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EFFICACY OF PLANT POWDERS OF *EUGENIA AROMATICA* AND *AZADIRACHTA INDICA* ON THE RICE MOTH, *Corcyra cephalonica* (STAINTON) (LEPIDOPTERA: PYRALIDAE) IN COCOA BEANS IN STORAGE

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ABSTRACT

The rice moth, *Corcyra cephalonica* is a notorious insect pest of stored cocoa beans causing quantitative and qualitative economic damage that can lead to total loss of stored beans if not controlled. The use of synthetic fumigants for control of the moth often leaves undesirable residues that could pose health and environmental hazards. Therefore, safer alternative like the use of plant powders could be used. Hence, plant powders of *Eugenia aromatica* and *Azadirachta indica* were evaluated for their efficacy as protectants of cocoa beans against the rice moth. Disinfested cocoa beans (50g/jar) were admixed with powders of *E. aromatica* and *A. indica* at levels of 0.5 w/w, 1 w/w, 2 w/w and 4 w/w in three replicates with an untreated control under laboratory conditions at ambient temperature of 28 ± 2 °C and relative humidity 72% ± 5%. A cohort of day old adults (n=20) of *C. cephalonica* were introduced into each container for bioassays. Total adult mortality of the F_1 progeny at seven days post-infestation was 100% for both powders at 4 w/w concentration. The LD_{50} of *E. aromatica* and *A. indica* three days after treatment were 1.09 and 11.41 while the LD_{95} were 2.36 and 149.85, respectively. Percentage bean damage ranged from 10% at 0.5w/w to 0% at 2.0 w/w for *E. aromatica* and a similar trend was recorded for bioassay with *A. indica* (26% at 0.5 w/w). Both plant powders averted damage of cocoa beans at 2.0 w/w and therefore, can be incorporated into a formidable control strategy of cocoa beans in storage.

Keywords: *Corcyra cephalonica*, *Eugenia aromatic*, *Azadirachta indica*, bioassay, cocoa beans

INTRODUCTION

Cocoa, *Theobroma cacao* Linnaeus is a major commodity crop cultivated mainly for its highly valuable beans. The cocoa beans constitute the raw materials of some agro based industries which manufacture semi-finished product, such as cocoa mass, used in making chocolate, biscuits and confectioneries. Also, from the cocoa beans are obtained cocao butter, intended for various food industries for sweetening products (Opeke, 1992). The cocoa butter is used for making sweets, chocolate, perfume, and in pharmaceuticals, and finished products that include, chocolate powder, bars of chocolate, cocoa cake and various chocolate-based products (Adegbola, 1990). The beans ere treated through a process of fermentation after they have been removed from the fruit. This process is necessary to moderate the initially bitter flavour of the cocoa beans and to develop their typical flavour. By fermentation, the highly bitter tannins present in the beans are oxidized, resulting in the formation of aromatic substances and the development of the typical brown to deep red-brown colour of the cocoa beans. As a result of the heat generated by fermentation, the cocoa beans lose their ability to germinate (Navarro *et al.,* 2007).

Cocoa in storage is exposed to rodent attack which leads to reduction in its market value. An estimated 40 percent of global annual cocoa production is lost to insects, pests and diseases, and currently, cocoa pests and diseases follow a regional pattern of distribution and are indigenous to specific geographic areas (Boateng, 2010). The main insect pests that attack cocoa beans in storage include the tropical warehouse moth (*Ephestia cautella*), cigarette beetle (*Lasioderma serricone*), corn sap beetle (*Carpophilus dimidiatus*), rusty grain beetle (*Cryptolestes ferrugineus*), coffee bean weevil (*Araeocerus fasciculatus*), red flour beetles (*Tribolium castaneum*) and the rice moth (*Corcyra cephalonica*) (Boateng, 2010). The most common storage pest in cocoa beans from Indonesia and South America is the cocoa moth or the tropical warehouse moth (*Ephestia cautella*), whereas the dominant species in cocoa beans from West Africa is the rice moth (*Corcyra cephalonica*) (Navarro *et al*., 2007).

Corcyra cephalonica (Stainton) is a serious pest of some important stored food commodities such as rice, wheat, maize, sorghum, groundnut, cotton seeds, coffee, spices and cocoa beans, especially in the tropics (Allotey, 1991). The larvae of *C. cephalonica* cause considerable damage to stored food commodities while feeding, leaving silken threads wherever they move. The webbing formed is noticeably dense and tough adding to the damage caused (Allotey and Azalekor, 2000).

Fumigants have been widely used for the control of insect pests in cocoa beans and also in other stored products to prevent economic and quality losses because without fumigation, these pests will reduce commodities to worthless frass. However, increased public concern over the adverse effects of insecticide residues in food and the environment has led to their partial substitution by alternative control methods. Consequently, non-chemical and environmentally user-friendly methods of pest control in the post-harvest sector are becoming increasingly important (Navarro *et al*., 2007). Methyl bromide has been phased out in developed countries since 2005 and in developing countries by 2015 because of its contribution to stratospheric ozone depletion (United Nations Environment Programme, 1998). However, phosphine is very popular in developing countries because of the ease of its application but many insects have developed resistance to phosphine over the last decade (Daglish and Nayak, 2012).

It has become necessary to minimize the amount of toxic materials released into the environment by increasing the search for cheap, easily biodegradable and readily available plant/natural products. The use of locally available plant material to protect stored products against pest damage is common practice in traditional farm storage systems in most developing countries (Boeke *et al*., 2004). It is against this background that the efficacy of clove fruit, *Eugenia aromatica* and neem seed, *Azadirachta indica* powders were tested as protectants of cocoa beans against the rice moth, *C. cephalonica*.

MATERIALS AND METHODS

Cocoa beans were obtained from a large commodity warehouse where the cocoa beans are temporarily stored before exporting to other countries. To safeguard against any unwanted infestation, cocoa beans were disinfested in an oven at a temperature of 55°C for 24hrs. Then, left under laboratory conditions at ambient temperature of 28 ± 2 °C and relative humidity 72% ± 5% for 24hrs for their moisture contents to stabilize before being use for the experiments. Adult *C. cephalonica* were obtained from laboratory cultures at the entomological unit of the Cocoa Research Institute of Nigeria (CRIN), Ibadan Oyo State and maintained at the laboratory in the Department of Zoology, University of Lagos. Mature, fallen neem seeds were gathered from Festac in Lagos while fruits of dried clove were obtained from the local market in Lagos State, Nigeria. The plant materials were sun-dried, decorticated and pulverized into powder using a manually operated mill (Philips Blender HR2056). They were then stored in a freezer at -18°C until ready to use.

Toxicity of powers on adult *Corcyra cephalonica* and F_1 emergents

For each plant material, 0.25, 0.5, 1 and 2 g of powders (*E. aromatic* and *A. indica*) were separately introduced into 50 g of cocoa beans in a 500 ml glass kilner jars corresponding to 0.5, 1, 2, and 4% w/w concentration, respectively. The containers were shaken thoroughly to ensure even mixing and the lid of each jar was covered with a muslin cloth, secured with a rubber-band. Three replicates of each treatment and untreated cocoa beans were set up. A lot of day old (n=20) adult moths were introduced into each container using a fine brush. Adult mortalities were recorded daily for 7 days post-infestation. After counting dead insects on the 7th day, live and dead insects were removed and the cocoa beans were left in the jars for weekly observation for F_1 emergence for a period of six weeks. Preliminary experiments showed that adult emergence usually started after four weeks.

Residual effect of powders on *Corcyra cephalonica* population increase and damage

In a similar study, concentrations of 0.5, 1, 2 and 4% (w/w) of each powder/50 g of cocoa beans were admixed as described above. A batch of day-old adult insects (n=20) was introduced into each jar containing treated or untreated cocoa beans. Control for each set of treatments consisted of cocoa beans without plant material. Each treatment was repeated three times. The containers were left undisturbed for three months after which, the number of live and dead moths were determined for each jar. Damage assessment was done by measuring the weight of the sieved powder and that of the grains without powder (final weight). The amount of cocoa bean powder (frass plus faeces) formed was expressed as the total powder minus the weight of plant powder used.

Percent weight loss was determined as follows:

$$\frac{\text{Initial weight - Final weight}}{\text{Initial weight}} \text{ X 100}$$

The number of damaged (beans with characteristic hole) and undamaged beans in a randomly selected 100 bean sample from each jar were counted and percent bean damage was estimated.

Statistical analysis

The data were analyzed using analysis of variance (ANOVA) procedure of the SPSS version 16.0 software and significant differences (P < 0.05) were separated using LSD at the 5% level.

RESULTS

Powders of plant material applied to cocoa beans at varying dosages showed significant toxicity (P< 0.05) against the F_1 progeny of *C. cephalonica* in storage (Table 1). Emergence of insect from the control was significantly higher than those of both plant powders irrespective of the dosages (Table

1). There was zero percent emergent on cocoa beans treated with *E. aromatica* at 2 and 4 g per 100 g of cocoa beans while *A. indica* recorded a similar feat at only 4 g/100 g cocoa beans.

The two plant powders were generally toxic to adult *C. cephalonica*, however, *A. indica* had a slower toxic action than was *E. aromatic*. Also, there were significant differences (P<0.05) in mortality between plants, among dosages and exposure time (Table 1). There was no insect mortality recorded in the control for both plant treatments during the seven days of toxicity assessment on the adult moth. Absolute control (100% mortality) was achieved with *E. aromatica* powder at 2 g and 4 g per 100 g cocoa beans on the 4th day after treatment (Table 1) while 100% mortality was obtained with *A. indica* at 2 g/100 on the 7th day after treatment (Table 1). At a dosage of 2% (w/w), about 88.5% mortality was attained with *E. aromatica* in just two days whereas it took *A. indica* six days to achieve 96.5% mortality.

Table 1. The F_1 adult emergents (mean ± SE) of *Corcyra cephalonica* on cocoa beans treated with plant powders of *Eugenia aromatic* and *Azadirachta indica*

Plant type/ Powder levels (g/100g cocoa beans)	Number (mean ± SE) of F_1 adult progeny
Eugenia aromatica	
0	594 ± 66.12a
0.5	190.67 ± 28.26b
1.0	6.33 ± 2.96c
2.0	0d
4.0	0d
Azadirachta indica	
0	668.33 ± 141.98a
0.5	256.33 ± 44.70b
1.0	37 ± 9.29c
2.0	2.33 ± 1.45d
4.0	0e

Means followed by the same letter in the same column are not significantly different (P > 0.05). Tukey's Studentized Range (HSD) Test.

Table 2 shows the LD_{50} and LD_{95} values of powders of *E. aromatica* and *A. indica* to *C. cephalonica* at 3, 5 and 7 days after treatment. The LD_{50} and LD_{95} values reduced with a corresponding increase in exposure periods from 1.090 to 0.871 and 2.357 to 1.662, respectively for *E. aromatica* while the LD_{50} and LD_{95} values reduced from 11.414 to 0.842 and 149.853 to 1.403, respectively. In other words, the toxicity factor at LD_{50} showed that *A. indica* became 13.56 times more toxic than *E. aromatica* on the 7th day after treatment while *E. aromatica* was only 1.25 times more toxic than *A. indica* on the first day.

The residual effect of powders of *E. aromatica* and *A. indica* on the moth populations after three months in storage are presented in Figures 1 and 2. Both plant powders showed a significant effect on the reproductive cycle in which F_1 progeny populations were reduced, and this was generally dose-dependent. At higher dosages of 2 and 4 % (w/w), the population of live and dead moths significantly reduced (P>0.05) to the barest minimum.

Table 2. Toxicity of powders of *Eugenia aromatica* and *Azadirachta indica* to *Corcyra cephalonica* at 3, 5 and 7 days after treatment

Days of treatment	*Eugenia aromatica*	*Azadirachta indica*
Day 3		
LD50	1.090 (0.144 – 8.232)	11.414
LD95	2.357 (1.363 – 6.030)	149.853
Day 5		
LD50	0.920 (0.646 – 1.348)	1.158 (0.215 – 3.385)
LD95	1.739 (1.233 – 6.902)	3.137 (1.696 – 8.376)
Day 7		
LD50	0.871 (0.668 – 1.145)	0.842 (0.769 – 0.922)
LD95	1.662 (1.234 – 3.910)	1.403 (1.232 – 1.716)

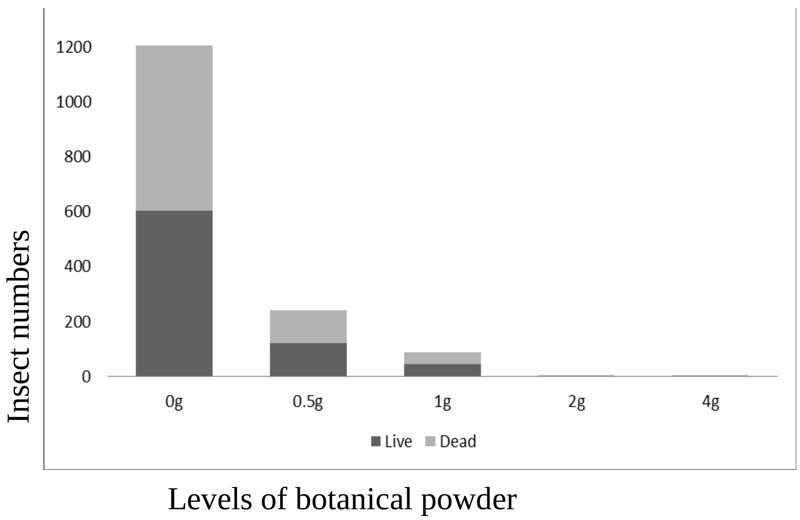

Figure1. Residual effect of powder of *Eugenia aromatica* on the moth population after three months in storage

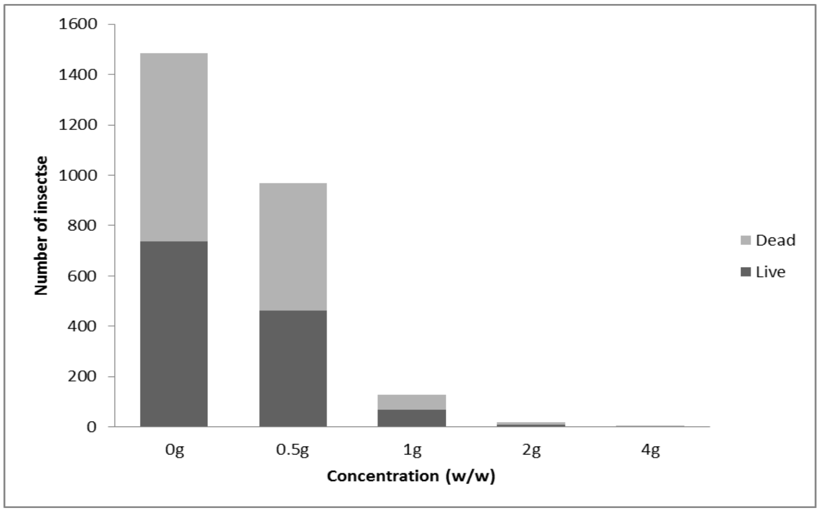

Figure 2. Residual effect of powder of *Azadirachta indica* on the moth population after three months in storage

The powders of *E. aromatica* and *A. indica* significantly reduce cocoa bean damage from attack by C. cephalonica (Table 3). At dosages of 2 and 4 % (w/w), E. aromatic averted the moth attack whereas at 4 % (w/w), A. indica completely prevented moth attack. Cocoa beans samples treated at these rates showed no sign of damage did not produce bean powders and lost no weight in three months of storage. There were significant differences between the control and treated cocoa beans as well as dosages for each damage parameter tested.

Table 3. Damage assessment of *Corcyra cephalonica* on cocoa beans treated with different powder levels of *Eugenia aromatica* and *Azadirachta indica* in storage

Plant type/ levels (g/100g bean)	Powder Damaged beans (g)	Powder weight (g)	Weight loss (%)
Eugenia aromatica			
0	41 ± 1.7a	4.65 ± 0.06a	18.0 ± 0.34a
0.5	10 ± 1.6b	0.33 ± 0.22b	2.1 ± 0.52b
1.0	4 ± 1.2c	0.09 ± 0.01c	1.2 ± 0.02c
2.0	0d	0c	0c
4.0	0d	0c	0c
Azadirachta indica			
0	39 ± 6.4a	3.96 ± 0.12a	21.5 ± 4.22a
0.5	26 ± 7.6ab	2.01 ± 0.34b	15.0 ± 2.16b
1.0	15 ± 0.4b	0.98 ± 0.05c	6.2 ± 1.25c
2.0	4 ± 0.5c	0.08 ± 0.02d	1.0 ± 0.22d
4.0	0d	0d	0d

Means followed by the same letter in the same column are not significantly different ($P > 0.05$). Tukey's Studentized Range (HSD) Test.

DISCUSSION

The use of plant powders for the protection of stored products is a common phenomenon amongst local farmers in developing countries because of the ease of its application as well as effectiveness in protecting stored produce. The number of progeny produced by *C. cephalonica* in the untreated control was significantly higher than the one treated with dosages of the plant powders. Cocoa beans treated with *E. aromatica* at 2 and 4 g per 100 g of cocoa beans and with *A. indica* treated at 4 g/100 g cocoa beans absolutely suppressed progeny emergence. Our findings are in consonance with that of Allotey and Goswami (1994) who reported that leaves of neem *(A. indica*) caused 100% mortality of *Plodia interpunctella* at 15 g/100 g groundnut and suppressed more than half the population of *Ephestia cautella* at 5 g/100 g groundnut. This study is in agreement with Senguttuvan *et al*. (1995) who reported the efficacy of a range of plant products, including neem leaf powder and edible oil in protecting stored groundnuts against the rice moth, *C. cephalonica* and noted that even though all the plant products and edible oils afforded protection, neem leaf powder and neem oil were most effective.

Several authors have also reported on the efficacy of *E. aromatica* in suppressing insect pest emergence. Olotuah (2013) reported zero *S. zeamais* adult emergence in treatment involving Eugenia dust of particle size 150, 212 and 300 m on 20 g of maize in storage. The mode of action of these plant powders could be due to feeding deterrence which subsequently leads to death of the insects or oviposition deterrence/ovicidal action leading to reduced progeny production.

This study further revealed that the powders of *E. aromatica* and *A. indica* were generally toxic to adult *C. cephalonica*. The plant powders possess some toxic bioactivity which caused significant mortalities of the moth. The bioactivity of the botanicals varied between species because *A. indica* was slower acting than *E. aromatica*. The toxicity of plant materials to insects depends significantly on the chemical composition, which varies across species and plant families and plant powders have been well documented to act slower than the extracts or the pure compounds (Tapondjou *et al*., 2005). The speed of action of the powder of *E. aromatica* was much greater than that of *A. indica* because it achieved 100% mortality at 2 g/100 g cocoa beans on the 4th day after treatment while *A. indica* at the same dosage achieved 100% on the 7th day after treatment. This is further corroborated by the lower LD50 and LD95 values of *E. aromatica* compared to those of *A. indica*.

The powders of *E. aromatica* and *A. indica* exhibited strong residual activity on the moth populations after three months in storage. At higher dosages of 2 and 4 % (w/w), the population of live and dead moths found in the cocoa beans significantly reduced. The results of this study have corroborated reports of previous workers on the effect of *E. aromatica* on other storage pests such as *C. maculatus* (Adedire and Lajide, 2001; Longe, 2004). The mechanisms of the protective action against the cowpea bruchid include direct toxicity to adults and eggs, and inhibition of oviposition by female beetles. It was reported that *E. aromatica* powder still manifested significant contact and fumigant insecticidal activity against the cowpea seed beetle four years after the dry flower buds was powdered. Likewise, *A. indica* has strong residual and asphyxiating effect due to its numerous insecticidal compositions, the most important of which is azadirachtin which has growth inhibiting effect (Koul et al., 1990).

The powders of *E. aromatica* and A. indica significantly reduce cocoa bean damage from attack by *C. cephalonica* and at dosages of 2 and 4 % (w/w), E. aromatic averted the moth attack while *A. indica* completely prevented moth attack at 4 % (w/w). Plant powders applied at these dosages ensured that no cocoa bean was damaged, no weight loss and no cocoa bean powder produced. This may be attributed to the plant powder extracts possessing antifeedant properties. It has been documented that insect damage in stored grains and pulses may amount to 10-40% in countries where modern technologies have not been introduced (Shaaya *et al.*, 1997). It is therefore a common practice among small scale farmers in Africa to protect their stored produce using plant materials. These plant materials are within the reach of the resource poor farmer. The use of clove and neem powders could be advocated for the protection of produce which include cocoa beans, grains and pulses in storage because they do not require high-tech for their application, and moreover, these plant powders can be integrated into a formidable integrated pest management (IPM) systems for stored produce.

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