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Fumigant Activities of Three Plant Powders against Stored Grain Beetles

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Abstract: The insecticidal properties of *Eugenia aromatica*, *Dennettia tripetala* and *Piper guineense* were assessed in the laboratory for their potential to protect stored grains against insect depredation using their fumigant action. Results from fumigant bioassay revealed that responses of test beetles to plant materials was dependent upon susceptibility of insect species, application rate and exposure time. *E. aromatica* powder was the most toxic; evoking the highest mean percentage mortality on the test beetles and this was significantly different ($P \leqslant 0.05$) from the mortalities obtained in treatments with two other plant powders and control. At the highest concentration of 0.47mg/cm^3, *E. aromatica* gave 63.75, 82.5, 67.5, 43.75, and 48.75% mean percentage mortalities on *S. zeamais*, *C. maculatus*, *T. castaneum*, *L. serricorne* and *O. mercator* respectively at four days after treatment. The plant powders used in this study have bioactive components, which are toxic to stored product insects, thus could serve as good substitute for synthetic chemical insecticides like Methyl Bromide. Resource-poor farmers in developing countries could harness the use of these plant materials for protecting their produce against insect attack.

Introduction

Developing countries in tropical regions are faced with problems of malnutrition, food shortage and scarcity due to their inability to protect crops from quality and quantity deterioration caused by the activities of microbes, rodents, and insects pests[1,2]. Food crops particularly cereals and grain legumes form the main diet and protein source among the people in developing countries[3]. However, insect pests usually attack all facets of the crops both in the field and soon after harvest[4].

The production of cereals and leguminous crops by peasant farmers in developing countries in the tropics suffer a setback due to their inability to afford the high cost for procuring synthetic chemical insecticides or effective and efficient storage facilities, which are used for protecting stored produce from insect pest attack[5,6,7]. Apart from the high cost of procuring chemicals and irregular supplies, synthetic insecticides leave residual toxicity on protected food and this could be harmful to man and his livestock [4,8]. Alternative controls aimed at reducing the use of synthetic insecticides are earnestly being sought and in recent decades[9], traditional pest control methods such as the use of plant derived insecticides have attracted researchers as good alternative control agents[10,11]. This paper examines the fumigant property of powders from three plant species and their contact toxicities against coleopterous pests of cereals and grain legumes.

Materials and Methods

Insect Cultures

They were disinfested by a method described by Adedire and Lajide (1999)[12], while the storage beetles (*Callosobruchus maculatus* (Fabricius); *Tribolium castaneum* (Herbst); *Sitophilus zeamais* (Mots.); *Oryzaephilus mercator* (Fauvel) and *Lasioderma serricorne* (Fabricius)) were obtained from established laboratory cultures raised in the storage research laboratory, Federal University of Technology Akure, Nigeria. The insect pests were originally obtained from International Institute for Tropical Agriculture Ibadan (IITA), before rearing in our laboratory. The diets used in rearing the test beetles

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and their bioassay are whole maize (*S. zeamais*), whole cowpea (*C. maculatus*), maize grits (*T. castaneum*), cocoa seed (*L. serricorne*), wheat (*O. mercator*) 750g food media were measured into 1litre Kilner jars. Those grains were obtained from Erekesan market, Akure, Nigeria during October, 2002. Twenty unsexed adults of each insect species were introduced into their respective culturing medium and covered with muslin cloth held tightly in place by rubber bands and was kept in the rearing chamber with a 12 – h photoperiod at ambient condition(28 2℃ and 755% R. H.). Newly emerged (teneral, 1 – 7 days) adult insects were used for each test. Beetles were certified dead when there are no movements after gently probing their abdomen several times with a sharp forceps.

Preparation of Plant Powders

Fruits of *Piper guineense* Thonn and Schum. , *Dennettia tripetala* Baker, and cloves of *Eugenia aromatica* (*Syzygium aromaticum*) Baillon used for this study were obtained fresh from Erekesan market in Akure. The fruits of the plants collected were first dried naturally on laboratory benches at prevailing tropical storage condition. The dried plant materials were pulverized into fine powder using Kenwood electric blender and sieve through a 10 m size mesh. The powder was kept in brown airtight bottle.

Contact Activity of Plant Powders on Beetle Mortality

Different concentrations (0. 1, 0. 2, 0. 3, 0. 4, 0. 5g per 20g food medium) of plant powder/grain mixture were prepared in 9cm diameter Petri dishes and twenty newly emerged adult insects were introduced into the treated grains in the dishes and covered. Weevil mortality was observed on a daily basis for four days and 50% mortality(LD_{50}) was determined. A control experiment was set up without powder treatment. All the treatments were replicated six times.

Fumigant Effect of Plant Powders on Beetle Mortality

The fumigant effects of powders of *E. aromatica*, *D. tripetala* and *P. guineense*, at dosages of 0. 8, 2. 5, 4. 2mg/cm^3 were evaluated on adults *S. zeamais*, *C. maculatus*, *T. castaneum*, *L. serricorne* and *O. mercator* in the laboratory at ambient conditions. Five pairs of newly emerged adult beetles were introduced inside a sac made of muslin cloth containing 20g food medium (Fig. 1). The bag was tied and suspended inside a Glass fumigating chamber (height = 12cm, diameter = 8cm) containing specified concentrations of plant powder. The fumigating chamber was corked and sealed. Each treatment was replicated six times. Beetle mortality was recorded at four days after treatment and the numbers of dead adult beetles were recorded. Beetles were certified dead if they did not respond to forceps probe.

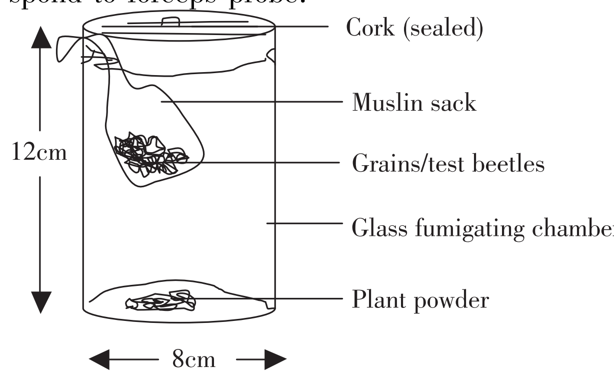

Fig. 1 Showing the fumigating chamber set up

Statistical Analysis

The data from contact toxicity bioassay were analyzed for LD_{50} values and their 95% confidence limits (95% CL) by probit analysis using DPS data processing system. The analysis of variance and Tukey's test for mean separations were also calculated using the DPS (version 3. 01) data processing system[13].

Results

Contact Toxicity

The powder of *E. aromatica* caused the highest mortality to *C. maculatus* (i. e. least LD_{50} 0. 22) at one day after treatment, followed by *D. tripetala* (LD_{50} 0. 24) and *P. guineense* (LD_{50} 0. 30) in that order(Table 1). *D. tripetala* and *P. guineense* gave similar LD_{50} (0. 26) but different confidence limits and slopes in *S. zeamais* infested food media at one day post treatment period. At day two after treatment, *E. aromatica* was still the most potent powder with the least LD_{50} followed by *D. tripetala* and *P. guineense* respectively. Similar trends of plant powder activities were observed at days 3 and 4 in treatments infested with *S. zeamais*, *C. maculatus*, *T. cataneum*, *L. serricorne* or *O. mercator*. The LD_{50} of each plant powder on test beetles decreases as the exposure period increases which is indicative of increase in potency of plant powders with time of exposure. All plant powders tested were effective, however the susceptibility of the insect species used varied with different plant materials. At four days after treatment *C. maculatus* appears the most susceptible insect to *E. aromatica* powder treatment ($LD_{50}=0.08g$) while the least susceptible beetle was *L. serricorne* $LD_{50}=0.24$.

Fumigant Effect of Plant Powders on Beetle Mortality

Fumigant effect of plant powders on the beetles is presented in Table 2. The lowest concentration ($0.8mg/cm^3$) of *E. aromatica* was not effective on test beetles, mortalities ranged between 516.25% at day four after treatment, while the highest concentration ($4.2mg/cm^3$) caused significant ($P \leqslant 0.05$ level, Tukey's test) mortality 43.75% - 82.50% across all the test beetles. Adult mortality in the controls was 0%. The mortality recorded in treatments with *P. guineense* was low (15% - 37.50%) compared to treatments with *E. aromatica* (43.75% - 82.50%) and *D. tripetala* (45% - 65%) at the highest concentration. *D. tripetala* had higher fumigant activity on *L. serricorne* than other plant materials tested. Based on analysis of variance (ANOVA), all the plant powders were effective fumigants at $4.2mg/cm^3$ with percentage mortality significantly ($P \leqslant 0.05$) different from the control (Table 2).

Discussion

Results of this study have revealed that powders of *D. tripetala*, *E. aromatica*, and *P. guineense* were effective as botanical insecticides against all the test beetles. However, their effectiveness was dependent on application rates and exposure periods. The test beetles showed varying degrees of susceptibility. *E. aromatica* had the highest contact toxic effect on the test beetles followed by *D. tripetala* powder while the least effective was *P. guineense.* The observed toxicity of *E. aromatica* is in agreement with findings of Lajide *et. al.* (1998)[14] who observed that *E. aromatica* evoked high contact toxicity on *S. zeamais.* The high beetle mortality observed in *E. aromatica* treated media could be due to the pungent asphixiating smell of its volatile components. In addition to direct toxic effect, plants powder could also produce odours that may confuse of repel beetles Boeke *et al.* (2004)[15].

Since particle size affects dispersion, powders of plant materials have the tendency of coating seeds more uniformly than whole plant or plant parts, thereby enhancing contact with the target pests[16]. The action of *E. aromatica* on these beetles may be as a result of stomach poisoning [14] or contact action. High toxic effect of *E. aromatica* on *S. zeamais* and *T. castaneum* may be as a result of the feeding habits of these pests during which lethal dose of the plant material were ingested [17]. *E. aromatica* contains eugenol, sesquiterpene and caryophylline [18]. Eugenol is toxic and could inhibit growth in insects [18,19].

The volatile components of *E. aromatica* could also result in blockage of spiracles and evoke respiratory impairment or reduce oxygen carrying capacity of the haemolymph. The effectiveness of *D. tripetala* on adult mortality of *C. maculatus*, *S. zeamais* and *T. castaneum* agrees with the reports of Okonkwo and Okoye (1996)[20] who observed that powders of *D. tripetala* resulted in 100% mortality of maize weevil *S. zeamais* and *C. maculatus*. According to Agbakwuru *et. al.* (1978)[21], *D. tripetala* contains (*phenylnitroethane* which is known to have insecticidal activity hence, the action of *D. tripetala* on these storage beetles could be attributed to this active principle found in the plant.

Although *P. guineense* evoked high percentage mortality on the test beetles, its efficacy is lower than those of *E. aromatica* and *D. tripetala.* Similar observations have been made by other workers [22,20,14]. The observed mortality could be ascribed to the presence of amides piperine, Chavicine, N - iso - butyloctadeca-trans - 2 - trans - 4 - dienamide, sylvatine, a (dihydro piperine and trichostachine in the fruits of *P. guineense*[23]. The biological activities of the powder have been linked to the presence of these active principles in the plant because some of these compounds, especially chavicine and piperine have contact toxicity and fumigant action on insects. Mbata *et al.* (1995)[24] had reported that 0.4g/5.0g of powdered seeds of *P. guineense* when admixed with maize, resulted in 50% adult *S. zeamais* mortality. In related study with plant powders Adedire and Akinneye (2004)[25] observed that powder of *Tithonia diversifolia* at 5% concentration impaired oviposition adult emergence and evoked 98% mortality on *C. maculatus.* These results agreed with the observations made in this study where it was observed that though *P. guineense* had some lethal effect on all the test beetles, it was however not as effective as *E. aromatica* and *D. tripetala*.

When used as fumigant, the powders of *E. aromatica* was the most effective of all the three plant materials evoking 82.50% adult mortality in *C. maculatus* followed by *D. tripetala*, while *P. guineense* was the least effective. The effectiveness of *E. aromatica* as bio fumigant may be due to its volatile component and pungent smell, which result in asphyxiating effect on the beetles. This is in line with the observations ofwho reported [26,27] that powders of some botanicals and aromatic plants have some fumigant or lethal effect on storage beetles.

This study has revealed *E. aromatica* as a botanical with high fumigant activity against stored product beetles. This property could be exploited and used to replace synthetic fumigants such as methyl bromide, phosphine gas, which are ecologically intolerable.

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Table 1. Contact toxicity effect(LD_{50} and Confidence limit) of three plant powders on five post harvest insect pests at days post treatment.

Days post treatment	Plant materials	*S. zeamais* LD_{50} (95% CL) * Slope ± SE	*C. maculatus* LD_{50} (95% CL) * Slope ± SE	*T. casternum* LD_{50} (95% CL) * Slope ± SE	*L. serricorne* LD_{50} (95% CL) * Slope ± SE	*O. mecartor* LD_{50} (95% CL) * Slope ± SE
1	*E. aromatica*	0.25 (0.23 – 0.28) * 3.06 ± 0.31	0.22 (0.07 – 0.35) * 3.48 ± 0.32	0.31 (0.28 – 0.34) * 4.03 ± 0.40	0.58 (0.46 – 0.85) * 2.47 ± 0.38	0.47 (0.40 – 0.59) * 3.34 ± 0.44
	D. tripetala	0.26 (0.24 – 0.28) * 3.64 ± 0.34	0.24 (0.22 – 0.26) * 3.13 ± 0.31	0.35 (0.31 – 0.40) * 2.95 ± 0.34	0.63 (0.44 – 5.89) * 11.29 ± 4.12	0.51 (0.43 – 0.70) * 2.58 ± 0.37
	P. guineense	0.26 (0.24 – 0.29) * 3.17 ± 0.32	0.30 (0.28 – 0.34) * 3.19 ± 0.34	0.44 (0.37 – 0.58) * 2.03 ± 0.31	0.81 (0.58 – 1.60) * 2.55 ± 0.48	0.88 (0.54 – 8.31) * 4.85 ± 1.60
2	*E. aromatica*	0.19 (0.17 – 0.21) * 3.90 ± 0.34)	0.13 (0.03 – 0.20) * 3.01 ± 0.32	0.23 (0.21 – 0.25) * 3.73 ± 0.34	0.40 (0.35 – 0.47) * 3.66 ± 0.43	0.32 (0.29 – 0.35) * 3.40 ± 0.35
	D. tripetala	0.23 (0.12 – 0.32) * 3.66 ± 0.33	0.20 (0.01 – 0.36) * 3.24 ± 0.31	0.28 (0.25 – 0.32) * 2.71 ± 0.31	0.48 (0.40 – 0.63) * 2.37 ± 0.39	0.37 (0.32 – 0.44) * 2.61 ± 0.32
	P. guineense	0.23 (0.21 – 0.26) * 3.11 ± 0.31	0.22 (0.12 – 0.31) * 3.48 ± 0.32	0.29 (0.19 – 0.60) * 2.67 ± 0.31	0.64 (0.45 – 2.81) * 9.90 ± 3.15	0.77 (0.54 – 2.01) * 4.41 ± 1.05
3	*E. aromatica*	0.14 (0.12 – 0.17) * 3.39 ± 0.33	0.09 (0.07 – 0.12) * 3.18 ± 0.38	0.17 (0.06 – 0.25) * 3.63 ± 0.33	0.30 (0.27 – 0.33) * 3.53 ± 0.35	0.24 (0.22 – 0.26) * 3.46 ± 0.33
	D. tripetala	0.21 (0.15 – 0.33) * 3.88 ± 0.34	0.16 (0.00 – 0.26) * 2.95 ± 0.30	0.22 (0.19 – 0.25) * 2.39 ± 0.28	0.34 (0.30 – 0.39) * 2.59 ± 0.31	0.28 (0.25 – 0.31) * 3.00 ± 0.31
	P. guineense	0.22 (0.13 – 0.30) * 3.50 ± 0.32	0.20 (0.07 – 0.29) * 3.81 ± 0.34	0.23 (0.21 – 0.25) * 3.27 ± 0.32	0.75 (0.53 – 1.82) * 4.42 ± 1.02	0.78 (0.56 – 1.64) * 3.25 ± 0.66
4	*E. aromatica*	0.11 (0.08 – 0.13) * 3.27 ± 0.37	0.08 (0.05 – 0.12) * 3.81 ± 0.55	0.14 (0.12 – 0.16) * 3.90 ± 0.36	0.24 (0.22 – 0.26) * 3.85 ± 0.35	0.17 (0.08 – 0.23) * 3.51 ± 0.33
	D. tripetala	0.20 (0.06 – 0.29) * 3.86 ± 0.34	0.12 (0.03 – 0.17) 3.38 ± 0.35	0.17 (0.14 – 0.19) * 2.57 ± 0.29	0.26 (0.23 – 0.29) * 2.85 ± 0.30	0.21 (0.19 – 0.23) * 3.14 ± 0.31
	P. guineense	0.20 (0.18 – 0.23) * 3.50 ± 0.32	0.18 (0.10 – 0.24) * 4.14 ± 0.35	0.18 (0.09 – 0.25) * 3.40 ± 0.323	0.75 (0.55 – 1.40) * 2.99 ± 0.56	0.61 (0.49 – 0.93) * 3.67 ± 0.60

Table 2. Fumigant effect of three plant powders on five post harvest insect pests at four days after treatment.

Plants Materials Used	Conc. (mg/cm³)	Percentage mean mortality ± standard error(% Mean ± S. E) *				
		S. zeamais	*C. maculatus*	*T. casternum*	*L. serricorne*	*O. mecartor*
E. aromatica	0.0	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]
	0.8	5.00 ±2.04[ab]	12.50 ±2.04[bc]	8.75 ±1.25[ab]	15.00 ±1.25[cd]	16.25 ±2.0[4b]
	2.5	32.50 ±1.25[d]	40.00 ±1.44[e]	41.52 ±2.39[de]	30.00 ±2.50[e]	30.00 ±2.39[c]
	4.2	63.75 ±1.25[e]	82.50 ±1.39[g]	67.50 ±2.04[f]	43.75 ±3.75[f]	48.75 ±2.04[d]
D. tripetala	0.0	1.25 ±1.25[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]
	0.8	12.50 ±1.25[bc]	21.25 ±1.25[cd]	11.25 ±2.39[ab]	12.50 ±3.15[bc]	3.75 ±1.25[a]
	2.5	18.75 ±3.15[c]	25.00 ±2.39[d]	25.00 ±3.15[c]	21.25 ±2.04[d]	18.75 ±2.04[b]
	4.2	61.25 ±1.44[e]	65.00 ±2.04[f]	50.00 ±2.04[e]	45.00 ±1.25[f]	48.75 ±2.04[d]
P. guineense	0.0	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]
	0.8	2.50 ±1.25[a]	6.25 ±2.04[ab]	7.50 ±2.04[ab]	2.50 ±1.25[a]	1.25 ±1.25[a]
	2.5	5.00 ±2.04[ab]	18.75 ±2.39[cd]	12.50 ±1.44[b]	6.25 ±1.25[ab]	2.50 ±1.44[a]
	4.2	28.75 ±3.75[d]	37.50 ±1.25[e]	35.00 ±1.25[d]	15.00 ±1.44[cd]	15.00 ±2.39[b]
CK	0.0	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]	0.00 ±0.00[a]

* Each value is the percentage mean ± standard error of six replicates. Means followed by different letter(s) vertically are significantly different at P≤0.05 by Tukey's test.

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