Journal of Science and Technology, Vol. 31, No. 3 (2011), pp 11-20 11

http://dx.doi.org/10.4314/just.v31i3.2

RESEARCH PAPER

SUSCEPTIBILITY OF SOME KERSTING'S GROUNDNUT LANDRACE CULTIVARS TO INFESTATION BY *CALLOSOBRUCHUS MACULATUS* (FAB.) [COLEOPTERA: BRUCHIDAE]

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ABSTRACT:
Seeds of five different landrace cultivars of Kersting's groundnut, Macrotyloma geocarpum (Harms) Marechal and Baudet, obtained from northern Ghana, were evaluated for their susceptibility to infestation and damage by the pulse beetle, Callosobruchus maculatus Fab. The completely randomized design was used to obtain data on the oviposition, developmental period, progeny emergence, weight loss in seeds and susceptibility index. The results showed that the Nakpanduri-white, Heng-mottle and Damongo-cream landrace cultivars proved to be the highly preferred hosts to C. maculatus, recording the highest egg load and progeny emergence, as well as the shortest egg-adult developmental period, and highest seed weight loss. The Najung-black landrace cultivar was the least preferred, followed by the Nakori-brown cultivars. Results from susceptibility indices further indicated that the Najung-black and Nakori-brown were the least susceptible to C. maculatus, while Nakpanduri-white was the most susceptible. Overall, the Najung-black and Nakori-brown landrace cultivars consistently demonstrated high tolerance to the pest and therefore, should be promoted or incorporated into breeding programmes to help minimize the high losses incurred by farmers during storage.

Keywords: *Kersting's groundnut, Landrace cultivars, Callosobruchus maculatus, Susceptibility.*

INTRODUCTION

Kersting's groundnut, *Macrotyloma geocarpum* (Harms) Marechal and Baudet, is an important grain legume cultivated in parts of West Africa (Duke *et al*., 1977). It is a neglected and underutilized crop believed to have originated from Nigeria where it is cultivated widely (Hepper, 1963; Okigbo, 1992). In Ghana, the crop is mainly produced along the guinea savanna belt, particularly in the Northern, Upper East and Upper West regions (Bayorbor *et al*., 2010). In most communities, Kersting's groundnut is produced mainly for its mature bean-like seeds which have high nutritional value and medicinal significance (Obasi and Ezedinma, 1991; Obasi and Agbaste, 1994). However, a major constraint to the postharvest preservation of Kersting's groundnut seeds in the savanna ecology of Ghana is infestation by the storage bruchid, *Callosobruchus maculatus* Fab., which is capable of rendering unprotected seeds unviable and unsuitable for utilization as food, within few months of storage (Amponsah, 2007). The control of this pest is crucial to safe storage, sustainable production and utilization of the crop among rural households in northern Ghana.

While there are several synthetic insecticides such as chemical grain protectants and fumigants for the control of this pest, their use has not been sustainable owing to their high costs, unavailability in local markets and associated health and environmental risks (Wolfson *et al*., 1991). The need to minimize over-reliance on synthetic insecticides for pest control in Kersting's groundnut has, thus, called for the need to search for resistant strains against bruchid infestation in storage. While improved varieties for Kersting's groundnut are yet to be developed in the ecology, existing landrace cultivars with resistant traits can serve as viable alternative to bruchid pest management (Asante and Mensah, 2007; Olakojo *et al*., 2007). Redden (1983) studied the inheritance of seed resistance factor to *C. maculatus* in cowpea and concluded that the trait is inherent in a recessive manner. According to Redden and McGuire (1984), host legume resistance to bruchids is conditioned by major genes with the presence of modifiers, and that trypsin inhibitors are associated with the resistance.

The development and use of resistant Kersting's groundnut cultivars will, thus, offer a simple and attractive means for reducing bruchid damage, since it requires little knowledge by farmers, free extra cost to farmers, and also increases the effectiveness of other pest management strategies such as cultural and biological control (Asante and Mensah, 2007). Hence, it is pertinent that bruchid responses to existing Kersting's groundnut landrace cultivars be known in order to make resistant landraces available for cultivation, and recommend susceptible ones for genetic improvement. Five different landrace cultivars of Kersting's groundnut were identified by Bayorbor *et al*. (2010) in the three northern regions of Ghana. These have been known to be high-yielding and adaptable to biotic and abiotic stresses in the ecology. This study evaluated the differences in susceptibility of their seed grains to infestation and damage by *C. maculatus*.

MATERIALS AND METHODS

Source of Kersting's Groundnut Landrace Cultivars

Seed grains of five different landrace cultivars of Kersting's groundnut were used for the study in the Biology Laboratory of the University for Development Studies, Nyankpala in the Northern Region of Ghana, between January and March, 2010. The landrace cultivars included local germplasm lines collected from farmers' fields around the Northern, Upper East and Upper West Regions of Ghana, during the 2009 harvesting season. These were identified on the basis of their places of origin and seed coat colours, namely Najung-black, Nakori-brown, Damongo-cream, Heng-mottled and Nakpnduri -white (Table 1).

These cultivars served as the indigenous genetic resource base which commenced to be regarded as the raw material for identifying useful traits for genetic combination and improvement. One of the commonly cultivated cowpea varieties in the ecology, known as Bengpela was used as a control.

The seeds were stored in the laboratory's cold room maintained at 10°C, 70-100% relative humidity (RH) to ensure that they were free

Table 1: Test landrace cultivars of Kersting's groundnut in northern Ghana

Landrace cultivars	Place of collection/origin	Seed coat colour
Najung-black	Najung, Upper East Region	Black
Nakori-brown	Nakori, Upper West Region	Brown
Damongo-cream	Damongo, Northern Region	Cream
Heng-mottle	Heng, Upper West Region	Mottle
Nakpanduri-white	Nakpanduri, Northern Region	White

from attack by any postharvest insect or pathogen. They were later conditioned to room temperature before being used for the experimental purposes.

Rearing of Experimental Insects

Experimental beetles were reared for both choice and no-choice experiments. Rearing of insects for the no-choice experiment followed the procedure described by Swella and Mushobozy (2007). The adults of *C. maculatus* were originally obtained from infested samples of Kersting's groundnut seeds in a laboratory stock. They were reared and bred under diet of Kersting's groundnut seeds inside a growth chamber at a temperature of 27 ± 2°C and 70 ± 5% RH. Initially, 50 pairs of newly emerged (1-24 hrs old) adults were placed in jars containing the various seeds. The jars were covered with perforated lids to allow for aeration, and a maximum of 3 days were allowed for mating and oviposition. The parent insects were removed afterwards and the seeds containing the eggs were transferred to fresh seeds in rearing jars which were covered with pieces of cloth, fastened with rubber bands to prevent the contamination of the seeds and escape of the beetles. The subsequent progenies emerging from the stock were then used as parental generation for the experiment.

Experimental insects for the choice experiment were reared from the culture of insects for the no-choice experiment described above. One glass jar each of a capacity of 1 kg contained respective seeds of the five Kersting's groundnut landrace varieties, and the cowpea. The aim was to precondition the bruchids so as to eliminate any short term changes in behavior associated with the change of host variety from that used for culturing to that being tested (Dobie 1974). The rearing procedure followed the method described by Swella and Mushobozy (2009).

Experimental Design and Procedure

The experimental procedure used for this study was based on Asante and Mensah (2007), Adam and Baidoo (2008) and Swella and Mushobozy (2009), with some modifications. For the no-choice experiment with seeds of the five different landraces of Kersting's groundnut, a completely randomized design (CRD) with four replications was used. For a choice experiment, seeds of the five landraces were mixed in all possible pairings, and each replicated four times.

Two hundred (200) sound seeds of each cultivar were then placed in a glass jar after being weighed to determine their initial weight. Five pairs of the bruchids (24 hrs old) emerging from the insect culture were selected and introduced into each seed sample in the glass jar. The jars were then closed with perforated covers and kept in an incubator maintained at conditions described above. The insects were then allowed for 48 hours to mate and lay eggs after which they were removed from the jars. Observations were then made for a maximum of four weeks during which period the appropriate data were collected. The observations were terminated 27 days from the date of the first adult emergence after which the final weight of seeds in each treatment was determined.

Data Collection and Analysis

The variables that were determined from the experimental set up were: number of eggs laid, mean development period, adult emergence, seed weight loss and susceptibility index. The number of eggs laid on the seeds of each sample were counted separately by following the method described by Lambert *et al*. (1985), and recorded for each treatment 7 days after the infestation, by which time most eggs had hatched and the larvae had bored into the seeds, leaving behind the cream-coloured shells. The various treatments were then examined daily for adult emergence (i.e. proportions of adults that emerged from the number of eggs laid on the seeds, including hatched and unhatched) following the method of Asante and Mensah (2007). The emerged adults were removed from each sample with an aspirator and counted daily under illuminated magnifier. Mean development period was recorded as the time period taken for the insects to develop from egg to adult stages. Percentage weight loss (PWL) in seeds was calculated using the method of Jackai and Asante (2003) as follows:

$$Percentage\ weight\ loss\ (PWL)\ = \left(\frac{Initial\ seed\ weight - Final\ seed\ weight}{Initial\ seed\ weight}\right)100\%$$

Index of susceptibility (SI) was determined using the formula given by (Dobie 1974):

$$SI = \left(\frac{Log_e F_I}{D}\right)100\%$$

Where, F_I = Total number of emerging adults, and D = Median developmental period (estimated as the time from the middle of oviposition to the emergence of 50% of the F_I generations).

Differences in susceptibility of seeds in the various treatments were examined based on the parameters estimated above, using the one-way analysis of variance (ANOVA). Numerical and percentage data were log and arc sine transformed respectively, before the analysis. Where AVOVA test indicated significant difference between the treatments, the least significant difference test (LSD) was used to separate the means.

RESULTS

Egg oviposition

Table 2 presents the mean number of eggs laid by the adult females of *C. maculatus* on seeds of the different Kersting's groundnut landrace cultivars in the no-choice experiment. The results showed that number of eggs laid ranged from 15.0 in the Najung-black to 52.7 in the Nakpanduri-white cultivar. Mean egg oviposition was highest (68.4) in the control treatment. Egg oviposition was significantly affected ($p < 0.005$) by the seeds of the different landrace cultivars. The number of eggs laid on seeds of the control was significantly higher ($p < 0.01$) than that of any of the cultivars. Among the landrace cultivars, seeds of Nakpanduri-white recorded the highest egg load and this was significantly different from any of the other cultivars ($p < 0.01$). The Najung-black and Nakori-brown landrace cultivars recorded the lowest numbers of eggs laid, and these were also significant when compared with that of the Damongo-cream or Heng-mottled.

The relative ovipositional preference by *C. maculatus* in the choice experiment indicated that among the landrace cultivars, mixtures containing the white and black seeds recorded the maximum and minimum number of deposited eggs, respectively. Seeds mixtures containing the control however, recorded the heaviest egg load (Table 3).

Table 2: Number of eggs laid and developmental period of *C. maculatus* on seeds of different Kersting's groundnut landrace cultivars in northern Ghana

Landrace cultivars	Mean number of eggs laid on seeds n = 50	Mean developmental period (days)
Najung-black	15.0 ± 0.8^{a}	27.5 ± 1.0^{a}
Nakori-brown	20.5 ± 1.0^{a}	26.3 ± 0.8^{ab}
Damongo-cream	31.0 ± 1.3^{b}	24.0 ± 0.6^{bc}
Heng-mottle	37.2 ± 1.4^{b}	24.4 ± 0.6^{c}
Nakpanduri-white	52.7 ± 1.9^{c}	21.8 ± 0.3^{d}
Control	68.4 ± 2.2^{d}	21.0 ± 0.2^{d}
LSD (5%)	*9.5*	*2.4*

n = number of eggs laid
Column means followed by different letters are significantly different at p < 0.05.

Table 3: The relative ovipotitional preference by *C. maculatus* on paired seed mixtures of different Kersting's groundnut landrace cultivars in northern Ghana

*Landrace cultivars	NJ-black	NK-brown	DG-cream	HG-mottled	ND-white	Control
NJ-black	9.2	12.5	10.0	13.5	14.0	12.7
NK-brown	13.4	13.6	14.1	16.0	19.5	13.0
DG-cream	12.0	14.5	12.4	15.8	15.0	17.7
HG-mottled	11.0	16.2	18.6	13.1	19.1	16.0
ND-white	14.0	19.0	22.5	21.0	34.3	28.3
Control	15.8	17.1	23.9	20.5	19.6	25.2

**NJ = Najung; NK = Nakori; DG = Damongo; HG = Heng; ND = Nakpanduri*

Developmental Period and Progeny Emergence

The mean developmental period of *C. maculatus* on seeds of the different Kersting's groundnut landrace cultivars are presented in Table 2. Egg-adult developmental period was found to range from 21.8 days on Nakpanduri-white to 27.5 days on Najung-black, and 21.0 days on the control. Analysis of variance indicated significant differences among the treatment means ($p < 0.02$). Among the Kersting's groundnut cultivars, significantly higher number of days was taken by *C. maculatus* to develop in seeds of Najung-black or Nakori-brown. However, developmental period in Damongo-cream and Heng-mottle landraces did not differ significantly. Moreover, significantly lower number of days was taken by the pest to develop on the Nakpanduri-white compared with the other cultivars. Developmental period in Nakpanduri-white was not significantly different from that of the control treatment.

The mean number of emerged adult weevils was lowest (15.2) in Najung-black landrace and highest (41.9) in Nakpanduri-white. Adult emergence was highest (52.0) in the control than all the five landrace cultivars (Table 4). There was significant difference ($p < 0.001$) among the treatments. Mean adult emergence in the Nakori-brown was significantly lower than that of the Damongo-cream or Heng-mottled. Weevil emergence in Heng-mottled was significantly lower than that of Nakpanduri -white, but statistically similar to that of Damongo-cream. Also, the number of emerged weevils from the control was significantly higher than that of Nakpanduri-white. Even though the percentage progeny emergence did not differ significantly ($p< 0.15$), the Heng-mottle and Najung-black recorded the highest (84.9) and lowest (74.0) percentage of emerged weevils, respectively (Table 4).

Seed Weight Loss and Susceptibility Index

The resultant weight loss in seeds due to *C.*

Table 4: Mean progeny emergence of *C. maculatus* from seeds of different Kersting's groundnut landrace cultivars in northern Ghana

Landrace cultivars	Mean number of adults emerged	Percentage progeny emergence
Najung-black	15.2 ± 0.5^{a}	74.0 ± 2.8
Nakori-brown	20.5 ± 0.8^{a}	76.6 ± 2.0
Damongo-cream	30.0 ± 1.3^{b}	83.3 ± 3.0
Heng-mottled	31.6 ± 1.0^{b}	84.9 ± 3.3
Nakpanduri-white	41.9 ± 2.1^{c}	79.5 ± 2.8
Control	52.0 ± 2.6^{d}	76.0 ± 2.1
LSD (5%)	*7.8*	*Ns*

Column means followed by different letters are significantly different at $p < 0.05$.

Table 5: The weight loss in seeds of different Kersting's groundnut landrace cultivars due to infestation by *C. maculates*

Landrace cultivars	Weight loss in seeds (g)	% weight loss
Najung-black	1.80 ± 0.05^{a}	8.0 ± 0.60^{a}
Nakori-brown	1.60 ± 0.08^{a}	9.3 ± 0.88^{a}
Damongo-cream	1.55 ± 0.07^{a}	9.1 ± 0.71^{a}
Heng-mottled	1.50 ± 0.30^{ab}	9.3 ± 1.25^{a}
Nakpanduri-white Control	2.40 ± 0.28^{c}	14.4 ± 1.00^{b}
LSD (5%)	3.47 ± 0.55^{d}	18.5 ± 1.77^{c}
	0.50	*4.0*

Column means followed by different letters are significantly different at $p < 0.05$.

maculatus infestation was found to be significantly affected by the different Kersting' groundnut landrace cultivars ($p < 0.03$). Seed weight loss was highest (2.4) in Nakpanduri-white and lowest (1.5) in Heng-mottled (Table 5). Seed weight loss in the control was higher (3.5) than any of the five landrace cultivars. Nakori-brown recorded a statistically similar weight loss compared with Najung-black or Damongo-cream. Seed weight loss in Heng-mottled was significantly lower than that of Nakpanduri-white, but statistically similar to that of Damongo-cream, Nakori-brown and Najung-black. Moreover, weight loss in seeds of the control was significantly higher than that of the Nakpanduri-white. Percentage weight loss in seeds indicated that Nakpanduri-white and Najung-black recorded the highest (14.4) and lowest (8.0) percentage weight loss, respectively with significant difference existing between Heng-mottled and Nakpanduri-white (Table 5).

The susceptibility indices (SI) of the different landrace cultivars to infestation by *C. maculatus* are presented in Table 6. SI was found to range from 4.3 in Najung-black to 10.0 in Nakpanduri-white, and 12.5 in the control. There were significant differences ($p < 0.003$) even among the landrace cultivars. SI of the Najung-black or Nakori-brown cultivars was significantly lower than that of Damongo-cream or Heng-mottle. The Nakpanduri-white cultivar recorded a significantly higher SI value compared with the other landrace cultivars. Overall, the highest susceptibility index was recorded in Nakpanduri-white while Najung-black recorded the lowest susceptibility index.

Table 6: The susceptibility indices of seeds of different Kesting's groundnut landrace cultivars to infestation by *C. maculatus*

Kersting's groundnut seeds	Susceptibility index
Najung-black	4.3 ± 0.15^{a}
Nakori-brown	5.2 ± 0.42^{a}
Damongo-cream	7.3 ± 0.78^{b}
Heng-mottled	7.2 ± 0.85^{b}
Nakpanduri-white	10.0 ± 1.34^{c}
Control	12.5 ± 1.90^{d}
LSD (5%)	*2.00*

Column means followed by different letters are significantly different at $p < 0.05$.

DISCUSSION

The results of this study have indicated that the egg deposition and development of *C. maculatus* on Kersting's groundnut seeds were significantly affected by the landrace cultivars. Egg deposition and development was significantly inhibited by the Najung-black and Nakori-brown cultivars, but significantly increased by Nakpanduri white (Table 2). This could be attributed to the differences in physiochemical characteristics of the seeds (Nwanze *et al.,* 1975). Adam and Baidoo (2008) observed that the nature and seed coat hardness is a major determinant for oviposition by *C. maculatus* on cowpea. According to Mbata (1992), legume varieties with smooth seed surfaces are more preferred for oviposition by *C. maculatus* compared to rough-coated ones. The Nakpanduri-white seeds actually had smoother coats compared with either Najung-black or Nakori-brown, which appeared more rough and wrinkled. Previous report by Nwanze *et al.* (1975) have also shown that the number of eggs oviposited by *C. maculatus* was found to be affected by seed size, curvature, colour, smoothness and thickness.

Moreover, Patil and Jadhav (1985) reported that the weight and volume of legume seeds could be additional factors responsible for oviposition preference. Seed hardness is due to chemical composition of the seed coat, which may have an effect on seed invasion by *C. maculatus* (Asiedu *et al.,* 2000). Differences in seed coat hardness and colour have been associated with differences in chemical composition of seeds of pulses (Adam and Baidoo, 2008). According to Obasi and Agbatse (1994), the chemical compounds found in Kersting's groundnut seeds include tannins, nontannins, lignins and polyphenolic compounds. The concentration of these compounds may differ depending on the level of colour pigmentation of the seed coat (Asiedu *et al.,* 2000). Morrison *et al*. (1995) reported high lignin levels in pigmented cowpea varieties, and this may be a factor conferring resistance. Coloured seed coats in the Kersting's groundnut landrace cultivars used in the study probably contained more of these chemical compounds and thus, making them unsuitable for oviposition than their white-seeded counterparts.

Ndlovu and Giga (1988) reported that the pattern of adult emergence and percentage adult emergence of *C. maculatus* in resistant cowpea ones were characterized by delayed, staggered and slow adult emergence while in the susceptible lines, adult emergence was relatively early and extremely rapid. This might explain the prolonged egg development and few progenies emerging from the Najung-black and Nakori-brown landrace cultivars recorded in this study. Obasi and Abgatse (1994) recorded higher protein content in dark-seeded Kersting's groundnut cultivars compared to the light-seeded types. The inability of the beetles to develop on the dark Kersting's groundnut seeds could be attributed to the high protein-carbohydrate ratio of the seeds, and in part, to their saponin content. This finding is in conformity with that of Applebaum *et al*. (1969) who noted that bean bruchids are not capable of attacking seeds with high fat content. The results also agreed with those of Swella and Mushobozy (2009) who reported that legume seeds with higher mean egg counts and percentage adult emergence correspondingly had shorter development period (Table 2).

Weight loss arising from the quantity of material consumed by the developing larvae was found to correlate positively with the susceptibility index. The differences in susceptibility of the various seeds showed their suitability or otherwise, as hosts for oviposition, feeding and development of *C. maculatus*. Mbata (1992) reported that weight loss in cowpea seeds was generally correlated with SI. It is possible that the number of emerging bruchids determines the extent of bruchid damage, as seeds permitting more rapid and higher levels of beetle emergence were more prone to infestation and damage by the storage bruchids. The findings also agree with those of Asante and Mensah (2007) who reported that cowpea cultivars that recorded high percentage emergence of *C. maculatus* suffered the greatest damage, weight loss and SI. Also, Swella and Mushobozy (2009) reported that SI has a positive correlation with bruchid emergence in different legume seeds.

Available reports have also shown that trypsin inhibitors are important factors conferring resistance to bruchid infestation in many pulse seeds (Baker *et al*., 1989). According to Borchers *et al*. (1947) the presence of trypsin inhibitors in leguminous seeds affect the ability of bruchids to digest proteins contained in the seeds. Thus, what confers resistance to a variety should be an intrinsic property that combats development even after *C. maculatus* oviposition. The results in the present study appeared to suggest that black- or dark-seeded landrace cultivars of Kersting's groundnut contain more inhibitors of trypsin compared to those of the white- or light-coloured seeds .

CONCLUSIONS

The results of this study have indicated that landrace cultivar variability in Kersting's groundnut seeds has a significant effect on the infestation and damage caused by *C. maculatus*. The Najung-black and Nakori-brown landrace cultivars were the least susceptible to infestation by *C. maculatus*, followed by Damongo-cream and Heng-mottle cultivars.

The Nakpanduri-white landrace cultivar was the most susceptible to the pest. This might be due to the difference in the various morphological and chemical properties of the seeds as discussed above. The cowpea seeds used as check however, proved to be the most preferred and susceptible host for *C. maculatus*. The Najung-black and Nakori-brown landrace cultivars consistently demonstrated high tolerance to the pest and therefore, should be promoted or incorporated into breeding programmes to help minimize the high losses incurred by farmers during storage. White- or light-seeded landrace cultivars would require more protection in storage in order to preserve seed quality.

Acknowledgements

This research was supported by the International Foundation for Science, Stockholm, Sweden, through a grant to Benjamin K. Badii.

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