EISSN: 2707-0425

East African Journal of Science, Technology and Innovation, Vol. 4 (1): December 2022

Influence of Phaseolus vulgaris L. (Fabaceae: Fabales) varieties on oviposition behavior and susceptibility to infestation by Acanthoscelides obtectus Say (Coleoptera: Bruchidae)

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Abstract

Despite beans being a vital source of cheap protein, its yield is reduced mainly by diseases, field and storage pests. An effective and sustainable control option against storage insect pests could be improving the genetic resistance of the host plant in relation to the behaviour of the pest. A laboratory investigation was conducted to determine effects of bean varieties on oviposition behaviour and susceptibility to infestation by *Acanthoscelides obtectus*. The experiments evaluated oviposition site preference, number of eggs laid, and number of progeny and emergence holes of *A. obtectus* per bean variety. The number of progeny and duration in days taken by 50 % of progeny to emerge was determined and used to calculate susceptibility index. Seed harness index, thickness of testae and surface area of bean varieties were determined and related with susceptibility indices. Results indicated that the number of eggs laid, progeny, and emergence holes, duration taken by progeny to emerge, susceptibility indices, seed surface area and hardness were statistically different. Mwezi moja variety was the most preferred host for oviposition whereas Mwitemania was the least preferred. Beetles laid the highest (84.5) number of eggs in Mwezi moja and the least (41.9) in Mwitemania. Number of progeny of *A. obtectus* emerging were the highest in Mwezi moja (73.7) and the least (36) in Mwitemania. Beetles took the shortest (36.75 days) period to emerge from Mwezi moja and the longest (38.63days) period from Mwitemania. Mwezi moja was the most susceptible variety to attack by A. obtectus with a susceptibility index of 7.43 and Mwitemania was the least susceptible with an index of 6.51. Nyayo had the highest (0.019) seed hardness index and Mwezi moja and Canadian wonder had the least (0.012) hardness index. Mwezi moja had the largest (278mm) surface area whereas Mwitemania had the lowest (171mm) surface area. Physical characteristics of bean varieties and oviposition behavior of beetles may be incorporated in bean breeding programs to produce a variety/varieties resistant to *A. obtectus*.

Keywords: *Bean varieties; Acanthoscelides obtectus; oviposition; susceptibility index; progeny*

Received:	31/01/22
Accepted:	05/12/22
Published:	16/12/22

Cite as: *Bett and Kiplagat, (2022)* Influence of Phaseolus vulgaris L. (Fabaceae: Fabales) varieties on oviposition behavior and susceptibility to infestation by Acanthoscelides obtectus Say (Coleoptera: Bruchidae). *East African Journal of Science, Technology and Innovation 4(1).*

Introduction

Bean varieties (*Phaseolus vulgaris* L.) (Fabales: Fabaceae) is the most cultivated grain legume in several developing nations, where its contribution as human and animal food and income is considered vital (Kusolwa 2007, Ayvas et al. 2010, Ogendo et al. 2012). However, bean yield is limited by such factors as diseases, field and storage pests (Ayvas *et al*., 2010, Ogendo *et al*., 2012). The contribution of stored product insect pests in stored grain loss is enormous since the insects damage grains in the field and storage. The predominant bruchid pest species in many countries cultivating beans are *Acanthoscelides obtectus* Say (Coleoptera: Bruchidae) and *Zabrotes subfasciatus*. The beetles are attributed to most damage in stored beans (Kusolwa 2007). *Acanthoscelides obtectus* causes most damage in many African countries cultivating beans. It is also considered to have a high genetic variability and a more geographic dispersal as compared to Z. *subfasciatus* (Rendón-Huertaet *et al.,* 2013). However, *A. obtectus* causes more damage with losses in the range of 20-100% of stored beans depending on the storage conditions (Baldin *et al.,* 2008, Ebinu *et al.,* 2016, Ogendo *et al.,* 2012).

Various alternatives for controlling storage insect pests such as chemical insecticides, cultural and physical control methods have proven effective. Chemical pesticides have been used against storage insect pests but their application has not been fully exploited in farming due to environmental, health and economic concerns (Isman, 2020). Besides, the intensive use of synthetic insecticides has resulted in pest resurgence and pests developing resistance (Ogendo *et al.,* 2012, Bett *et al.,* 2016). In this scenario, host plant resistance seems to be an alternative method of controlling seed beetles including *A. obtectus*, since it is environmentally friendly. In addition host plant resistance technologies are easier to transfer to small-scale farmers than more complex knowledge-based agronomic and crop protection practices (Ebinu *et al.,* 2016, Ogendo *et al.,* 2012). Furthermore, host resistance may be used as a constituent of integrated pest management because it is compatible with cultural, chemical, physical and biological control measures (Keneni *et al.,* 2011). However, the majority of crop breeding programs undertaken in the past were based mainly on improving seed yield, resistance to disease and field pests. Few of the studies focused on improvement of crop for resistance against storage insect pests (Keneni *et al.,* 2011).

Host plant resistance against insect pests is associated with physical and chemical characteristics of host plant and is associated with non-preference and tolerance by the pest (Edwards and Singh 2006). According to Lattanzio *et al.,* (2005) and Edwards and Singh (2006) the resistant qualities of a host plant can be physical, physiological and chemical that interrupt normal growth and proper development of affected pest species. The shape of seed and color are some of the physical characteristics, whereas the physiological and chemical qualities include secondary metabolites and anti-feeding substances influencing physiological processes in bruchid species (Lattanzio *et al.,* 2005, Somta *et al.,* 2007, War *et al.,* 2017). The chemical process employed by herbivores to defend themselves against predators come in different forms, highly dynamic, and are intervened both by indirect and direct defense mechanisms. The protective chemicals are released in response to plant damage and influence nutrition, reproduction, and longevity of herbivores. The relationship between leguminous plants bruchid beetles is based on concurrent evolution of both with each developing defensive mechanisms against each other. In this scenario, legumes have developed poisonous biochemical to repel or destroy bruchid beetles. The bruchid beetles on the other hand, have developed adaptive approaches to counter the properties of these deadly biochemical substances in legumes (Somta *et al.,* 2007, War *et al.,* 2017).

Despite the application of morphological, biochemical, and molecular traits in breeding and adoption of high yielding bean varieties with resistance to diseases and pests in the field, more research is yet to be done to produce varieties resistant to stored product insect pests. There are various challenges related to lack of efficient selection strategy, proper gauging of selection pressure for new biotypes and breakdown of resistance and lack of knowledge of genetic bases of resistance (Keneni *et al.,* 2011). Moreover, the emergence of genetic variability in pest species being targeted in breeding programs is an additional constrain encountered by plant breeders. The occurrence of biotypes of a pest species has been attributed to failure of resistance in legume crops bred against bruchid beetles (Fox *et al.,* 2010). In this case, there is need to constantly screen varieties for resistance to counter damaging effects of evolving pest biotypes in order to improve the resistant traits.

In this context, the current paper reports the assessment of oviposition behavior of *A. obtectus* and susceptibility to infestation of bean varieties popularly cultivated by farmers in East Africa.
In particular the experiments focused on testing the hypothesis that; (1) oviposition site preference by adult *A. obtectus* differed with bean varieties, (2) number eggs laid and progeny, emergence holes of *A. obtectus* and susceptibility to infestation varied according to bean variety (3) physical characteristics (seed hardness, thickness of testae and surface area) of bean varieties have a relationship with susceptibility indices.

Materials and methods

Experimental conditions and design

The experimental room was maintained at temperatures of 28±2°C, relative humidity of 65±5% and photoperiod of 12h darkness: 12 h light which are optimum conditions for rearing stored product insect pests. The bean varieties (Mwitemania (GLP 92), Rose coco, Nyayo (GLP2), Canadian wonder (GLP 24), and Mwezi moja (GLP 1004)(Figure. 1) used for the study were sourced from Kenya Agricultural and Livestock Research Organization (KALRO) (Horticultural Research Centre, Thika, Kenya). The beans were selected based on size and consumer preferences (Korir *et al.,* 2005). Clean dry bean grains of each variety, used for the experiments, were placed in the oven at 100°C for 24 h in order to eradicate previous pest infestation. The percent moisture content of bean varieties was evaluated with the aid of a moisture meter and moisture content was 12 ± 1.5 %. Beans were preconditioned to experimental conditions for seven days before being used in bioassays.

Rearing of test insects

Samples of *A. obtectus* were obtained from a stock maintained by Egerton University Integrated Biotechnology Laboratory (Kenya). The stock had been maintained for six years but new genetic stock being added regularly after every two years. Beetles for the experiments were reared on Red haricot variety to ensure they did not get conditioned to the varieties being used in the experiments. One kilogram (1kg) of beans and 100 unsexed adult beetles were placed in 1L Kilner Jars and kept in experimental room. The adult beetles laid eggs for 10 days and thereafter the beans were sieved to remove adult beetles allowing the larval stages to continue developing. In order to obtain adult beetles of similar ages, newly emerged beetles were isolated on different jars on a daily basis for 5 days.

Adult A. obtectus oviposition site preference

Adult *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 larvae and that would ensure that development is successful to adult stage. In these experiment, effect of different bean varieties on ovipostion site preference by *A. obtectus* was investigated. The experimental design was randomized complete block design (RCBD) with five replicates. This was also a choice assay where in the first part of the experiment 30g of each bean variety were placed in pill boxes (Diameter: 4cm and Height:2cm). Each variety had five replicates. A petri-dish containing 300 unsexed one day old beetles and damaged beans were placed at the centre of a cage made of muslin cloth. The pill boxes containing bean varieties were randomly placed around the petri to form a block. Five other blocks were also prepared. The petri-dish was to act a source of infestation for the bean varieties. Beetles could enter or leave petri-dishes at will. After 24h the number of beetles settled in each pill box were counted.

1. Mwitemania

2. Rose coco

3. Nyayo

4. Canadian wonder

5. Mwezi moja

Figure 1. Bean varieties used in the study (Mwitemania, Rose coco, Nyayo, Canadian wonder, Mwezi moja)

The distribution of exit holes on each seed has been used as indication of suitability of varieties of legumes as medium of development of bruchids. It is also known that in bruchid beetles that feed on seeds the number of exit holes on each seed follow a Poisson probability distribution model (Parsons and Gredland 2003, Paukku and Kotiaho 2008). In the second part of the experiment the number of exit holes on each variety were also determined. Beetles were provided with a mixture of all five bean varieties. The containers were left in the experimental room until adults of next generation emerged. The number of emergence holes per seed per variety were counted.

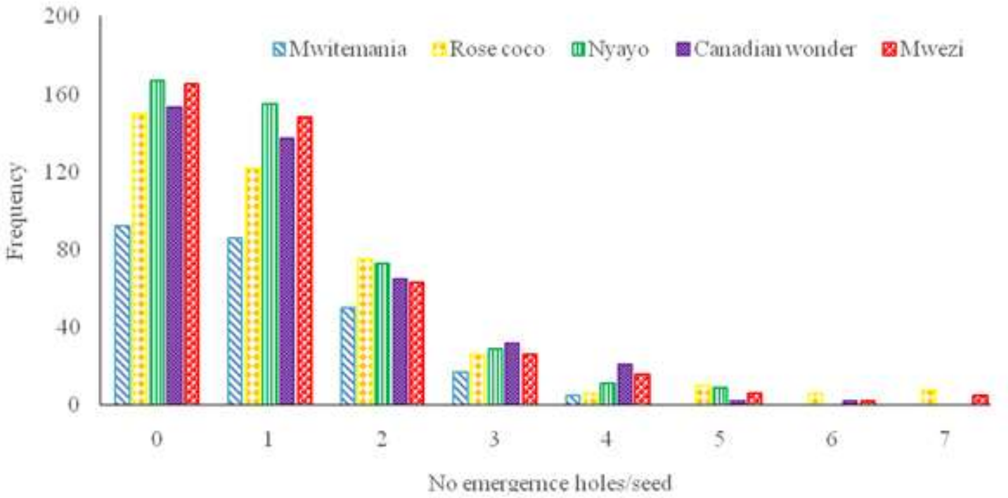

Figure 2. Frequency of number of emergence holes per seed per bean variety

Effects of bean varieties on egg laying in A. obtectus

Beans are the most preferred oviposition site for *A. obtectus*. This experiment was set up to investigate whether fecundity by *A. obtectus* females is influenced by different bean varieties. The study design used for this studies was a randomized block design (RBD) with treatments having four replicates. This was a no-choice experiment where for each variety, 20g of seed were placed in a pill box. One female and one male one (day old) beetles were introduced into pill boxes. As a negative control beetles were placed in pill boxes containing beads. The fecundity of beetles was determined by counting eggs laid by the beetles, 24 hours post introduction of adults. Counting continued for 7-8 days depending on bean variety until all beetles had died. Similarly, the egg laying duration of each female was also noted.

Assessment of susceptibility of bean varieties to attack by A. obtectus

This experiment was set up to test whether there is variation in susceptibility of different varieties of beans when infested with *A. obtetus*. The study design used for this studies was a randomized block design (RBD) experimental design with treatments having four replicates. This was no-choice assay where for each variety 250 bean seeds were mixed with 20 females and 20 males (one day old beetles) in 500 ml plastic containers. Containers were kept in the experimental room. After 30days emergence of progeny was checked. When the first adults emerged, they were counted and discarded. The process of counting continued for 35-39 days depending on variety until emergence of progeny stopped. The number of progeny and duration in days taken by 50 % of progeny to emerge was used to calculate susceptibility index. These indices were determined as in equation1 (Dobie, 1974; Gudrups *et al.,* 2001):

$$SI = \frac{LnF*100}{DME} \quad \ldots\ldots 1$$

Whereby SI =Susceptibility Index, Ln =Natural logarithm, F = Total progeny, and DME = Duration(days) taken by 50 % of progeny to emerge

Physical factors that are likely to affect vulnerability of bean cultivars to infestation by A. obtectus

The experiment was 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 index and harness of seed, thickness of testae and size (surface area) of seed were also considered.

The method of Gudrups *et al.,* (2001) was used to determine seed hardness. Hundred grams of each variety replicated four times were ground using an electric grinding mill at constant speed for 3 minutes. The powdered seed was sieved with a sieve (aperture size of 500µm). Fractions that were retained and those that passed through the sieve aperture were weighted separately. The seed hardness index (HI) was computed using equation2 (Gudrups *et al.,* 2001):

$$HI = \frac{Weight\ of\ retained\ part\ of\ flour}{Weight\ of\ sieved\ part\ of\ flour} \quad \ldots\ldots 2$$

Testae of bean varieties were removed using a scalpel with each variety having four replicates. The thickness of pieces of test pealed out were measured using a micrometer screw gauge.

Surface area was used as a measure of size. The surface area of beans was computed using the equation of determining the surface area of a cylinder (Equation 3) (Paukku and Kotiaho, 2008):

$$SA=2\square h + 2\square r^2 \quad \ldots\ldots 3$$

Where: SA= surface area, r = radius and h = height.

At the end of these experiments, the relationship between susceptibility index, thickness of testae, hardness and surface area of seed was computed using regression analysis.

Statistical data analysis

The data on susceptibility index was transformed using arcsine transformation before being analyzed using ANOVA. Data on number of adults distributed in bean varieties, eggs laid, egg laying duration(days), duration(days) taken by 50% of progeny to emerge, susceptibility index(transformed), seed hardness index, thickness of testae and surface area were analyzed using ANOVA (SAS, 2017). In the ANOVA(model 1) analysis the independent variables were varieties whereas the dependent variables were number of adults, eggs laid, egg laying duration, duration taken by 50% of progeny to emerge, susceptibility index (transformed), seed hardness index, thickness of testae and surface area. In ANOVA model 2 analysis the number of emerged progeny was the dependent variable while day and variety were the factors. The expected number of emergence holes per seed per variety were generated using the Poisson probability distribution model (SPSS 2013, Ebinu *et al.,* 2016). The obtained expected frequencies were then compared with observed frequencies using chi-square test (test for homogeneity) (SPSS 2013). The relationship between susceptibility index and seed hardness index and susceptibility index and surface areas of each bean variety was analyzed using regression analysis (Stata 2019). The differences between means were determined using Tukey's honestly significant difference (HSD) at 5% significance level.

Results

Preference of bean varieties as oviposition site by adult A. obtectus

The results indicate that bean varieties significantly influenced the distribution of adults bean bruchids ($F=5.12$; $df=4, 15$; $P<0.001$). The number of beetles settled in Mwezi moja variety were double that settled in Mwitemania variety (Table 1). The results also show that numbers were significantly different between Mwezi moja and all varieties except Canadian wonder (Tukeys HSD) (Table 1). The numbers of adults that settled in Mwitamania was also significantly different from Canadian Wonder Tukey's HSD) (Table 1).

Table 1. Mean number of beetles settled in varieties, eggs laid and egg-laying duration in A. obtectus per bean variety

Bean variety	***No of *A. obtectus***	***No. of eggs laid**	***Egg laying duration**
Mwitemania	36.00±7.7^{a}	41.9±0.7^{a}	7.1±0.3^{a}
Rose coco	43.75±6.9^{b}	50.39±0.7^{a}	7.4±0.4^{a}
Nyayo	44.75±11.75^{b}	49.3±0.7^{a}	7.9±0.4^{a}
Canadian wonder	57.50±10.36^{c}	56.2±0.7^{b}	7.6±0.4^{a}
Mwezi moja	73.75±22.4^{d}	85.4±0.7^{c}	8.0±0.3^{a}

Means in a column followed with the same letter are insignificant

*(Mean ± SD)

Number of eggs laid and egg-laying duration (Days)

The numbers of eggs laid by *A. obtectus* per day on different varieties are shown in Fig. 1. The beetles laid highest (85.4) number of eggs in Mwezi moja and lowest (41.9) on mwitemania (Fig. 2). Number of eggs laid per day was significantly different between varieties (F=548.04; df=4,135; *P<0.001*). The results also show that numbers of eggs laid per day in Mwezi moja were significantly different from all varieties except Canadian wonder (Table 1). Similarly, the numbers of eggs laid in Mwitamania variety was significantly different from Canadian Wonder (Tukeys HSD). Egg laying duration days was insignificant between the different varieties (F=0.62; df=4,15; *P=0.15*). Egg laying duration (days) was shortest (7days) in Mwitemania and longest (8 days) in Mwezi moja. The mean number of eggs laid by a female per day without beans (Control) was 0.6±0.2.

Duration (days) taken by 50% A obtectus progeny to emerge and susceptibility index

Duration (days) taken by 50% of progeny of *A. obtectus* to emerge was statistically significant in bean varieties (F=18.66; df=4,15; *P<0.001*). The shortest (35 days) duration taken by 50 % progeny to merge in Mwezi moja variety whereas the longest (38.8 days) duration was in Mwitemania variety (Table 2).

From the results on susceptibility index, Mwezi moja had the highest (7.43) and Mwitemania the lowest (6.51) susceptibility index. Susceptibility indices were significantly different between bean varieties (F=15.6; df=(4,15); *P=0.01*). Mwezi moja variety was different from Mwitemania (Tukeys HSD).

Table 2. Duration (days) taken by 50% of progeny to emerge, total progeny and susceptibility index of bean varieties

Bean variety	***DTPE$_{50}$**	**Total progeny**	***SI**
Mwitemania	38.63±0.85^{a}	364	6.51±0.29^{a}
Rose coco	35.13±0.48^{b}	446	7.43±0.34^{b}
Nyayo	38.8±0.75^{a}	376	6.53±0.16^{b}
Canadian wonder	38.63±1.1^{a}	427	6.70±0.27^{b}
Mwezi moja	36.75±1.6^{c}	458	7.53±0.16^{c}

Means in a column followed with the same letter are insignificant

DTPE$_{50}$= Duration (Days) taken by 50% of progeny to emerge, SI=Susceptibility Index

*(Mean ± SD)

Emergence holes per seed and Poisson Probability Distribution

Results of number of emergence holes per seed perforated by progeny of *A. obtectus* in bean varieties showed emergence holes per seed ranged from 0 to 7 per seed. The mean number of emergences holes per seed was lowest (1.4) in Mwitemania and highest (1.7) in Rose coco. The results also indicated that frequency of number of emergence holes per seed per variety followed a Poisson probability distribution model (Fig.2). The test for homogeneity (chi-square) of observed and expected number of emergence holes per seed showed that all varieties were statistically significant (x^2=24.69-228.53; df= 3; $P<0.001$) (Table 3).

Table 3. Mean no. of emergence holes of A. obtectus per seed and variety and Chi-test output of expected and observed frequencies of emergence holes per seed

Bean variety	***Emergence holes/seed**	x^2	**d.f**	**Probability**
Mwitemania	1.4±0.7	24.69	7	*P=0.001*
Rose coco	1.7±0.5	228.53	7	*P=0.001*
Nyayo	1.5±0.2	85.11	7	*P=0.001*
Canadian wonder	1.5±0.5	70.69	7	*P=0.001*
Mwezi moja	1.5±0.8	132.61	7	*P=0.001*

x^2=Chi-square, d.f=Degrees of freedom, *(Mean ± SD)

Seed hardness index, thickness of seed testae and seed surface area

Results on seed hardness index of bean varieties indicated that the harness index was significantly different between varieties (F=6.9; df=4, 20; $P<0.001$). Nyayo had the highest (0.019) seed hardness index whereas Canadian wonder and Mwezi moja had the least (0.012) hardness index (Table 4). This meant that Nyayo had the hardest seeds whereas Canadian wonder and Mwezi moja had the soft seeds. Seed hardness index was significantly different between Mwitemania and Nyayo and Mwezi moja (TukeysHSD).

Table 4. Mean (± SD) seed harness index, thickness testae, surface area in bean varieties

Bean Variety	***Seed hardness index(X 10^{-1})**	***Thickness of testa (mm) (X 10^{-2})**	***Surface area(mm^2)**
Mwitemania	1.7±0.3^a	11.5±3^a	171.75±1.7^a
Rose coco	1.7±0.2^b	9.1±0.2^a	195.25±2.5^a
Nyayo	1.9±0.1^b	10.1±0.2^a	175.00±6.0^a
Canadian wonder	1.2±0.5^b	11.4±2^a	258.75±3.1^b
Mwezi moja	1.2±0.1^c	9.9±2^a	277.75±1.7^c

Means in a column followed with the same letter are insignificant

*(Mean ± SD)

Thickness in testae was significantly insignificant between bean varieties (F=1.7; df=4, 20; $P=0.18$). On the other hand**,** results on surface area of bean seed varieties indicated Mwezi Moja had the largest (278mm^2) surface area whereas Mwitemania had the lowest (171mm^2) surface area (Table 4). The surface area was significantly different between varieties (F=608.8; df=4, 20; $P<0.001$). All varieties were different from each other (Tukeys HSD).

Results of regression analysis comparing susceptibility index and seed hardness index and susceptibility index and surface areas of each bean variety shows that, in all the varieties there was linear relation between the variables. The linear relationship in susceptibility index and seed hardness was insignificant in Mwitemania, Nyayo and Rose coco (r=0.148-1.24, t=0.148-0.78, *P=0.10-0.517*) and significant in Mwezi moja and Canadian wonder (r=3.29-4.63; t=2.13-4.56; *P=0.011-0.045*). Similarly, the linear relationship in susceptibility index and surface area was insignificant in Mwitemania, Nyayo and Canadian wonder (r=0.148-1.24; t=0.028-0.068; t=0.25-1.85; *P=0.241-0.825*) and significant in Rose coco and Mwezi Moja(r=0.135-0.094; t=6.25-10.28; *P=0.01-0.025*). It is important to note here that the thickness of testae was not subjected to regression analysis since there was insignificant differences in thickness of testae between bean varieties.

Discussion

Generally, many bruchid beetles are known to be selective with regards to variety and size of beans on which they deposit eggs (Hall *et al.,* 2003). The initial contact between bruchids and host plants is during egg laying process where the insect pest's acceptance or non-acceptance of host plant as oviposition site will affect degree of susceptibility or resistance of the host plant species. If the host plant becomes a suitable oviposition site then this will result in laying of many eggs, population increase and more damage to the plant. On the contrary, any unfavorable conditions will result in insect laying fewer eggs, harmful effects resulting slower population increase and lesser damage to plant. Therefore, the appropriateness of the host plant as an oviposition site for the insect pest will be an indicator of suitability for development and survival of the young generation (War *et al.,* 2017). From the results of this investigation it was found that beetles were attracted to varieties with large seeds compared with those with small seeds. This was supported by the fact that the number of beetles attracted to beans was higher where bean varieties had large seeds. It is therefore possible that *A. obtectus* exhibited the same behavior and tended to select bean varieties 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, Keneni *et al.,* (2011) demonstrated that number of larval stages and adult of insect species attracted to a cultivar when given a free choice could be used as a measure of susceptibility or resistance of the cultivar to the insect pest. Thus, it is possible that the relatively few beetles attracted to some varieties could be an expression of variable susceptibility to the beetles.

Reproductive decisions such as oviposition in many insects' species can determined by availability of food and population density, consequently affecting survival fitness of the species. In many species insects females oviposit eggs on areas with plenty of food resources or they may aggressively collect food into a central zone on which to ultimately lay eggs (Janz *et al.,* 2005, Paukku and Kotiaho, 2008). Generally, it is known that survival of larvae of bruchids is differentially affected by size and quality of beans and that selective pressures could favor females that discriminate among bean varieties and avoid overloading a given bean (Paukku and Kotiaho, 2008). In the present study, it is likely that beetle avoided overloading seeds and therefore relatively large seeds could have been preferred. Results of the present investigations are similar with those of other researchers that total number of eggs laid by *A.* obtectus differ with variety and conditions. For instance, it has been reported that a female is capable of laying 61.6+2.2 eggs in her life time at 28^{o} and 70% relative humidity (Parsons and Gredland, 2003). Similar results have also been obtained by Savkovic *et al.,* (2019) that the female lay an average of 42.29 eggs. Investigations on the effects of bean varieties on fecundity of the beetles revealed that the beetles laid more eggs on some varieties than others and eggs laid ranged between 41.9 and 85.4. Therefore, the differences of eggs reported in this study and those of previous researches could be attributed to differences in bean varieties and experimental conditions used because the bean varieties used here are different from those of previous researchers.

It is known that bruchids that feed on seeds, the number of eggs laid and emergence holes of progeny per seed follow a Poisson probability distribution (Siemens and Johnson, 1996; Parsons and Gredland, 2003, Paukku and Kotiaho, 2008). The median number of eggs per seed in many bruchids will range from 4 to 8.5, even though many seeds will have zero eggs. Furthermore, Ebinu *et al*. (2016) found that *A. obtectus* damaged 77% of seeds, with seeds bearing 1 - 3 exit holes comprising 55% of the total damage. In contrast, *Z. subfasciatus* damaged 93% of the seeds, with seeds bearing more than 4 exit holes constituting 58% of the damage. Therefore, the number of emergence holes per seed may be used as a measure of susceptibility/resistance of legumes to damage by bruchids. If data of expected frequencies of emergence holes per seed is analyzed using Chi-square test is found to be statistically significant, this may imply that the development of beetles is variable in different varieties. In the current study the results show that the varieties had variable emergence holes per seed. From the results, it can be deduced that in *A. obtectus* expected frequencies of emergence holes per seed per variety analyzed using Ch-qsuare were statistically significant. The implications of these is that the development of *A. obtectus* in the bean varieties was variable. However, the development of beetles in bean varieties could also be due to toxic or inhibitory biochemical constituents. Therefore, the number of emergence holes may be used to support other data (Kusolwa, 2007; War *et al.,* 2012).

Furthermore, it has been found that characteristics of a host species of an insect may prevent oviposition by failing to provide the appropriate stumuli that finally results in deposition of eggs. In other research reports, oviposition in *A. obtectus* has been found to be influenced within a particular cultivar of beans by minute differences, for example size and harness of seed, seed coat roughness and stoutness (Somta *et al.,* 2007; Keneni *et al.,* 2012; War *et al.,* 2012). Therefore, the differences in number of eggs laid between varieties could be due to failure by some of the varieties to provide the complete releasing stimuli that could have resulted in deposition of more eggs. It is also noted in this study that the majority of females laid their eggs within one week of adult emergence. The above observations support the findings of Parsons and Gredland, (2003) who found that eggs were laid within a week of adult emergence. Beans have been reported to regulate reproductive activity in *A. obtectus* and that in absence of a suitable medium, females delay oviposition by maintaining 30 to 40 developing oocytes in their oviducts until a preferred host is available. Therefore, the fact that egg laying duration was variable could suggest that some varieties induced egg laying more than others. 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 females with the hope of finding a better host. Furthermore, it has also been reported that chemical signals emanating from beans are captured by sensory structures of these beetles and they perhaps control reproductive processes through the nervous system (Mitchel 1975; Parsons and Gredland, 2003).

The total number of progeny that eventually emerged were significantly different between varieties. Several factors could have been responsible for the bean 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 center of the seed such that progeny is unable to emerge after pupation (Keneni *et al.,* 2011). Furthermore, Desroches *et al.,* (1995) found in *Vicia faba* the seed coat may act as an obstacle for entry by *C. maculatus* and *C. chinensis* larvae. They observed that about 45–58% of the larval stages penetrated the seed coat to reach the cotyledons. A related kind of was resistance also observed by Eddie et al. (2003) *C. maculatus* and cowpeas. On the other hand, resistance to *C. maculatus* in three varieties of cowpea has been found to be contributed mostly by a blend of decreased egg laying and hatching which may be a manifestation of biochemical other morphological traits of the seed (Lale and Kolo, 1998).

It is likely that some varieties used in this study were unsuitable food media so that some progeny failed to emerge. Thus the differences in number of progeny emerging from different bean varieties. The duration taken by 50 % of progeny to emerge was significantly different between varieties. It is known that the duration of the life cycle is a good measure of suitability of an environment for growth and development of an insect. In general, adverse conditions prolong developmental period and few adults survive (Schoonhoven and Cardona, 1983; Parsons and Gredland, 2003; Paukku and Kotiaho, 2008). Therefore, it can be suggested that the period of the development of the bean bruchids was a measure of the suitability of 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 also been reported to retard growth and development and to kill larvae of various bruchid species. The manifestation of resistance to bean bruchid in bean cultivars is usually associated with biochemical components of the beans such as protease inhibitors phytohemagglutinins, and alpha-amylase. In addition, different forms of the protein arcelin, is associated with deleterious effects on larval stages and adult insects. Defence lectins found accumulated in legume seeds are also known to be anti-nutritional may affect development of larvae in bruchids (Dobie, 1990; Edwards and Singh, 2006; Baldin *et al.,* 2008; Zaugg *et al.,* 2012). Despite the presence of such biochemical constituents and others were not being investigated, it possible that there were certain chemical components which may have retarded growth and development. Therefore, the differences in developmental period of beetles in various bean varieties could be attributed to the existence of such chemical components.

In this study susceptibility indices were considered as indicators of different degree of tolerance of bean cultivars to damage by *A. obtetus* (Gudrups *et al.,* 2001). The greater the susceptibility index, the higher the vulnerability of the beans to damage by *A. obtectus*. The determination of the susceptibilities of the bean cultivars to infestation by *A. obtectus* using a susceptibility index relating duration of development and progeny revealed that the bean varieties showed significant differences to their susceptibilities to damage by the bean bruchids. Indeed, Desroches *et al.,* (1998) attributed the differences to thickness to testae. In other findings, Dobie *et al*., (1990) also suggested that resistance could be affected by the incapacity of the larvae stages to enter the seed coat or by biochemical characteristics. In other studies, resistance was associated biochemical arcelins, alpha amylase inhibitor and phytohaemagglutinins (Kusolwa, 2007; Keneni *et al.,* 2011, Zaugg *et al.,* 2012). Although, the presence of such biochemical constituents and others were not investigated, it possible that there were certain chemical components which may have retarded growth and development and probably killed some of the larvae. Therefore, the differences in developmental period, number of progeny emerged and consequently differences in susceptibilities could be reflections of such varietal differences.

Physical characteristics of beans have also been reported to be responsible for their resistance to insects of grain legumes. Genetic variability of grains has been associated with various agronomic characteristics, such as seed size, color, and texture (Beaver *et al.,* 2003; Hall *et al.,* 2003; Sibakwe and Donga, 2015). The above physical characteristics of the seed are responsible for differences in susceptibility to bruchids among different bean varieties compared to medium seeded and large seeded varieties (Mei *et al.,* 2009). Furthermore, Gudrups *et al.,* (2000) found that the size of the kernel in maize varieties is the most important morphological feature influencing resistance to infestation by *S. zeamais.* The maize cultivars with large kernels were relatively more resistant compared than smaller ones. The phenomenon 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 bean bruchids (Beaver *et al.,* 2003, Hall *et al.,* 2003, Sibakwe and Donga 2015). In the present study, it was found that some physical characteristics such as size and hardness of seed affected susceptibility. For instance, seed hardness affected susceptibility in Rose coco and Canadian wonder whereas seed surface area also affected susceptibility in Rose coco and Mwezi moja.

Conclusion

The study showed that the *A. obtectus* displayed apparent marked preference for certain bean varieties for oviposition purposes. Fecundity and number of progeny emerging and was also affected by bean varieties. Contrary to expectation, observed and expected number of emergence holes per seed departed from those expected in a homogenous distribution an indicator of variable development of *A. obtectus* in bean varieties. Duration taken by 50% of progeny to emerge and Susceptibility indices were also different between varieties. Furthermore, physical traits of bean varieties for instance seed hardness and surface area of bean varieties contributed to susceptibility to attack by *A. obtectus*. Overall, considering all the factors studied, Mwezi moja may be considered the most susceptible variety while Mwitemania was the more tolerant variety to infestation by *A.obtectus*. From the findings of this study, the authors recommend more research to be carried out especially on chemical components of bean varieties in order to understand the additional causes of the differences in the above varieties. More field studies may also be carried out on yield, consumer preferences, structure, size, and nutritional content of seed, plant type and the maturity period among others. Armed with such information, a variety/varieties could be bred and selected and recommended for cultivation in different regions.

Acknowledgement

We wish to appreciate the support of Dr. Symon K. Kiprop of Department of Economics, Egerton University, Kenya for assisting in statistical data analysis. In addition, we wish to thank all the staff of Integrated Biotechnology, Egerton University, Kenya for the technical support during the study.

Ethical approval

This article did not involve experiments on human subjects or vertebrate animals. Therefore, ethical approval is not applicable

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