

EFFICIENCY OF GINGER, PEPPER FRUIT AND ALLIGATOR PEPPER POWDERS IN THE CONTROL OF COWPEA BRUCHID

Supported by

(Callosobruchus maculatus) (FABRICIUS, 1775)

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Abstract:

Cowpea seeds are legumes particularly for consumptions by populations in tropical and sub-Saharan Africa. Effectiveness of powders of ginger (*Zingiber officinale*), alligator pepper (*Aframomum melegueta*), and pepper fruit (*Dennettia tripetala*) in single and mixed forms were evaluated for the control of cowpea weevils (*Callosobruchus maculatus*) under laboratory conditions. Toxicity bioassay adopted contact method at concentrations of 0.00, 0.05, 0.10, and 0.20% corresponding to 0.00, 1.25, 2.5 and 5.0 grams/25 grams of cowpea seeds in tripled replicates. Ten adult weevils were assigned to plant powders in petri dishes and adult mortality recorded after 30 days treatment exposures and seed viability was assessed. The results demonstrated that increased concentration of plant powders from 0.05-0.20% caused significant mortality of adult weevil (p<0.05). Highest mean mortality was achieved in 0.2% of mixtures of alligator pepper plus pepper fruit and pepper fruit plus ginger (mean= 2.00) with LC₅₀= 3.31 and 3.39 g/25g cowpea seeds respectively. The number of seeds with damages, mean numbers of eggs deposited and weight loss across concentrations was 0.00 to 9.17 gram percent, 2.51, and 1.78%, respectively. Powders of *D. tripetala* in 0.1 and 0.2% concentrations also caused notable mortalities. The mixed forms recorded no eggs, seed damages and weight loss (mean= 0.00). Seed viability showed 50-100% germination and differences were not significant (p>0.05). Overall, this present study indicated that higher concentrations of plant powders in mixed forms could significantly reduce infestation of cowpea seeds.

Keywords: Alligator pepper, biopesticides, *Callosobruchus maculatus*, ginger and pepper fruit

Introduction

Cowpea (Vigna unguiculata L.) is one of the vital food crops that accounts for a considerable measures of protein in human diet. Like other plant proteins, cowpea grains can provide a cheaper protein alternative compared to animal proteins in Nigeria. According to Somta et al. (2008), cowpea seed contains high level of protein which makes them a major alternative source of natural dietary supplement to staple crops including cereals, roots, tubers and fruits. Cowpea is the largest and most popularly consumed protein product in the sub-Saharan Africa. Several losses from post-harvest and reduction in seed quality caused by associated pests are the major problems facing the biosecurity of grains on a general note. Most especially security of cowpea seeds in developing countries such as Nigeria (Adedire and Ajayi, 1996).

The cowpea weevil, Callosobruchus maculatus (Coleoptera: Chrysomelidae) is a storage pest of unprotected grains and their infestations has been shown to reduce the production of cowpea in Africa (Radha and Susheela, 2014). Cowpea infestations do not only occur in field conditions but prevail also in stored vessels where pest activities increase in high rates with optimal conditions. Farmers in Nigeria have long relied on the use of different classes of pesticides. hermetically sealed bags and other viable control means in order to secure grains, prevent losses from post-harvest, and tangibly manage infestations by C. maculatus on field and stored cowpea (Sanon et al., 2010; Baoua et al., 2012; Murdock et al., 2012). The use of pesticide control cause injury to non-target species, toxicity to the environment, accumulations in stored food, health-related conditions in consumers and insecticide resistance in pests. Thus, these implications have led to the search for newer, safer alternative and acceptable methods for the control of stored product pests. These methods include the promotion of the use of plant products which poses little or no side effects to grain consumers (Ileke et al., 2014). Studies on insect pest control using plant derivatives have diverted the focus on the use of chemical pesticide.

Today, the application of many botanical oils and plant-based insecticides are known to have a range of useful biological protection against insect pests (Shaaya *et al.*, 1991; Isman,

2000; Kim et al., 2003; Lee et al., 2003; Aslan et al., 2005; Cetin and Yanikoglu, 2006; Negahban et al., 2007; Maribet et al., 2008, Ayvaz et al., 2009; Vetrivel et al., 2009; Al Qahtani et al., 2010; Napoleao et al., 2013). Botanicals are known to possess secondary metabolites which act as anti-feedants, oviposition deterrents, larvicidal and insect growth regulators (Pugazhvendan et al., 2009). Information on the combined effects of powders of Alligator pepper; Aframomum melegueta, Pepper fruit; Dennettia tripetala, and Ginger; Zingiber officinale against stored cowpea weevils is lacking. Hence, the need for this research. This study is designed to determine the effectiveness of powders of Aframomum melegueta, Dennettia tripetala, and Zingiber officinale in single and mixed forms against Callosobruchus maculatus infesting cowpea grains.

Materials and Methods

Insect culture

Cowpea seeds (obtained from Abraka Major Market, Abraka, Delta State, Nigeria) were used to rear *C. maculatus*. The adult weevils from an infested stock of untreated cowpea seed was sexually identified and used to grow more weevils in 800 ml glass jars. Glass jar was covered with fine gauze of 0.5 mm mesh size to avoid escape of emerging weevils.

The present study was carried out in May and June, 2019. Stock culture of weevils was done in an Entomological chamber at the Department of Animal and Environmental Biology, Delta State University, Abraka, Nigeria, under ambient temperature (23.05±5.75°C) and relative humidity (77.05±2.75%) measured using Voltcraft temperature and humidity data logger (model DL-121TH, Voltcraft, Hirsham, Germany). One to two weeks age adults were used for the toxicity experiment. Sex determination was carried out using examination of the elytra pattern as described by Tiroesele *et al.* (2015); females are maculated with four elytra spots whereas males are plain with less distinct spots.

Cowpea seeds

Five hundred (500) grammes of cowpea seeds for the experiment were also purchased. Meanwhile, the seeds were physically examined for holes, frass, eggs or any suspicious material indicating the presence of infestations prior to

commencement of the experiment. Physical examination was done by visual observation. Damaged seeds were sorted out by handpicking leaving healthy seeds. Seeds were air dried properly and preserved for the experiment.

Plant collection and preparation

Alligator pepper seed (*Aframonum melegueta*), pepper fruit seed (*Dennettia tripetala*) and ginger rhizome (*Zingiber officinale*) were obtained from Abraka market. The plant materials totaling ca. 100g were left to air dry in an open space in the laboratory at room temperature. The plant materials were then grounded into powder using Philips electric blender (Cucina HR 1731/37, 2L/400w.220v-50/60Hz.), sieved with 0.5 mm sieve and preserved in plastic containers with air tight lid until needed for use. This method was adopted from Ojianwuna and Umoru (2010).

Toxicity bioassay

Powders of alligator pepper, pepper fruit and ginger were measured in 1.25, 2.5 and 5.0 grams corresponding to 0.05, 0.1, and 0.2 g/g in petri dishes. Twenty-five grams (25 g) of cowpea seeds were measured in petri dishes containing the various concentrations of plant powders in tripled replicates. Powders were in the proportion of 100% in single form and 50:50 in mixed form for the various concentrations. There were a total of 28-32 seeds in 25 g of cowpeas (mean number \pm SE of seeds, 30 ± 2). Aspirator was used to introduce 10 adult crawling *C. maculatus* into each petri dish containing treated seeds. The petri dishes were subsequently covered to avoid weevils from crawling out during introduction. Petri dishes were then tightly sealed with a paper cello tape to prevent entry and exit of weevils.

The experiment was closely monitored and adult weevil mortality, numbers of egg deposited on seeds, seeds with damages, and weight loss recorded. The minimum concentration of plant protectant was adopted from Adesina *et al.* (2015) with modifications as a dose that was effective against *C. maculatus* adults to determine efficacy immediately after 30 days post treatment. The adults were exposed using protocols mentioned above. After 30 days of treatment exposure, cowpea seeds in each treatment were examined under a light microscope to record the number of eggs deposited on cowpea seed and the number of peas with damages. Cowpea weevils were considered dead when they fail to respond to gentle pressure using the forcept and fingertip.

Seed viability

The effect of crude extracts of alligator pepper, pepper fruit, and ginger on seed viability was assessed by germination test using method adopted from Udo (2008). A pair of seed from the triplicated concentrations was randomly selected for seed viability test. Seeds were soaked in water for 2 minutes and placed on moist cotton wool in a Petri dish. Germination was observed from the first day and up to the five days.

Statistical analysis

Data of adult mortality of *C. maculatus*, number of eggs laid, percentage weight loss in seed and number of seeds with damages were presented in means. The data were equally

subjected to one-way ANOVA to determine differences among the different concentrations. Treatment means were separated using Turkey's test at 5% probability level. Corrected mortality data (Abbott, 1925) of various concentrations were subjected to probit analysis using the XLSTAT, 2019 statistical software for determining the concentration responsible for 50% (LC₅₀) and 95% (LC₉₅) mortality of *C. maculatus*. Seed viability through germination test was presented in percentages. Results were considered significant at p<0.05.

Results and Discussion

Mortality records, egg deposition, seed damage, and weight loss on treated seeds

This study demonstrated the efficiency of three plant powders in single and mixed forms against adult mortality, oviposition, seed damages and weight loss of cowpea infested with C. maculatus (Table 1). Adult mortality was recorded in the various concentrations of plant powders but highest mean mortality was recorded in 0.2% of A. melegueta plus D. tripetala, and D. tripetala plus Z. officinale, respectively. The mean mortality of adult weevil exposed to treated seeds ranged from 0.70 to 2.00 (Table 1). Similar finding of adult mortality was reported by Uwamose and Okolugbo (2016) where higher concentrations of methanolic extract of C. citratus caused 100% mortality in Callosobruchus maculatus. The number of eggs deposited on untreated cowpeas as well as seed damages was a mean ± SE of 2.71±0.14 eggs and 12.66±0.02 damages after 30 days of treatment exposure, respectively (Table 1).

Egg deposited on cowpeas treated with individual and mixed concentrations of plant extract and the damages on seeds decreased with increasing concentration. Differences in the number of eggs deposited (P< 0.05) and the mean damages on seed in grammes (P< 0.05) were significant among the different concentrations. Records of seed with damages, numbers of eggs and weight loss after treatment exposures to powders compared favourably reports in other studies with similar conditions (Ukeh *et al.*, 2009; Ukeh *et al.*, 2010; Okpako *et al.*, 2013; Adesina *et al.*, 2015).

The mean weight loss in on treated cowpeas relative to that on untreated cowpeas ranged from 0.00 to 2.15%. The mean weight loss differs significantly among concentrations of plant extracts (P< 0.05). Mean seed damages and mean number of eggs ranged from 0.00 to 9.17 and 0.00 to 2.51, respectively. The mean \pm SE weight loss of untreated cowpeas was 9.10 \pm 0.02 loss. The number of deposited eggs, damages on seed as well as weight loss was lowest in 0.2% of pepper fruit plus ginger and alligator pepper plus pepper fruit (Table 1). Seeds treated with higher concentrations of plant extracts in Boakye *et al.* (2016) recorded minimum weight loss. This is in line with cowpea seeds exposed to higher concentrations of *A. melegueta* plus *D. tripetala* and *D. tripetala* plus *Z. officinale* which recorded no weight loss.

Table 1: Effect of various concentrations of individual and mixed plant powders on mean numbers of eggs, seed damages and weight loss of cowpea infested with *C. maculatus*

Plant powders	Conc. (%)	Mean adult mortality	Mean No. of Eggs	Mean Seed damages (% gram)	Mean weight loss (%)
Control	0.00	0.00±0.37a	2.71±0.14 ^a	12.66±0.02 ^a	9.10±0.02 ^a
A. melegueta 0.05		0.80 ± 0.37^{ab}	2.51 ± 0.20^{a}	8.14 ± 0.02^{c}	1.75 ± 0.02^{c}
	0.10	1.00 ± 0.37^{ab}	2.13±0.21ab	7.04 ± 0.02^{d}	0.46 ± 0.02^{h}
	0.20	1.10 ± 0.37^{ab}	2.10 ± 0.24^{ab}	5.60 ± 0.02^{h}	0.53 ± 0.02^{gh}
Z. officinale	0.05	0.70 ± 0.37^{ab}	2.20 ± 0.17^{ab}	6.81 ± 0.02^{e}	2.15 ± 0.02^{b}
	0.10	0.90 ± 0.37^{ab}	2.33 ± 0.17^{ab}	6.05 ± 0.02^{g}	0.96 ± 0.02^{e}
	0.20	1.10 ± 0.37^{ab}	1.31 ± 0.24^{ab}	5.13 ± 0.02^{i}	0.16 ± 0.02^{jk}
D. tripetala	0.05	1.20 ± 0.37^{ab}	2.40 ± 0.20^{ab}	9.17 ± 0.02^{b}	1.78 ± 0.02^{c}
	0.10	1.90 ± 0.37^{b}	1.48 ± 0.17^{ab}	6.77 ± 0.02^{e}	0.57 ± 0.02^{g}
	0.20	1.90 ± 0.37^{b}	0.69 ± 0.15^{ab}	$6.22\pm0.02^{\rm f}$	0.20 ± 0.02^{ij}
A. melegueta plus	0.05	1.40 ± 0.37^{ab}	2.17 ± 0.38^{ab}	1.81 ± 0.02^{k}	0.62 ± 0.02^{fg}
D. tripetala	0.10	1.80 ± 0.37^{ab}	0.39 ± 0.27^{ab}	0.22 ± 0.02^{0}	0.29 ± 0.02^{i}
·	0.20	2.00 ± 0.37^{b}	0.00 ± 0.26^{b}	0.00 ± 0.02^{q}	0.00 ± 0.02^{1}
A. melegueta plus	0.05	0.90 ± 0.37^{ab}	2.06 ± 0.31^{ab}	2.15 ± 0.01^{j}	1.14 ± 0.01^{d}
Z. officinale	0.10	1.10 ± 0.37^{ab}	1.94 ± 0.33^{ab}	1.44 ± 0.01^{1}	0.58 ± 0.01^{g}
	0.20	1.20 ± 0.37^{ab}	2.06 ± 0.33^{ab}	0.41 ± 0.004^{n}	0.11 ± 0.004^{k}
D. tripetala plus	0.05	1.40 ± 0.37^{ab}	1.63 ± 0.47^{ab}	1.17 ± 0.02^{m}	$0.71\pm0.02^{\rm f}$
Z. officinale	0.10	1.70 ± 0.37^{ab}	0.00 ± 1.32^{b}	0.11 ± 0.02^{p}	0.17 ± 0.02^{jk}
	0.20	2.00 ± 0.37^{b}	0.00 ± 1.32^{b}	0.00 ± 0.02^{q}	0.00 ± 0.02^{1}

Means of the same superscript letter do not differ significantly (p<0.05) using Turkey test within a column

Table 2: Probit regression estimates (mean \pm SE) for *C. maculatus* exposed to different plant powders

Tuble 2: 11 objet regression estimates (mean ± 52) for c. macanatus exposed to different plant powders								
Plant powders	Conc. (%)	Intercept	Slope	% Probit kill	LC50	LC95	χ2	P-value
A. melegueta	0.05	0.00 ± 0.00	0.00 ± 0.00	8.02	12.02	4394.22		
	0.10	0.25 ± 0.40	0.12 ± 0.19	10.00			0.40	0.529
	0.20	0.37 ± 0.40	0.18 ± 0.19	10.99			0.88	0.347
D. tripetala	0.05	0.00 ± 0.00	0.00 ± 0.00	12.02	3.80	15.14		
	0.10	1.30 ± 0.53	0.61 ± 0.25	18.81			6.13	0.013*
	0.20	1.30 ± 0.53	0.61 ± 0.25	18.81			6.13	0.013*
Z. officinale	0.05	0.00 ± 0.00	0.00 ± 0.00	7.03	12.59	1198.21		
	0.10	0.26 ± 0.40	0.12 ± 0.19	9.01			0.41	0.52
	0.20	0.51 ± 0.40	0.24 ± 0.19	10.99			1.59	0.21
A. melegueta plus	0.05	0.00 ± 0.00	0.00 ± 0.00	13.85	3.31	12.30		
D. tripetala	0.10	0.72 ± 0.48	0.34 ± 0.23	17.77			2.27	0.13
	0.20	1.79 ± 0.75	0.85 ± 0.35	19.78			5.77	0.02*
D. tripetala plus	0.05	0.00 ± 0.00	0.00 ± 0.00	13.84	3.39	12.59		
Z. officinale	0.10	0.50 ± 0.45	0.23 ± 0.21	16.78			1.19	0.27
	0.20	1.79 ± 0.74	0.85 ± 0.35	19.77			5.90	0.02*
A. melegueta plus	0.05	0.00 ± 0.00	0.00 ± 0.00	9.03	8.32	3032.44		
Z. officinale	0.10	0.26 ± 0.41	0.12 ± 0.19	11.01			0.39	0.53
	0.20	0.38 ± 0.41	0.18 ± 0.19	11.99			0.88	0.35

Toxicity bioassay of various concentrations

Probit regression estimates of *C. maculatus* exposed to plant powders at different concentrations is shown in Table 2. Concentrations of *D. tripetala*, *A. melegueta* plus *D. tripetala*, *D. tripetala* plus *Z. officinale* significantly caused adult mortality in 50% lethal dosage (P< 0.05). The powders are arranged in the order of effectiveness as follows: *A. melegueta* plus *D. tripetala* > *D. tripetala* plus *Z. officinale* > *D. tripetala* > *A. melegueta* plus *D. tripetala* was 3.31 and 12.30 respectively. The percentage probit kill was best recorded with 0.1 and 0.2% of *D. tripetala*, 0.05 and 0.2% of *A. melegueta* plus *D. tripetala*, and 0.1 and 0.2% of *D. tripetala* plus *D. tripetala*. Natural mortality was greater than 10% in *A. melegueta* plus *D. tripetala* and *D. tripetala* plus *Z. officinale* respectively

compared to other treatments (Fig. 1). Adult mortality started 24 hours after treatment with various concentrations.

Seed viability

The effect of different concentrations of plant powders on cowpea seed viability show a range of 50 to 100% for individual concentrations and 60 to 100% for mixed concentrations (Table 3). The differences in the germination test of treated seeds in single forms (P= 0.75) and mixed forms (P= 0.65) were not significant. Seed viability test corroborate those reported by Boakye *et al.* (2016) which showed seed viability in all the treatment exposures. They equally assert that treated seeds with highest concentration of liquid extract was not viable compared to lower concentrations. Treated seeds in this study were viable in germination test.

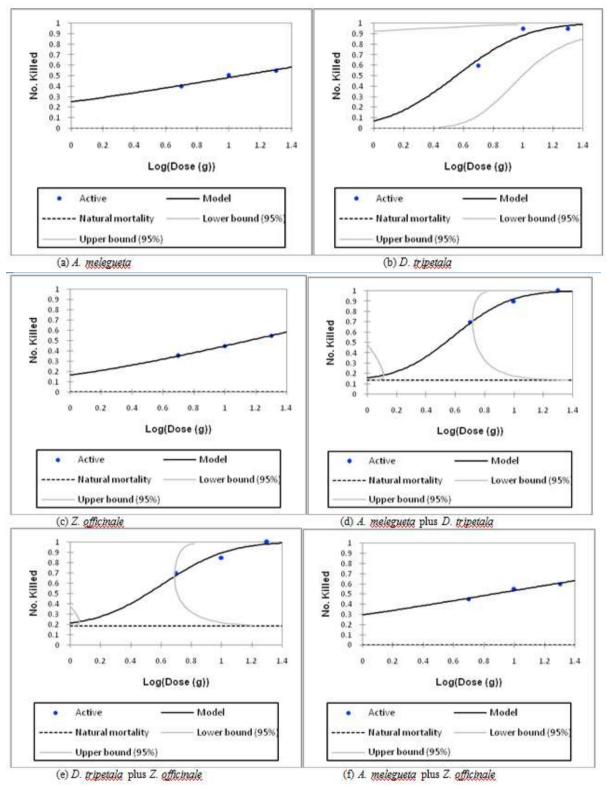


Fig. 1: Effect of plant powders in individual and mixed forms on mortality of C. maculatus

Table 3: Cowpea seed viability exposed to different treatment concentrations

	% seed viability							
Dose (%)	A. melegueta	D. tripetala	Z. officinale	A. melegueta plus Z. officinale	D. tripetala plus Z. officinale	A. melegueta plus D. tripetala		
Control	100a	100a	100a	100a	100 ^a	100 ^a		
0.05	80^{a}	100^{a}	60^{a}	80^{a}	100 ^a	60 ^a		
0.1	70^{a}	100^{a}	60^{a}	70^{a}	100 ^a	80^{a}		
0.2	50 ^a	50 ^a	100^{a}	60^{a}	60^{a}	100^{a}		

Means of the same superscript letter do not differ significantly (p<0.05) using Turkey test within a column

Grain losses are common occurrences in Nigeria, and to avoid storage losses and grain poisoning due to pesticide use, various plant products with insecticidal potentials have been used as storage protectants over time in Tropical Africa (Ito and Ukpohwo, 2018). The use of plant materials is due to their availability, edibility and low records of human toxicity (Oni, 2014). The mortality of adult weevil may be linked to active compounds in powders which caused spiracle blockage, amongst other physiological stress on the insect (Adedire et al., 2011; Fernando and Karunaratne, 2012; Sarwar et al., 2012). Furthermore, it is probable that plant powder disrupted the developmental cycle of insects causing water loss through cuticle stiffening, which led to reduced productivity (Sousa et al., 2005). The high mean mortality of adults recorded in higher treatment concentrations suggests that insect activities were suppressed. Damages on seed, weight loss, and number of deposited eggs were recorded low compared to control across treatment concentrations. Apart from using plant protectants, seed coating was earlier related to grain protection deterring insect attack and infestations (Abulude et al., 2007; Osipitan and Odebiyi, 2007).

Conclusions

The plant powders in this study have demonstrated optimal potentials for insecticidal activity against *C. maculatus*. The findings revealed that best concentrations of pepper fruit (0.1 and 0.2%) and the mixtures of 0.2% of alligator pepper plus pepper fruit and pepper fruit plus ginger led to highest records of adult weevil mortality. The number of deposited eggs, percentage weight loss, and damages on treated seeds were reduced compared to when seeds were left untreated. Thus, these plant powders could be adopted as protectants for stored cowpea seeds. In Nigeria, these plant species are available, affordable and edible culinary spices and herbs, thus posing no detrimental effects on the environment and life to the user. More studies are needed on effectiveness of plant protectants against adult cowpea weevils.

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Conflict of Interest

Authors have declared that there is no conflict of interest reported in this work.

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