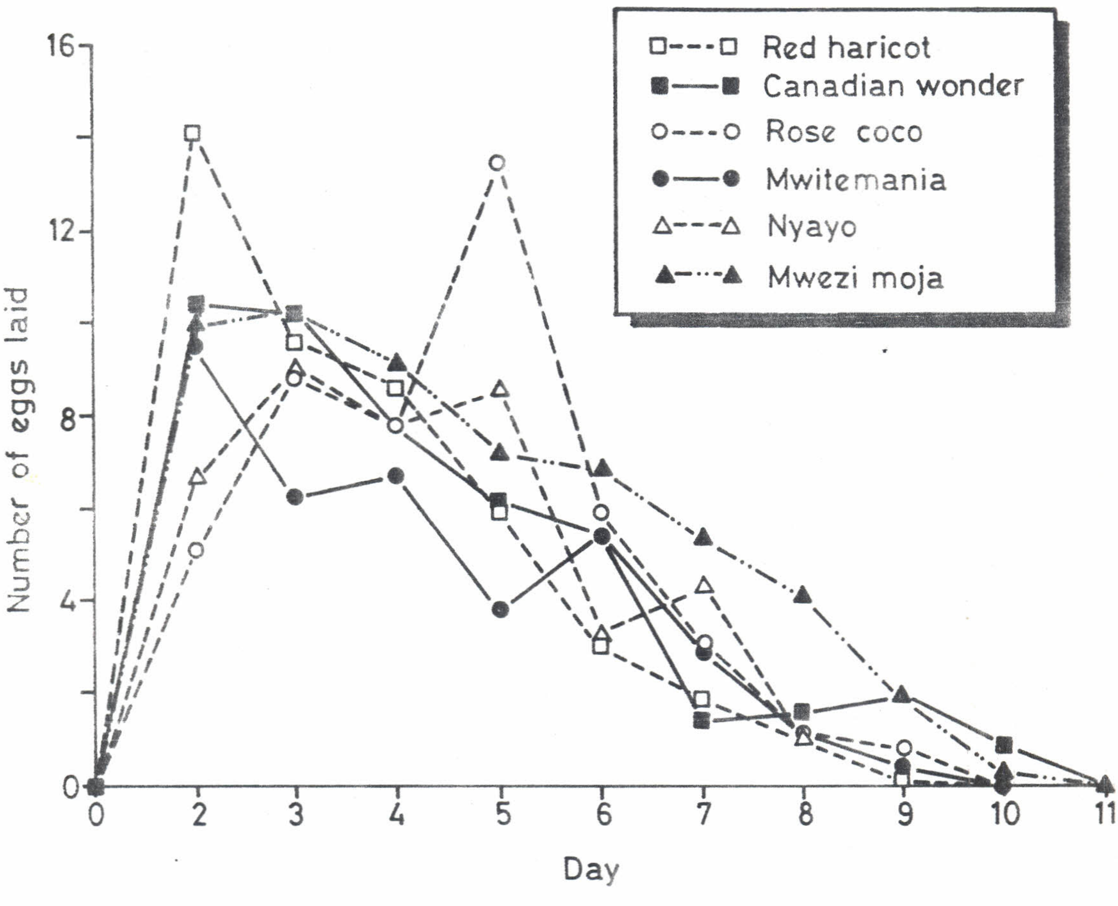


Fig. 5. Mean number of eggs laid by A. obtectus per day on bean varieties.

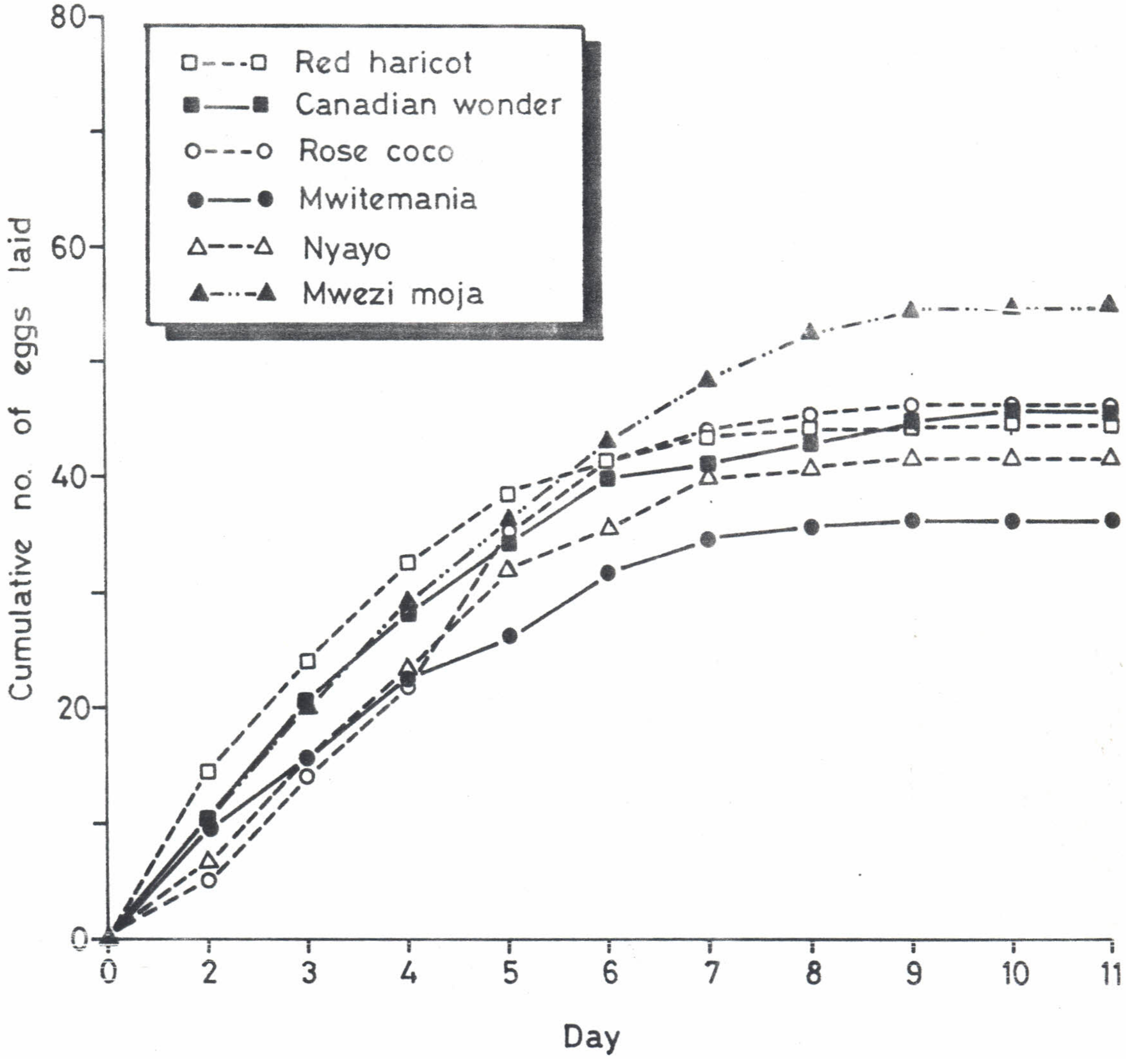


Fig. 6. Mean cumulative number of eggs laid by A. obtectus on bean varieties.

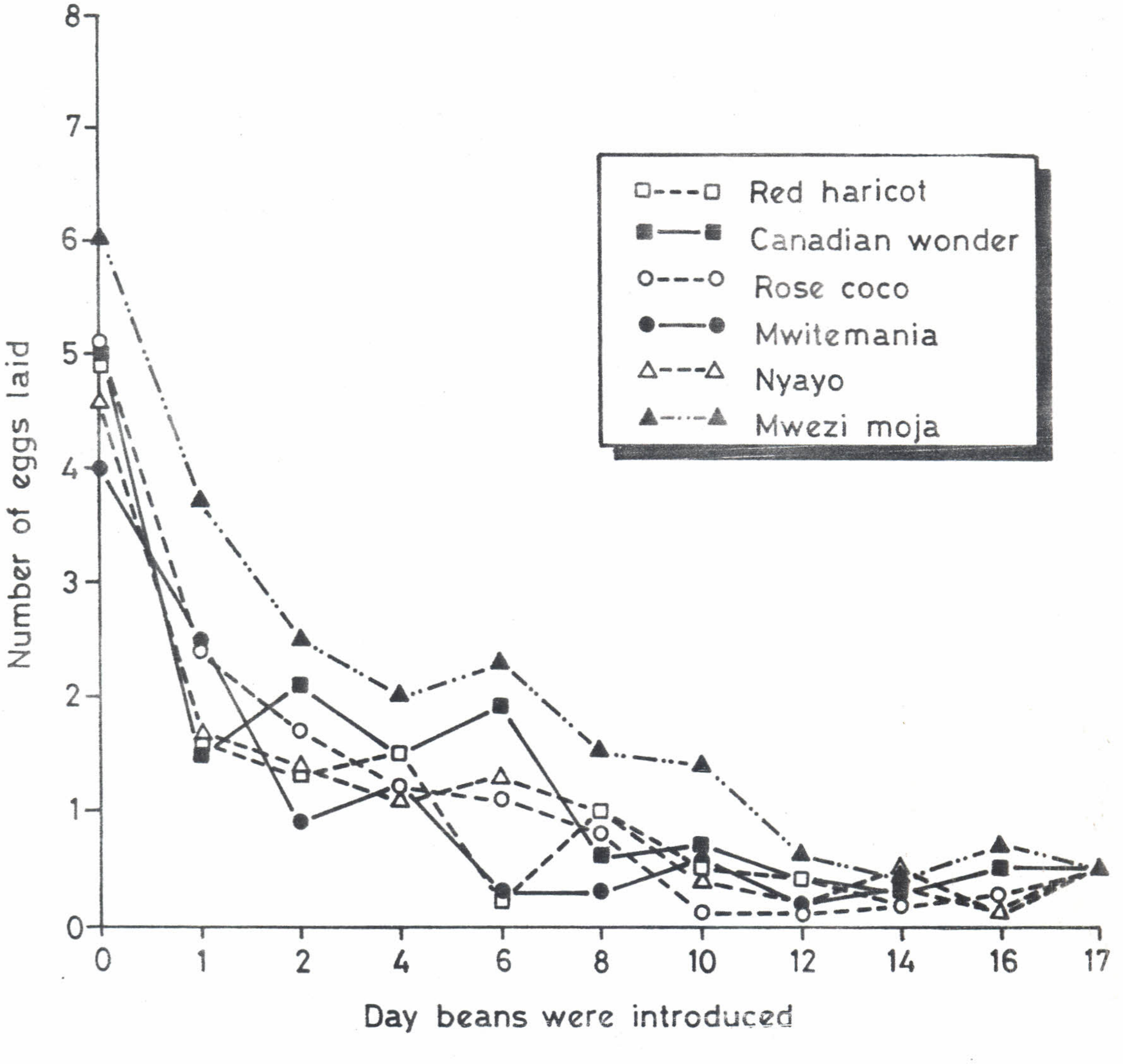


Fig. 7. Mean number of eggs laid by *A. obtectus* when provision of bean varieties was progressively delayed for 0 - 16 days.

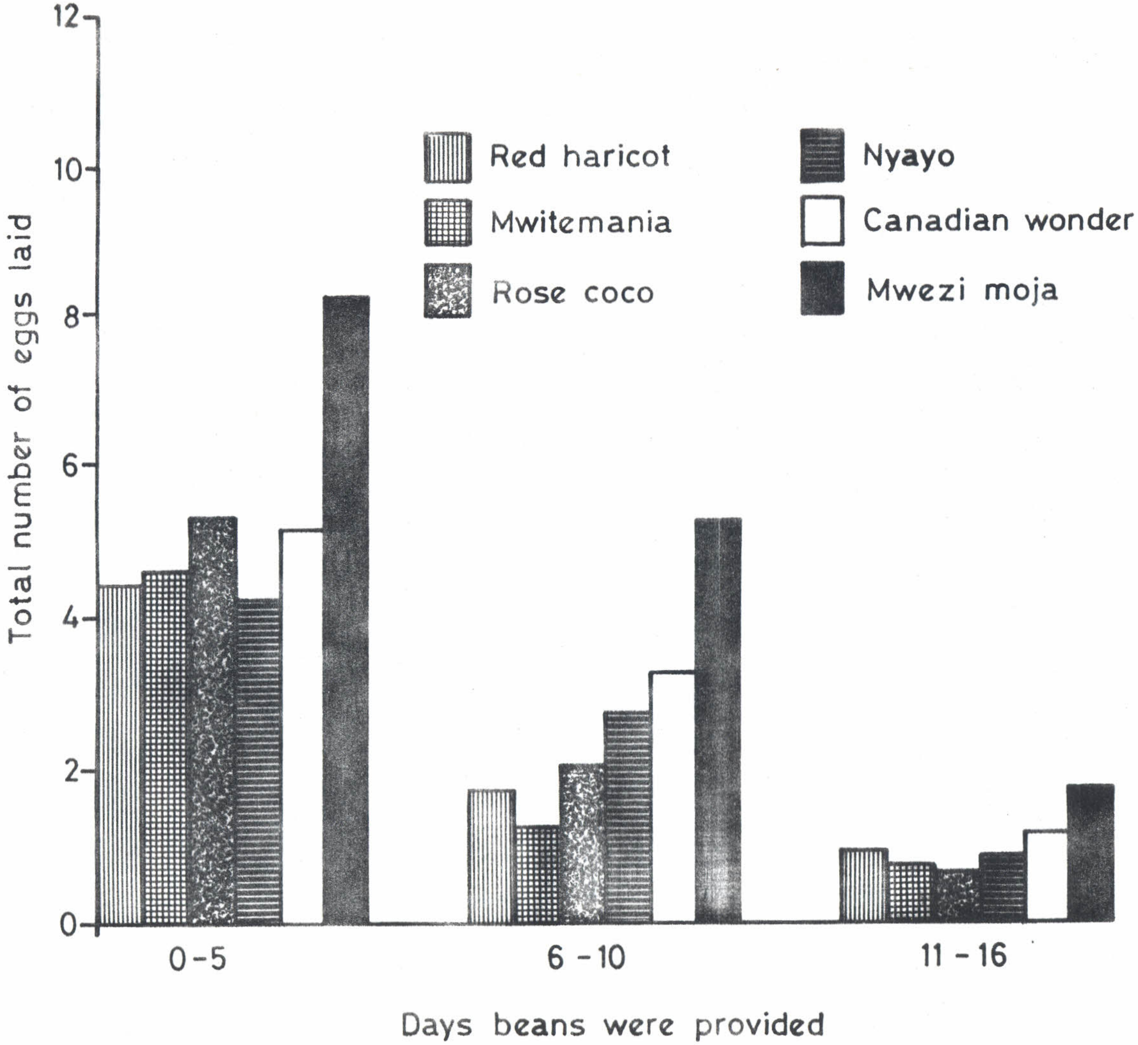


Fig. 8. Mean number of eggs laid by A. obtectus when provision of bean

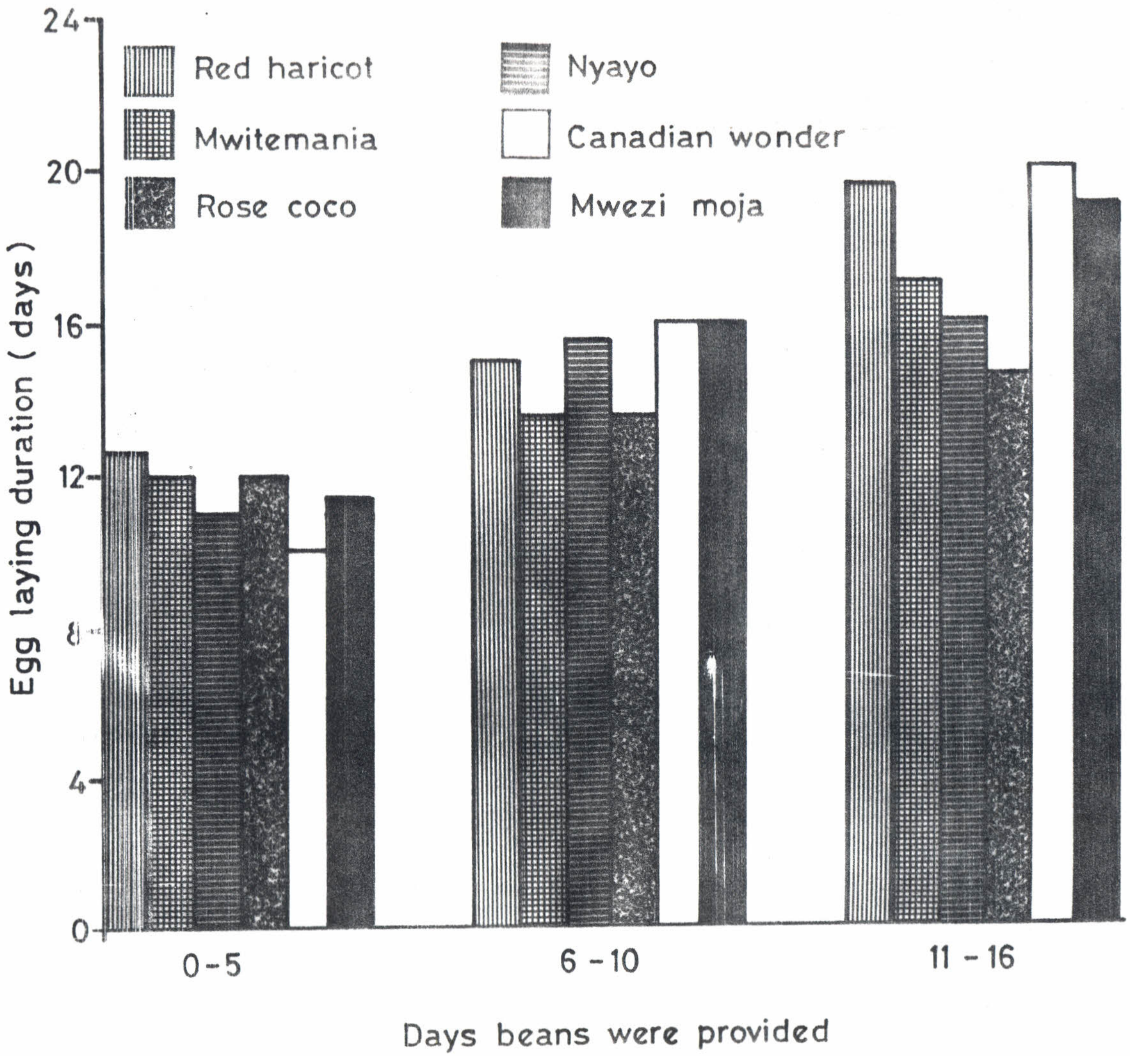


Fig. 9. Egg laying duration (days) of A. obtectus when provision of bean varieties was delayed for between 0 - 5, 6 - 10 and 11 - 16 days

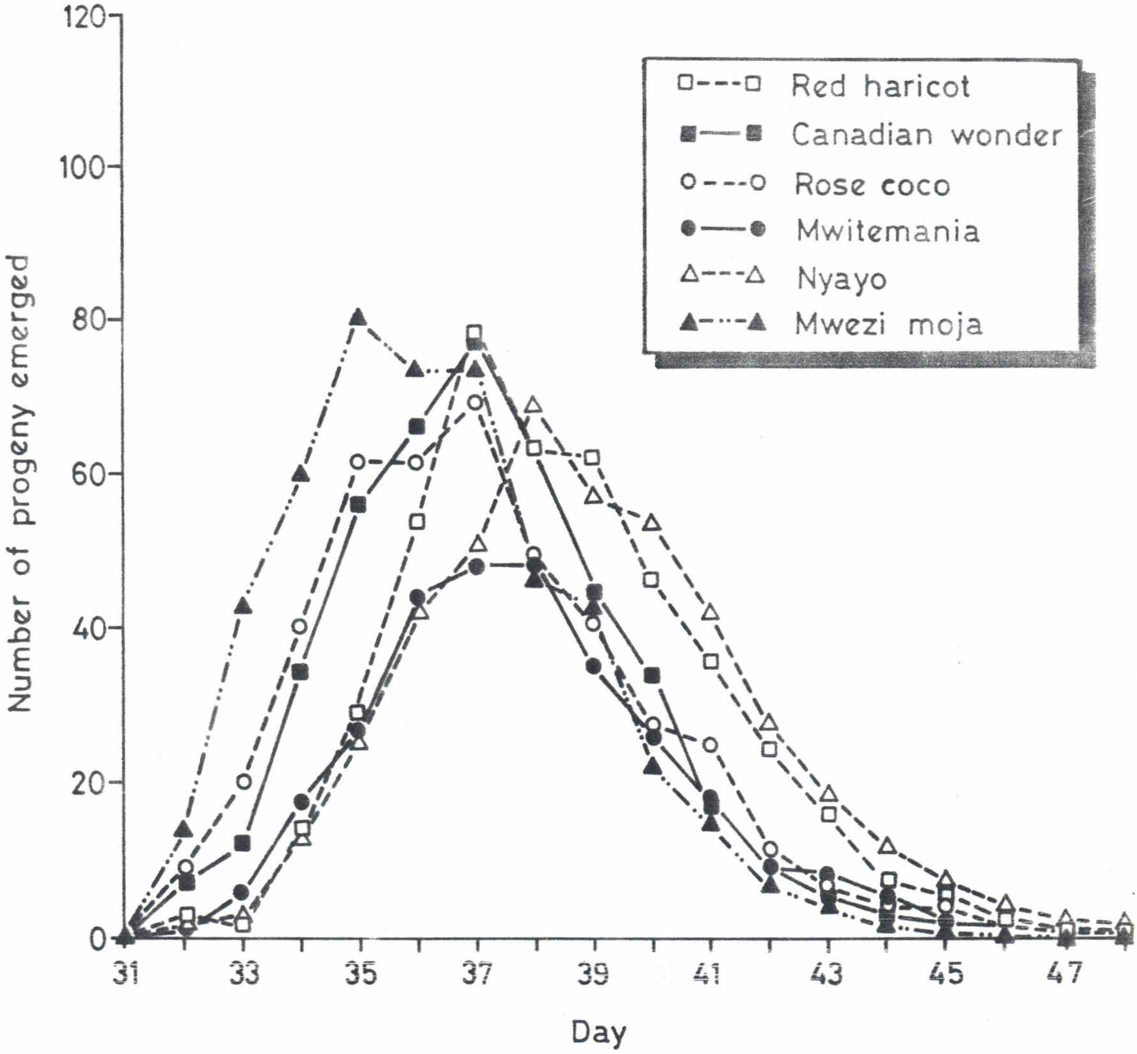


Fig. 10 Mean number of progeny of A. obtectus emerged per day from bean varieties after emergence of the first adults.

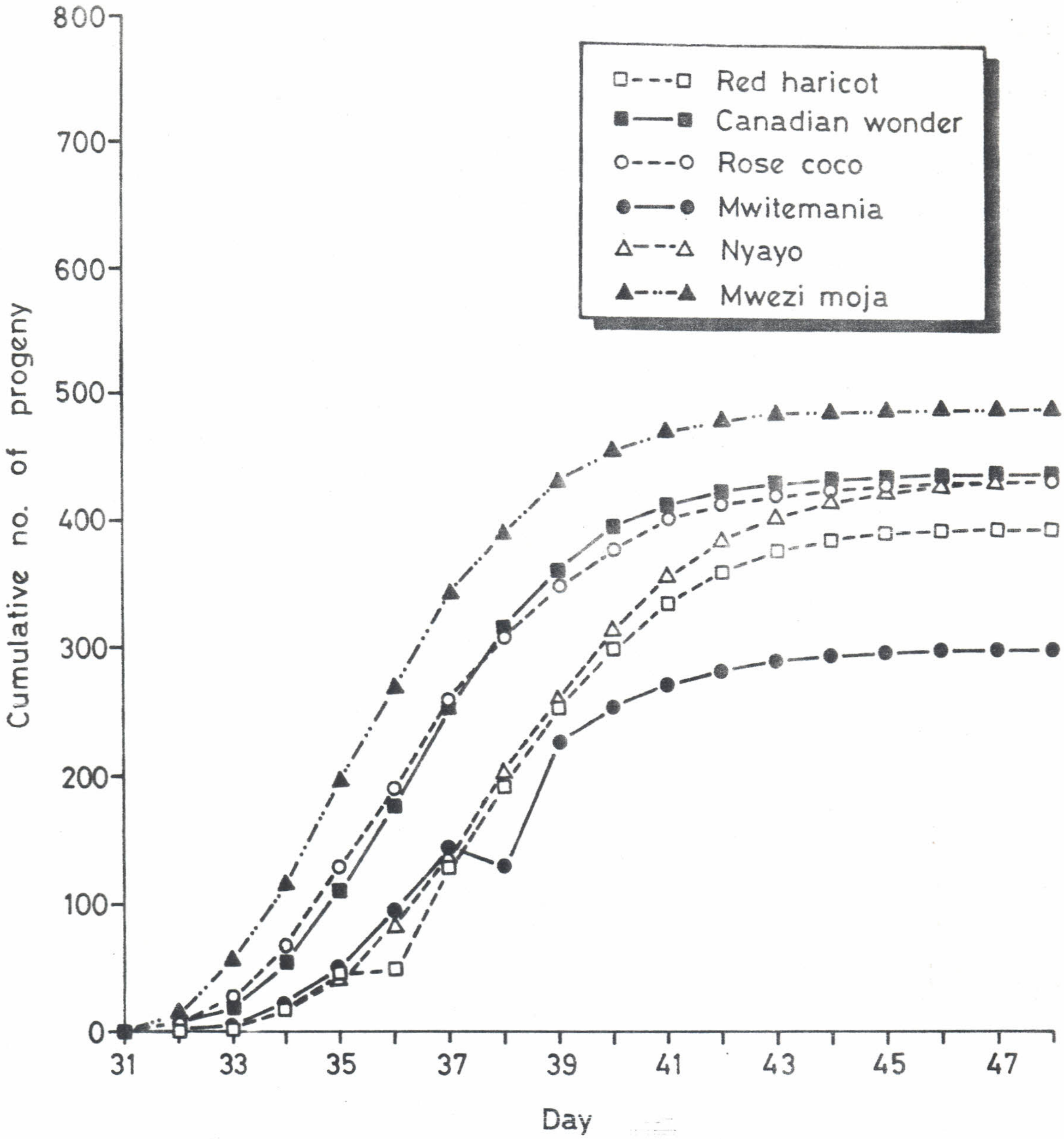


Fig. 11. Mean cumulative number of progeny of A. obtectus emerging from bean varieties after emergence of first adults.

CHAPTER FOUR

4.10 DISCUSSION

Generally, many bruchid beetles are known to be selective with regard to the variety and size of bean on which they deposit eggs (Jakhmola and Singh, 1971). In *C. chinensis*, females oviposit eggs on the largest seeds of beans first and in so doing avoid overloading small seeds with eggs (Mitchel, 1975). From the results of these investigations, it was found that the beetles were attracted more to varieties with large seeds compared to those with small seeds. This was supported by the fact that the number of beetles attracted to the beans was higher where the variety had large seeds. It is therefore possible that *A. obtectus* exhibited the same behaviour and tended to select beans with large seeds. Naturally, large seeds have more surface area for oviposition than small ones and this could in turn even provide enough food and space for pupation.

Furthermore, Dahms (1972) reported that the number of adult insects or larvae attracted to a cultivar when given a free choice, could be used as a measure of resistance of the cultivar to the insect pest. Thus, it is possible that the relatively few beetles attracted to some of the varieties could be an expression of resistance by the varieties to the beetles.

During this study, the number of emergence holes were used as a method of assessing the preference of the bean varieties by the bean bruchids. However, it should be noted that the difference in number of emergence holes between varieties could be due to other factors such as nutrient content and presence of toxic or inhibitory biochemical constituents that affect larval development as reported by Dobie *et al.* (1990), Hussey *et al.* (1969), Lukando (1978) and Gatehouse *et al.* (1987). Thus caution has to be taken when using the emergence holes as a measure of preference. In this case, the number of emergence holes were only used to supplement the observed number of beetles attracted to the bean varieties. The results indicated that some varieties had more emergence holes than others.

Generally, it is known that survival of larvae of bruchids is differentially affected by size and quality of beans and that selective pressures would favour females that discriminate among beans and avoid overloading a given bean seed (Mitchell, 1975). In the present study, it is likely that the beetles avoided overloading seeds and therefore relatively large seeds could have been preferred. Furthermore, it has been reported that testae of some bean varieties may be so thick that the newly hatched larvae die before reaching the cotyledon (Howe and Currie, 1964). Thus, mwitemania which had the thickest testa and the least number of emergence holes per seed could suggest that the thick testa adversely affected the larvae. Moreover, the fact that number of emergence holes per seed were significantly different between varieties could suggest that some varieties supported more larvae than others.

Investigations on the effects of bean varieties on fecundity of the beetles revealed that the beetles laid more eggs on some varieties than others. It has been reported that a female was capable of laying 40 - 60 eggs in her life time at 28°C and 70% relative humidity (Hill, 1975) . Similar observations have also been reported by Centro International de Agricultura Tropical (1986) that a female could lay an average of 63 eggs. Therefore the differences in number of eggs laid reported in this study and those of previous researchers could be attributed to differences in bean varieties and experimental conditions used because the bean varieties and experimental conditions used here were different from those used by other researchers.

Furthermore, it has been reported that characteristics of a host species of an insect pest may prevent oviposition by failing to provide the appropriate releasing stimuli that finally results in deposition of eggs (Beck, 1965). It has also been reported that oviposition in *A. obtectus* may be affected within a single variety of beans by small differences of surface smoothness, plumness of seed coat and by size and hardness of seed. Therefore, the differences in number of eggs laid between varieties could have been due to failure by some of the varieties to provide the complete releasing stimuli that could have resulted in the deposition of more eggs. It seems also that thickness of testa affected oviposition in the beetles in that mwitemania which had the thickest testa had the least number of eggs laid. It was also noted in this study that majority of the beetles laid most of their eggs within one week following adult emergence and that the egg laying duration was not significantly different between varieties. These results are in agreement with those of Davies (1959) and Howe and Currie (1964) who reported that eggs were laid by the bruchids within a week of adult emergence. Beans have been reported to regulate reproductive activity in *A. obtectus* and that in the absence of beans, females do not lay eggs but retain 30 to 40 Oocytes in the lateral oviducts (Huignard, 1974). Therefore, the fact that egg laying duration was not significantly different and the number of eggs laid was significantly different between varieties could suggest that some of the varieties induced least oviposition.

It has also been further reported by Teixeira (1956) that *A. obtectus* can retain most eggs until a suitable oviposition host is found. Therefore, in the present study, it is likely that the differences in number of eggs laid on different bean varieties was due to the delay by the females with the hope of finding a better host.

Furthermore, it has also been reported that signals coming from beans are picked by sensory organs of the beetles and they probably regulate the reproductive function through the nervous system and haemolymph (Jermy, 1974). In the present study, it was found that delaying provision of beans to the beetles resulted in reduced number of eggs laid and prolonged egg laying duration. It was also noted that in absence of beans, the beetles laid very few eggs. Therefore, this could suggest that the different bean varieties produced different effects on the reproductive activity of the beetles and that beans are essential for oviposition. For example, mwezi moja induced highest oviposition whereas mwitemania induced least oviposition.

First progeny of the beetles emerged from beans on about the 31St. to the 39th. day after introduction of parents into beans. Peak emergence of progeny was between the 35th. and 37th. day. Strong *et al.* (1968) rearing beetles on red kidney beans at 27 ± 1^{0}C and 55 ± 5% relative humidity found that in this species emergence of progeny began on the 29th. day and reached its peak on the 33rd. day after introduction of adults into beans. It is also known that growth and development in this species depends on host plant, variety of beans, temperatures, relative humidities among other factors (Strong *et al.*, 1968; Howe and Currie, 1964; Hill, 1975; Olubayo, 1980). The findings of the current study are in agreement with these reports in that beetles took different periods to emerge from the different varieties and the conditions used were also different from those of previous researchers.

The total number of progeny that eventually emerged were significantly different between varieties. Several factors could have been responsible for these varietal differences in number of progeny emerged. It has been reported that in unsuitable food media, the pupal cell of the beetles may be formed at the centre of the seed such that progeny are unable to emerge after pupation (Howe and Currie, 1964). It is likely that some varieties used in this study were unsuitable food media so that some of the progeny failed to emerge. Thus, the differences in number of progeny emerging from different bean varieties.

The duration in days taken by 50% of the progeny to emerge was significantly different between varieties. It is known that the duration of the life cycle is a good indication of the suitability of an environment for growth and development of an insect. In general, adverse conditions prolong developmental period and few insects survive (Sokoloff *et al.* 1966; Howe, 1971). Thus, it can be suggested that the duration of the life cycle of A. obtectus was a measure of the suitability of the bean varieties for growth and development and that mwezi moja was the most suitable environment for the development of the beetles compared with other varieties.

Furthermore, presence of toxic or inhibitory biochemical constituents in beans have been reported to retard growth and development and to kill larvae of various bruchid beetles. Applebaum *et al.* (1970) found that a heteropolysaccharide isolated from beans decreased the rate of development and the number of eggs laid by *C. chinensis*. Moreover, it has also been reported that phytohemagglutinins extracted from black beans can kill larvae of *C. maculatus* (Janzen and Juster, 1976). Dobie *et al.* (1990) has also reported that some varieties of bean, are resistant to *A. obtectus* and the resistance was associated with the presence of a lectin-like protein (LLP) and a novel protein. Although, the presence of such biochemical constituents in these bean varieties were not investigated, it is possible that they were certain chemical components which may have retarded growth and development. Therefore, the differences in developmental period of the beetles in the various bean varieties could be attributed to the existence of chemical components.

The assessment of the susceptibilities of the bean varieties to attack by *A. obtectus* using a susceptibility index revealed that the bean varieties showed significant differences in their susceptibilities to attack by the beetles. Indeed, Lukando (1978) and Rheenem *et al.* (1983) found that some bean varieties under cultivation in Kenya were different in their susceptibilities to attack by *A. obtectus*. They attributed the differences to thickness of testa, presence of crystals in the testa and alkaloids. Elsewhere, Gatehouse *et al.* (1987) found that resistance was associated with a heteropolysaccharide. Hussey *et al.* (1969) also reported that a pest's reproduction may be reduced by specific nutrient deficiencies or the presence of toxic substances. Dobie *et al.* (1990) also suggested that resistance could be caused by the inability of the larvae to penetrate the testa or by biochemical features.

Although, the biochemical characteristics of the bean varieties were not analysed during the current study, it is possible that some biochemical constituents of the bean seeds retarded growth and development and probably killed some of the larvae. Therefore, the differences in developmental period, number of progeny emerged and consequently the differences in susceptibilities could be reflections of such varietal biochemical characteristics.

Physical characteristics of beans have also been reported to be responsible for their resistance to insect pests of stored grain legumes. These phenomena could also provide possible explanation to the differences in susceptibilities of the bean varieties to attack by *A. obtectus*. Moreover, it has been reported that testa characteristics and hardness of seed could affect susceptibility of bean varieties to attack by the bean bruchids (Howe and Currie, 1964). In the present study, it was found that some physical characteristics of seeds of some of the varieties affected susceptibility. For instance, seed size affected susceptibility in canadian wonder. It was also found that the combined effect of seed size, seed hardness and thickness of testa affected susceptibility in canadian wonder. It seems here that the effect of one physical factor on susceptibility can magnify the effects of other factors.

4.20 CONCLUSIONS

A number of conclusions can be drawn from the results of these investigations. First and foremost, it was apparent that *A. obtectus* showed a marked preference for certain bean varieties for oviposition purposes. Mwezi moja attracted the largest number of beetles whereas mwitemania attracted the least.

Secondly, fecundity was affected by bean varieties. Beetles laid the largest number of eggs on mwezi moja and the least on mwitemania. Egg laying duration was the same in all bean varieties. Thirdly, beans induced oviposition in the majority of the female beetles. Some varieties induced more egg laying than others. Thus, the largest number of eggs was oviposited on mwezi moja, while mwitemania had the least effect on oviposition. Delayed bean provision reduced fecundity and prolonged the egg laying duration.

Fourthly, number of progeny of *A. obtectus* emerging were different between varieties. The highest number of progeny emerged from mwezi moja and the least from mwitemania. Duration taken by 50% of the progeny to emerge was different between varieties. Beetles took the longest period to emerge from red haricot and the shortest from mwezi moja.

Susceptibility index was also different between varieties. Mwezi moja was the most susceptible variety to attack by *A. obtectus* whereas mwitemania was the least susceptible. Last but not least, thickness of testa, size and hardness of seed played a subordinate role in susceptibility in most varieties.

From the findings of this study, it is apparent that *A. obtectus* has shown preference for certain bean varieties as oviposition sites and that bean varieties affected fecundity. It is also clear that some varieties are more susceptible to attack by the beetles than others. More research could be carried out with a view to understanding the major causes of the above differences between the bean varieties. Field surveys could also be carried out on field pests, agronomic factors and consumer preference for bean varieties. With such information, a variety or varieties could be bred, selected and recommended to farmers for cultivation in Kenya.

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Appendix 1. Anova table for the mean number of *A. obtectus* settled on bean varieties when provided with a random choice of bean varieties.

Source of variation	d.f	sum of squares	mean square	F ratio
Between groups	5	251.8	50.4	8.13
Within groups	36	222.9	6.19	
Total	41	474.7		

Appendix 2. Anova table for the susceptibility indices of the bean varieties .

Source of variation	d.f	sum of squares	mean square	F ratio
Between groups	5	33.7	6.73	3.33
Within groups	54	109.0	2.02	
Total	59	142.7		

Appendix 3. Anova table for the thicknesses of testae of the on bean varieties. varieties.

Source of variation	d.f	sum of squares	mean square	F ratio
Between groups	5	0.0084	0.0017	15.2
Within groups	78	0.0087	0.0001	
Total	83	0.0171		