Effect of Seed Powder of Three Pepper Species on the Bionomics of Cowpea Bruchid, *Callosobruchus maculatus* Fabricius

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Abstract- Seed powder of *Afromomum melegueta*, *Piper guineense* and *Xylopia aethiopica* at the rates of 0.5, 1.0 and 1.5 g/ 20 g seeds of cowpea were assessed for insecticidal activities against *Callosobruchus maculatus* Fabricius in the laboratory under fluctuating ambient temperature and relative humidity in Makurdi, Benue State, Nigeria. *P. guineense* caused the highest toxicity (68.53%) at 1.5 g/ 20 g cowpea seeds after 96 hours of exposure. Oviposition deterrency, larvicidal effect and suppression of adult emergence recorded for *P. guineense* was considerable but failed to match the conventional synthetic insecticide, Permethrin ($P > 0.05$). *A. melegueta* at test concentrations and at short storage duration (< 5 days) was not toxic to the bruchid. However, it significantly ($P<0.05$) deterred oviposition in *C. maculatus* females when compared to unprotected cowpea seeds. In the study, *X. aethiopica* protected cowpea seeds better than *A. afromomum* and gave minimal support to the bionomics of the bruchid. The study seriously suggests an increased need for search for botanical insecticides that can effectively control the resistant cowpea bruchid. Any natural material that cannot give good control of *C. maculatus* at an economically justified concentration in 24 hours may not be sustainable.

Index Terms- Afromomum melegueta, Piper guineense, Xylopia aethiopica, Oviposition deterrency

I. INTRODUCTION

Cowpea, *Vigna unguiculata* (L.) Walp is an important legume crop of Africa (Agboola, 1997). Nigeria accounts for 70 % of the world's cowpea production (Blade *et al*., 1997). Cowpea seeds are rich in protein and amino acids and therefore serve as a valuable source of protein for human consumption (Quin, 1997; Somta *et al*., 2008). In addition, the crop is a source of livestock feed in the tropics (Adedire and Lajide, 1999) and a source of revenue (Alghali, 1991).

It is attacked by a number of field and post harvest pests with *C. maculatus* (Coleoptera: Bruchidae) ranked as the most notorious post harvest pest (Caswell and Akibu, 1980). Damage and loss in stored cowpea due to *C. maculatus* infestation is a very serious problem to farmers and traders (Rees, 2004). Infestation culminates in substantial reduction in the quantity and quality of seed. Damaged cowpea seeds are unsuitable for human consumption and cannot be effectively used for agricultural and commercial purposes (Somta *et al*., 2008).

Synthetic insecticides have for been used to effectively control cowpea bruchids (Somta *et al*., 2008). However, economic, health and environmental implications has necessitated reorientation of research focus to the development of alternative control agents that are relatively cheap, easy to apply, safe and eco-friendly (Adedire and Lajide, 1999). Already, plant materials have been investigated for *C. maculatus* management in stored cowpea and some of the results are encouraging (Adedire and Lajide, 1999; Sarfraz and Keddie, 2005).

Among plant species so far investigated for contact toxicity against bruchids' and weevils' infestations in stored grains, the peppers seem to hold very strong promise (Asawalam and Emosiarue, 2006; Babarinde *et al*., 2008). Therefore, the present study sought to assess the effect of seed powders of three indigenous pepper plants on the toxicity and reproductive fitness of *C. maculatus* infesting stored cowpea.

II. MATERIALS AND METHODS

Insect Culture

Adult *Callosobruchus maculatus* used to establish the culture were obtained from naturally infested cowpea seeds procured from North Bank Market, Makurdi Benue State, Nigeria. The bruchids were mass reared on the susceptible local variety, Sokoto white under fluctuating ambient temperature and relative humidity. Newly emerged adult bruchids (1 – 2 days old) were used for the experiment.

Pepper Species and Powder Preparation

The three pepper species tested were *Afromomum melegueta* Schum (alligator pepper), *Piper guineense* Schum (black pepper) and *Xylopia aethiopica* Dunal (Ethiopian pepper). These species belong to families Zingiberaceae, Piperaceae and Annonaceae respectively. Five grams each of dried seeds of the pepper species were separately ground into powder and used immediately for the bioassays.

Bioassays

Twenty grams of disinfested and standardized (using the method of Sulehrie *et al*., 2003) cowpea variety, IFE BROWN were separately weighed into thirteen highly transparent plastic containers. The first three containers were added sequentially 0.5, 1.0 and 1.5 g of *A. melegueta* seed powder. The fourth, fifth and sixth containers received sequentially 0.5 1.0 and 1.5 g of seed powder of *P. guineense* while the seventh, eighth and ninth containers were added 0.5, 1.0 and 1.5 g of *X. aethiopica* respectively. The tenth, eleventh and twelfth containers received 0.5, 1.0 and 1.5 g of permethrin insecticide (Rambo: 0.6 %

permethrin) respectively. The thirteenth container received no treatment and served as an untreated control. Cowpea seeds were vigorously shaken for adequate mixing with pepper powders and allowed for 15 minutes (Southgate *et al*., 1957) before the introduction of five pairs of freshly emerged male and female adult *C. maculatus* into each container. The containers were covered with muslin cloth (fixed with cut-edge of container lid) for aeration and to prevent exist of the bruchids and entry of unwanted organisms. Each set-up was replicated four times and arranged in completely randomized block design on a laboratory bench at fluctuating ambient temperature and relative humidity. Adult mortality was assessed daily up to 96 hours and the number of dead bruchids was converted to percent mortality.

Six days after treatment with pepper species and synthetic insecticides, all dead (those that did not respond to probe with a pin) and live adult bruchids were removed and discarded. The number of eggs laid in a no choice condition was recorded. Egg count was done on ten randomly selected seeds. Cowpea seeds with hatched eggs/ larvae were examined and recorded and adult emergence was recorded beginning 21 days after oviposition until there was no emergence for three consecutive days (Sulehrie *et al*., 2003).

Statistical Analysis

Data were subjected to analysis of variance (ANOVA) using SPSS statistic 17.0 software. Mean separation was done using new multiple range test (FLSD) as 5 % level of probability.

III. RESULTS

Table1 presents the results of toxicity of seed powder of three pepper species compared to permethrin against adult *C. maculatus*. The results showed that mortality of the cowpea bruchid varied among treatments as well as within rates of a particular pepper species. Adult mortality also varied with post treatment period. There was no contact toxicity on adult bruchids with *A. melegueta* at all tested rates over short storage duration (< 5 days). Among the pepper species, seed powder of *P. guineense* was the most toxic to the bruchid, killing over 50 % in 96 hours at all tested rates, followed by *X. aethiopica* (when compared with results from untreated control). However, over short duration of exposure, no pepper seed powder matched the efficacy of the synthetic insecticide in killing the bruchids (P > 0.05). Only the seed powder of *P. guineense* was half-as-effective as permethrin.

Table 1: Comparative Contact Toxicity of Seed Powder of Three Pepper Species against Adult *Callosobruchus maculatus* Fabricius

Treatment	Rate (g/20 g seeds)	% Mortality at indicated hour*			
		24	48	72	96
A. melegueta	0.5	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a
	1.0	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a
	1.5	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a
P. guineense	0.5	3.33±4.72a	16.66±7.50b	33.33±6.10b	50.00±3.30b
	1.0	10.00±2.30b	28.25±4.74b	19.00±3.80b	58.24±7.00b
	1.5	18.69±2.80b	37.20±1.00b	60.00±0.50b	68.53±0.10b
X. aethiopica	0.5	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a
	1.0	0.00±0.00a	0.00±0.00a	0.00±0.00a	2.50±2.50a
	1.5	1.21±0.10a	4.00±0.10a	6.66±0.20a	7.48±0.21a
Permethrin	0.5	100.00±0.00b	100.00±0.00c	100.00±0.00c	100.00±0.00c
	1.0	100.00±0.00c	100.00±0.00c	100.00±0.00c	100.00±0.00c
	1.5	100.00±0.00c	100.00±0.00c	100.00±0.00c	100.00±0.00c
Untreated Control	0.0	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a

* All values are means of four replicates followed by standard error of the means. Means of the four treatments were compared same test concentration by same test concentration. Means followed by the same letter are not significantly different (P> 0.05) by Fischer's Least Significant Difference (Multiple range test).

Table 2 presents the results of comparative effect of seed powder of three pepper species on oviposition of adult female *C. maculatus* after six days of exposure. Similarly, variations were recorded in the mean number of eggs laid by the cowpea bruchid amidst pepper treatments as well as within tested rates of a particular pepper. Oviposition deterrence among pepper species increased with treatment rate. Eggs were not laid on cowpea seeds protected with permethrin in six days. On the contrary, unprotected seeds (in a sample of ten) had about 162 eggs. The most effective pepper seed powder for oviposition deterrence against gravid *C. maculatus* was *P. guineense*. Among the pepper species tested, highest number of eggs was laid on cowpea seeds treated with *A. melegueta.*

Table 2: Comparative Effect of Seed Powder of Three Pepper Species on Oviposition of Female *Callosobruchus maculatus* Fabricius in a No choice Condition

Treatment	Rate (g/20 g seeds)	Number of Eggs Laid* (n** = 10)
A. melegueta	0.5	45.00±1.50a
	1.0	44.00±0.95a
	1.5	40.23±3.34a
P. guineense	0.5	33.67±6.32a
	1.0	25.00±7.50b
	1.5	14.33±0.23b
X. aethiopica	0.5	41.67±10.42a
	1.0	27.00±5.27b
	1.5	16.33±4.05b
Permethrin	0.5	0.00±0.00b
1.0	0.00±0.00d	
1.5	0.00±0.00d	
Untreated Control	0.0	161.67±2.50c

* All values are means of four replicates followed by standard error of the means. Means of the four treatments were compared same test concentration by same test concentration. Means followed by the same letter are not significantly different (P> 0.05) by Fischer's Least Significant Difference (Multiple rang test).
**Number of randomly selected cowpea seeds for oviposition assessment.

Table 3 presents the larvicidal effect of seed powder of three pepper species on *C. maculatus*. Variations were recorded in the number of hatched larvae among pepper treatments and also within test concentrations of a particular species. *P. guineense* produced the highest larvicidal effect among the pepper species, but did not show any clear pattern with advancing concentration. More larvae hatched in cowpea seeds treated with seed powder of *X. aethiopica* than in seeds protected with *A. melegueta*_seed powder. No larvae hatched in seeds treated with permethrin insecticide. Unprotected cowpea seeds had about three hatched larvae and this matched (P > 0.05) the number that hatched in seeds protected with *X. aethiopica* at a concentration of 0.5 g /20 g seeds (2.5 % w/w).

Table 3: Larvicidal Effect of Seed Powder of Three Pepper Species on *Callosobruchus maculatus* Fabricius

Treatment	Rate (g /20 g seeds)	Mean number of Larvae* (in 10 sampled seeds)
A. melegueta	0.5	2.09±0.30a
	1.0	1.80±0.27a
	1.5	0.90±0.22a
P. guineense	0.5	0.90±0.25b
	1.0	1.29±0.29a
	1.5	1.00±0.23a
X. aethiopica	0.5	2.57±0.44a
	1.0	2.24±0.31b
	1.5	1.76±0.29a
Permethrin	0.5	0.00±0.00c
1.0	0.00±0.00c	
1.5	0.00±0.00c	
Untreated Control	0.0	2.92±2.24a

*All values are means of four replicates followed by standard error of the means. Means of the four treatments were compared same test concentration by same test concentration. Means followed by the same letter are not significantly different (P> 0.05) by Fischer's Least Significant Difference (Multiple rang test).

Table 4 presents the result of adult emergence from cowpea seeds as influenced by different treatments and concentrations. Even though, the least number of bruchids emerged from the conventional synthetic insecticide, permethrin, *P. guineense* was best in suppressing *C. maculatus* adult emergence among the test pepper species. Seed powder of *X. aethiopica* reduced adult emergence more than *A. melegueta*. At the rate of 0.5 g /20 g seeds, *A. melegueta* did not significantly differ (P > 0.05) from number of emerged bruchids in unprotected cowpea seeds. The number of emerged bruchids reduced with higher proportion of the treatments. Permethrin was about 35 % better than *P. guineense* in suppressing *C. maculatus* adult emergence.

Table 4: Adult Emergence of *Callosobruchus maculatus* Fabricius from Cowpea Seeds Treated with Seed Powder of Three Pepper Species and Synthetic Insecticide

Treatment	Rate (g /20 g seeds)	Mean number of Emerged Adults*
A. melegueta	0.5	63.00±0.22a
	1.0	54.00±0.53be
	1.5	50.00±0.60a
P. guineense	0.5	27.00±0.10b
	1.0	20.00±0.00c
1.5	19.00±0.72b	
X. aethiopica	0.5	48.00±3.20c
	1.0	43.00±7.61e
	1.5	40.00±6.73a
Permethrin	0.5	8.00±0.95d
1.0	6.00±1.23d	
1.5	4.00±0.78c	
Untreated Control	0.0	65.00±0.12ab

*All values are means of four replicates followed by standard error of the means. Means of the four treatments were compared same test concentration by same test concentration. Means followed by the same letter are not significantly different (P> 0.05) by Fischer's Least Significant Difference (Multiple rang test).

IV. DISCUSSION

The result of this study indicates that seed powder of *Piper guineense* showed the highest insecticidal activity against *Callosobruchus maculatus.* After 24 hours of exposure, its toxic effect generally became noticeable and considerable from 72 hours of exposure (at 1.5 g/ 20 g cowpea seeds) when compared to permethrin. *P. guineense* larvicidal effect and ability to suppress bruchid adult emergence were considerable when compared to the conventional synthetic insecticide, permethrin. This agrees with the conclusion that members of the family Piperaceae show some form of insecticidal activity (Adedire and Ajayi, 1996; Adedire and Lajide, 1999; Asawalam *et al*., 2006). Okonkwo and Okoye (1996) reported that the bioactive chemicals in *P. guineense* are piperine and chavicine and these are highly insecticidal to various crop pests. Lale (1995) however considers piperidine and alkaloids as the major active ingredients in *P. guineense* seeds. However, the plant's seed powder failed to match oviposition deterrence exhibited by permethrin.

That *Afromomum melegueta* did not show contact toxicity against *C. maculatus* at short storage duration corroborates the findings of Adedire and Lajide (1999). However, seed extracts of the plant in particular have been shown to be toxic to bruchids and some other stored product pests (Escoubas *et al*., 1995; Lale, 2002; Ntonifor *et al.*, 2010). Adedire and Lajide (1999) and Lale (2002) reported paradol, an alkyl phenol as the major insecticidal constituent of *A. melegueta*. The plant significantly deterred oviposition when compared to the unprotected cowpea seeds but not when compared to permethrin. Ofuya (1990) had reported that both seed powder and extract of *A. melegueta* did not significantly affect oviposition and egg hatchability of *C. maculatus.*

Xylopia aethiopica did not show any considerable oviposition deterrency effect when compared with unprotected cowpea seeds. However, it significantly reduced adult fecundity and increased adult mortality of *Sitophilus zeamais* on partially resistant stored maize (Babarinde *et al*., 2008).

Contact and fumigant activities might be responsible for the obtained results as reported by (Asawalam *et al* 2007; Tchoumbougnang *et al*., 2009; Ukeh *et al*., 2010; Ntonifor and Mona, 2011). It is interesting that *P. guineense* that most effectively protected against bruchid infestation is edible and as such poses no risk to the health of man and the environment.

More importantly, the results of this study strongly suggest an increased need to research rapidly for plant derived insecticides that will control the notorious *C. maculatus* in stored cowpea.

REFERENCES

[1] Adedire, C.O. and Ajayi, T.S. (1996). Assessment of the insecticidal properties of some plants extracts as grain protectants against the maize weevil S. zeamais Motschulsky. Nigerian Journal of Entomology 13: 93-101.

[2] Adedire, C.O. and Lajide, L. (1999). Toxicity and oviposition deterrency of some plants extracts on cowpea storage bruchid, Callosobruchus maculatus Fabricius Journal of Plant Diseases and Protection 106: 647-653.

[3] Alghali, A.M. (1991). Studies on cowpea farming practices in Nigeria, with the emphasis on insect pest control. Tropical Pest Management 37: 71-74.

[4] Asawalam, E.F. and Emosairue, S.O. (2006). Comparative efficacy of Piper guineense (Schum and Thorn) and pirimiphos methyl as poisons against Sitophilus zeamais (Motsch.). Electron Journal of Environmental and Agricultural Food Chemistry 5: 1536-1545.

[5] Babarinde, S.A., Adebayo, A.A. and Oduyemi, K. (2008). Integrating varietal resistance with Xylopia aethiopica (Dunal) seed extract for the management of Sitophilus zeamais Motschulsky in stored maize. African Journal of Biotechnology 7: 1187-1191.

[6] Caswell, G.H. and Akibu, S. (1980). The use of pirimipho-methyl to control bruchids attacking selected varieties of stored cowpea. Tropical Grain Legume Bulletin 17/18: 9-11.

[7] Lale, N. E.S. (1995). Laboratory study of comparative toxicity of products from three spices of maize weevil. Postharvest Biology and Technology 2: 51-64.

[8] Lale, N.E.S. (2002). Stored Product Entomology and Acarology in Tropical Africa. 1st Edn. Mole Publication, Maiduguri, p. 204.

[9] Ntonifor, N.N., Brown, R.H. and Muller-Harvey, I. (2010). Extracts of tropical African spices are active against Plutella xylostella. Journal of Food and Agricultural Environment 8: 16-26.

[10] Ofuya, T.I. (1990). Oviposition deterrence and ovicidal properties of some plant powders against Callosobruchus maculatus in stored cowpea (Vigna unguiculata) seed. during storage. Agricultural Science Cambridge 115: 343-345.

[11] Okonkwo, E.O. and Okoye, W.I. (1996). The efficacy of four seed powders and essential oils as protectant of cowpea and maize grains against infestation Callosobruchus maculatus (Fabricius) (Coleoptera: Curculionidae) in Nigeria. International Journal of Pest Management 42: 143-146.

[12] Quin, F.M. (1997). Introduction. In: Singh, B.B., D.R. Mohan Raj, K.E. Dashiell, K.E., L.E.N. Jackai (eds.): Advances in Cowpea Research. Copublication of International Institute of Tropical Agriculture (IITA) and Japan International Research Center for Agricultural Sciences, pp. ix-xv.

[13] Rees, D. (2004). Insects of stored products. CSIRO Publishing, Canberra, Australia.

[14] Sarfraz, M. and Keddie, B.A. (2005). Conserving efficacy of insecticides against Plutella xylestella (L.) (Lepidoptera: Plutellidae). Journal of Applied Entomology 129: 149-157.

[15] Somta, C, Somta, P., Tomooka, N., Ooi, P.A.C., Vaughan D.A. and Srinivies, P. (2008). Characterization of new sources of mungbean (Vigna radiata (L.) Wilczek) resistance to bruchids, Callosobruchus spp.(Coleoptera: Brunchidae). Journal of Stored Products Research 44: 316-321.

[16] Tchoumbougnang, F.J., Dogmo, P.M., Sameze, M.L., Ndifor, F., Wouatsa, N.A.V., Zoll, P.M.A. and Menut, C. (2009). Comparative essential oils composition and insecticidal effect of different tissues of Piper capense (L.), Piper guineense Schum Thorn, Piper nigrum L. and Piper umbellatum L. grown in Cameroon. African Journal of Biotechnology 8: 424-431.

[17] Ukeh, D.A., Birkett, M.A., Bruce, T.J., Allan, E.J., Pickett, J.A. and Luntz, A.J. (2010). Behavioural responses of the maize weevil Sitophilus zeamais to host (stored grain) and non-host plant volatiles. Pest Management Science 66: 44-50.

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