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RESEARCH ARTICLE

ENTOMOCIDAL CAPABILITY OF SOME SPICE POWDER ON THE CONTROL OF COWPEA BEETLE (*CALLOSOBRUCHUS MACULATUS*) INFESTATION ON STORED COWPEA SEEDS (*VIGNA UNGUICULATA*)

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ABSTRACT

Callosobruchus maculatus, is an undisputedly major insect pest militating against legume food grains availability and security. This experiment was conceived to evaluate the toxicity effect of some spice powder (*Capsicum annum* and *Piper guineense*) in the control of cowpea beetle (*C. maculatus*) on stored cowpea seeds arranged in a completely randomized design (CRD) with four treatments: 5g of dried *Capsicum annum* fruit, 5g of *Piper guineense* leaves, 5g of *Piper guineense* seeds and the control (no treatment) and replicated five times. The spice powders were assessed using the following parameters: percentage mortality of adult weevils at 24, 48, and 72 hours, progeny emergence after 35 days of storage, percentage weight loss, percentage of damaged grains and percentage germinability of treated and untreated cowpea seeds. The results obtained showed that at 72 hours, *C. annum* fruit had the highest mortality (50%), followed by *P. guineense* seed powder (45%) and was significantly more potent than *P. guineense* leaves (25%) and the untreated control (2%), respectively. The percentage of adult emergence in untreated cowpea seeds was 40.9%. While *C. annum* fruit powders and *P. guineense* seed powders completely inhibited adult emergence. *C. annum* fruit powder provides significant highest protection of the treated grains, followed by *P. guineense* seeds compared to the untreated seeds. The plant powders did not adversely affect seed germinability, suggesting that seeds treated with *P. guineense* seeds and *C. annum* can be used as an alternative to chemical pesticides against *C. maculatus* infestation and suppression of seed damage by smallholder farmers. Further study, recommended to evaluate the powders shelf life to determine if additional application is required after a specific amount of time to maintain the powder efficacy.

KEYWORDS

Capsicum annum, efficacy, infestation, *Piper guineense*, seed germinability, shelf life

1. INTRODUCTION

In many underdeveloped countries, including Nigeria, *Callosobruchus maculatus* Fabricius (Coleoptera: Chrysomelidae) infestations wreak havoc on cowpea storage, accounting for over 90% of pest damage. Cowpea seed storage at smallholder levels is limited by beetle infestations, which are often a very harmful and destructive pest of pulse seeds (Hamzei et al., 2023; Sanusi and Ibrahim, 2024). Through emerging adult escape holes, the insects inflict significant quantitative and qualitative losses, including damage to seeds that reduces their nutritional content, weight, market value, and seed germination potential (Oluwafemi, 2012; Hamzavi et al., 2022).

The use of synthetic chemical insecticides as dusts or fumigants has been the most effective control method for minimising losses and damage caused by *C. maculatus* to stored cowpea seeds (Younis et al., 2022). Despite their effectiveness, the use of synthetic insecticides and fumigants is hampered by a number of issues, including the potential for human and livestock toxicity, environmental pollution, the development of resistant insects, and more (Campos et al., 2013; Soujanya et al., 2016; Kalpna et al., 2022). As a result, researchers are looking for alternative methods for managing stored produce insects in order to overcome these issues. A group reseachers stated that utilising botanical insecticides can be an alternative method of controlling *C. maculatus* (Rohimatun et al., 2023). Plant products such as powders, extracts, and essential oils contain a variety of phytochemicals which affect a range from adult insect mortality, oviposition inhibition, developmental toxins, hatching inhibition, ovicides, larvicidal, and emergence inhibitors (Yang et al., 2004; Bakkali et al., 2008; Baz et al., 2024).

In Nigeria, there is are variety of spices that can be used as botanical insecticides such as *Syzygium aromaticum, Allium sativum, Piper nigrum, P. guineense, Capsicum annum,* etc. Limited evidence is available concerning the usefulness of these plant materials in averting insect pest infestations of stored food grains. Therefore, the focus of this study was to assess the potency of dried *Capsicum annum* fruit, *P. guineense* leaves, and seed powder under laboratory settings and validate their usage in the control of *C. maculatus* infestation and inhibition of seed damage.

2. MATERIALS AND METHODS

2.1 Experimental Location and Conditions

This experiment was conducted at the Entomology laboratory of the

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Federal College of Agriculture, Ishiagu, Ebonyi State (latitude 05°56'N and longitude 07°41'E) under ambient laboratory conditions of 28 + 2 °C, 80 + 5% relative humidity and 12 h D: L.

2.2 Experimental Materials

The leaves and fruits of *Piper guineense* and the fruits of *Capsicum annum* were collected from the vegetable farm of the Federal College of Agriculture, Ishiagu, Ebonyi State. The materials were washed in clean water to remove dirt, air dried, and thereafter ground into fine powder using the Silver Crest 2L 8500W 9525 Model electric blender. The powder was sieved through a $1mm^2$ perforation mesh to obtain the finest powder and subsequently, stored in an airtight container separately and kept in a refrigerator till use to prevent volatilisation of their bioactive chemical ingredients (Anaele et al., 2024).

2.3 Culturing of Cowpea Beetle (*Callosobruchus maculatus*)

The adult *C. maculatus* used in the experiment was derived from infested grains gotten from food grains sellers in Eke market, Ishiagu, Ebonyi State, cultured on 400g of drum variety in a 2L plastic container under laboratory conditions of 28 + 2 °C temperature, 80 + 5% relative humidity and 12 h darkness and light. Muslin fabric fastened tightly with a rubber band was used to cover the plastic container to allow for aeration and prevent the escape of the adult weevils. The culture was maintained for 25-30 days to give room for enough oviposition, and newly emerged 0-2-day-old adults were used for the experiment (Anaele et al., 2024; Adesina and Mobolade-Adesina, 2020).

2.4 Infestation of Cowpea Seeds and Application of Treatments

Clean seeds of drum cowpea cultivar with no visible signs of eggs and presence of adults or exit holes used for the study were handpicked and sterilised by keeping the bagged seeds in a deep freezer for one week to kill any hidden infestation. Before use, the seeds were removed, air-dried and left to equilibrate at room temperature for 4 h on the laboratory workbench to avoid mouldiness (Adesina, 2012; Adesina, 2022). About 50g of the sterilised cowpea seeds were introduced into 20 kilner jars, each arranged in a Completely Randomised Design and replicated five (5) times. Five (5) grams of *C. annum* fruit powder, 5g *P. guineense* leaf powder, and 5g *P. guineense* seed powder were admixed with the cowpea seeds, painstakingly shaken to ensure that the powder adequately covered the seed surface. Ten (10) unsexed adult beetles were introduced into each of the Kilner jars, tightly covered with the lids to prevent the insects from escaping and were arranged on a laboratory table for observations and data collection (Adesina, 2022). A control experiment was also set up, containing untreated seeds with spice powder.

2.5 Toxicity Effect of *P. guineense* and *C. annum* Powders Against *C. maculatus* Infestation and Seeds Damaged

Mortality assessment was done at 24, 48, and 72 hours after adult insect exposure to the treatment. At each observation, the dead insects were removed from the jars. Dead adult beetles were assessed as those which showed no visible movement after 60 seconds of observation. To avoid the possibility of death mimicry, the insects were watched for 2 min and again subjected to gentle pressure (Adesina et al., 2015). Before every count, the containers were kept in the freezer for 5 minutes to incapacitate the beetles. At the end of 7 days, all adult weevils were removed from the containers in anticipation of emerging progeny. The percentage of adult insect mortality was calculated as: number of dead beetles per container/total number of beetles introduced x 100.

Thirty-five (30) days post-exposure, the cowpea seeds were properly sieved, and the number of newly emerged adult beetles from both treated and control dishes was counted and recorded (Adesina et al., 2011). At the end of the experiment, the seed with adult emergent exit holes was counted and recorded per treatment to calculate the percentage of damaged seeds: Number of seeds with adult emergent holes (punctured seeds)/total number of seeds x 100 (Ileke and Oni, 2011). The seeds were reweighed, and the final weight of each of the treatments was recorded to calculate percentage loss in weight of the seeds (Babarinde et al., 2010): % Weight loss = W_I - W_f/W_I x 100. Where WI = Initial weight and Wf = final weight.

2.6 Percentage Germinability of Cowpea Seeds Treated with *P. guineense* and *C. annum* Powders

Percentage seed germinability was obtained by placing 10 randomly selected seeds handpicked from the seeds with no visible adult exit holes on cotton wool moistened with water in a Petri dish, replicated three times under ambient laboratory conditions suitable for germination. Forty-eight (48) hours after, the number of germinated seeds out of the total number sown was counted and recorded to evaluate the percentage germinability rate of the seeds using the formula: number of germinated seeds/ total number of seeds sown x 100 (Adesina et al., 2019a, b).

2.7 Data Analysis

The data generated was subjected to analysis of variance (ANOVA). Significant treatment means were separated and compared using Fisher's Least Significant Difference (LSD) at 5% level of probability. Data on percentage mortality, seed damage, weight loss and seed germination were arcsine transformed while data on adult emergence were square root transformed to ensure normality and homogeneity of variance, before analysis (Asiry and Zaitoun, 2020).

3. Results

3.1 Toxicity Effect of Some Spice Powders Against *C. maculatus* Infestation on Stored Cowpea Seeds

All the tested powders significantly (P<0.05) reduced the longevity of adults on treated cowpea grains; the spice powders showed beetle mortality ranging from 0 to 50% (Table 1). Adult mortality increased with the length of exposure. *C. annum* fruit and *P. guineense* seed powders were most effective against bean beetle induced 45% and 25% mortality, respectively; the least was the *P. guineense* leaves, which recorded 10% mortality after 48 hours of exposure. At 72 hours, there were significant (P < 0.05) differences in mortality. Among the treatments, *C. annum* fruit had the highest mortality of 50% followed by *P. guineense* seed powder, which recorded 45% and was significantly more potent than *P. guineense* leaves and the untreated control, which had values of 25% and 2%, respectively.

The different plant powders significantly (P<0.05) reduced the progeny of bean beetle (Table 2) after 35 days of storage. *Capsicum* fruits and *P. guineense* seed prevented the emergence of the *maculatus* population compared to *P. guineense* leaf powder and control. The percentage of adult emergence in the untreated cowpea seeds was 40.9%. While *C. annum* fruit powders and *P. guineense* seed powders completely inhibited the development of *C. maculatus*, making the emergence from seeds the least, with zero (0) adult insects.

Table 1: Effect of Spice Powders on the Percentage Mortality of Adult *Callosobruchus maculatus* in Stored Cowpea Seeds

Treatments	Percentage Mortality		
	24 hours	48 hours	72 hours
C. annum fruit powder	0	45	50
P. guineense leaf powder	0	10	25
P. guineense seed powder	0	25	45
Control	0	0	0
LSD	NS	1.54	1.78

3.2 Effect of Some Botanical Dust on Percentage Weight Loss and Damage Assessment in Stored Cowpea Seeds.

On the percentage weight loss, there was a significant (P<0.05) difference among treatments; the highest weight loss of 39.3% was recorded from cowpea grains in the untreated control, followed by those treated with *P. guineense* leaf powder, which recorded 14.45%. (Table 2). There was no weight loss in the cowpea seeds treated with *C. annum* fruit powder, which was statistically similar to the value of 0.5% obtained from *P. guineense* seed powder. Results obtained also showed that cowpea seeds treated with botanical powders showed a significant difference (P<0.05) in the reduction of damage caused by *maculatus. T*reatments differed in their protection against damage by the beetle (Table 2). Seeds with emergent holes were counted as damaged. The highest percentage of damaged seeds was recorded on untreated control grains. No damage was observed on the stored grains treated with *C. annum* fruit powder. Stored grains protected by *P. guineense* seed powder had recorded a damage of 0.5%.

3.3 Effect of Some Botanicals on Germinability Assessment on Stored Cowpea Seeds.

The germinability test conducted at the end of the storage revealed no significant (P>0.05) difference among treatments (Table 2). The effect of botanical powders on the viability of treated grains showed that none of the plant powders adversely affected the viability of the cowpea grains when compared with the untreated control. Almost all the seeds germinated, and seeds treated with spice powder exhibited a non-significant germination percentage when compared. The untreated control cowpea seeds had the least germination percentage (68.93%), and *C. annum* fruit and *P. guineense* seed powder had values *of* 98.2 and 95.1%respectively.

Table 2: Effect of Spice Powders on the Progeny Emergence, Seed Damage and Germinability of Stored Cowpea Grains

Treatments	% Progeny emergence	% Weight loss	% Damaged grains	% Germinability
C. annum fruit powder	0.00	0.00	0.0	98.2
P. guineense leaf powder	10.5	14.45	9.0	93.0
P. guineense seed powder	0.00	10.56	0.5	95.1
Control	40.9	33.9	23.5	68.93
LSD	9.43	7.54	5.24	5.16

4. Discussion

The safe use of insecticides derived from plant products has been receiving increasing attention as an effective alternative to synthetic insecticides in the management of insect pests commonly associated with infestations of stored products. The outcome of this study indicates that powders of *C. annum* fruits, *P. guineense* leaves, and seeds showed pronounced toxicity against *C. maculatus* infestation and suppression of damaged seed.

The resultant high mortalities of adult bean beetles observed on cowpea grains treated with the spice powders compared to untreated grains could be due to the high toxic effect of the product on adult bean beetles which could be ascribed to their pungent and peppery odour that asphyxiate insects by blocking the spiracles and impair their normal respiratory triggering choking and ultimately leading to the insect death (Ufele et al., 2015). The higher mortality of adult *C. maculatus* in stored cowpea seeds treated with *P. guineense* leaves and seeds powder was recorded with *P. guineense* seeds powder, which is more effective than *P. guineense* leaves. This may be due to their difference in percentage of chemical compounds to abrasive action. The mortality caused by spice powders compared to the controls could also be credited to contact toxicity since the weevils crawling over the grains, the chemical constituents of the powder could lodge between cuticular segments and increase water loss through abrasion of the cuticle. A similar observation was made (Alberto and Ulises, 2025). The significantly high mortality rate designates the credible presence of lethal bioactive ingredients with insecticidal properties in the spice powders against *C. maculatus*.

These chemicals in the test plants may be responsible for the pungent odour that repelled the insect pest. The repellency of insect pests may generally be attributed to the chemosensory effects of plants' secondary metabolites as terpenes, which insects take up through their respiratory system.

The significantly low progeny emergence of *C. maculatus* from cowpea seeds treated with *P. guineense* and *C. annum* could be due to the high toxic effect of these products and the presence of alkaloid, steroid and cardiac galacosides groups on both egg, larval and adult stages (Ekeh et al., 2018). According to this study, stated that the insecticidal properties of any plant products depend on their active constituents (Idoko and Adesina, 2012). Most of researchers reported the presence of piperine, chavicine, alphapinene, limonene, linalool, piperidine and alkaloids in *P. guineense* (Okonkwo and Okoye, 1996; Golob et al., 1999; Lale, 1995). While Madhumathy *et al.* (2007) and Oni (2010) reported the presence of seven capsinoids, including capsaicin and hydrocapsain, in *C. annum*. According to a study, the proteins found in *C. annum* leaves, such as a protease inhibitor, can disrupt the weevil post-hatching development by decreasing oviposition, larval bulk, and altering digestive enzymes and energy stores, resulting in insect death, suppression of adult emergence and seed damage (Bellei et al., 2023). The presence of active constituents in this plant material is a major contributor to their toxic properties and seems to be accountable for its insecticidal properties against *C. maculatus*. The decrease or zero rate of emerged adults noticed from the study could also be as an import of the admixture of cowpea seeds with the spices powder, which fills the insect intergranular air spaces and prevents adult free movement for mating and oviposition (Adesina et al., 2024).

The disruptive effect of *P. guineense* and *C. annum* powders as observed in the present study aligned with many findings exhibited significant adult mortality, inhibited egg laying, hatchability, adult emergence, and suppression of seed damage against different stored products insect pest infestation (Akinwumi et al**.**, 2007; Oni, 2010; Mailafiya et al., 2014; Ekeh et al., 2018; Manju et al., 2019; Paikaray et al., 2021; Rosulu et al., 2022; Wilberforce et al., 2024).

The observed non-emergence of the F1 generation of bean beetle on cowpea and lower seed damage from *P. guineense* seed and *C. annum* seed powders could be a result of high mortality of adult insects, which deterred females from laying eggs and complete suppression of the developmental stages of insects. This resulted in significantly reduced seed damage compared to untreated cowpea. This corroborates the report of who opined that the population of emerged adult *C. maculatus* from treated seeds is directly related to the volume of damaged seeds, with a low emerged adult population translating to reduced metabolic and larval feeding activities within the grain (Adesina et al., 2024). In addition, antifeedant properties of the spice powder that impede the larvae from feeding on the treated grains could also lead to reduced damaged seed and weight loss in the seeds treated, as the adult *C. maculatus* do not feed on the grains (Hamzavi et al., 2022).

The fact that the powders did not affect the germinability of treated seeds is of great importance to the users who may wish to adopt this technology for the preservation of cowpea seeds. This may be due to the non-lethal nature of the botanicals on the seeds and the capability of the spice powder to preserve the treated seeds against *C. maculatus* infestation and seed damage. Similar effects of plant products on grain germinability of treated seeds have been observed in the study conducted (Oparaeke and Dike, 2005; Adesina et al., 2019a, b).

5. Conclusion

According to this study, the spices have some toxic mechanisms that significantly induced the mortality of *C. maculatus* and prevented adult emergence, which has a greater negative impact on the weevils than the controls. As a result, the spices have the greatest potential as protectants for stored grain legumes. Since these spice powders greatly reduce infestation and seed damage, they should be included in resource-poor farmers' grain protection practices. Furthermore, it is necessary to evaluate the powders shelf life to determine whether additional application is required after a specific amount of time.

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