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Evaluation of the dust and methanol extracts of *Garcinia kolae* for the control of *Callosobruchus maculatus* (F.) and *Sitophilus zeamais* (Mots)

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Abstract: Insecticidal effects of different doses of the dust and methanol extracts of *Garcinia kolae* on *Callosobruchus maculatus* and *Sitophilus zeamais* were tested. The dust had no significant effect on the two insects; none of them died even at 3 d after treatment. The methanol extracts, however, had rapid lethal effects on both *C. maculatus* and *S. zeamais*. The mortality of *C. maculatus* by the lowest concentration of methanol extracts ranged from 95%~100% whereas in *S. zeamais*, the mortality ranged from 87.5%~100% and 70%~100% in concentrations of 1 g extract+3 ml methanol and 1 g extract+5 ml methanol, respectively, from 24 to 48 h. The least concentration of 1 g extract+15 ml methanol had no significant lethal effect on *Sitophilus zeamais*.

Key words: Insecticidal effect, *Garcinia kolae*, *Sitophilus zeamais*, *Callosobruchus maculatus*
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INTRODUCTION

Crop products are the end result of a sequence of husbandry operations in the field, starting from land preparation to harvesting. After harvesting, it has become necessary to store the products for some period before consumption or use for a variety of purposes that have been given by Adesuyi (1977). However, in the post-harvest period, stored crop products are usually liable to depreciation by pest organisms especially insects. Most important storage pests are Coleopterans and some Lepidopterans.

Varying estimates of losses caused by storage insect pests abound in literature. Appert (1987) reported total crop losses (from harvesting to consumption) to be 40% of production in the hot, humid regions of the world and more than 10% in the dry regions. Estimates of post-harvest crop losses worldwide have also been given as 10%~20% but 25%~40% for the tropics by other researchers (Hill and Waller, 1990). In storage, *Callosobruchus maculatus* causes a major loss. Infestation of stored cowpea could be as much as 98% in markets and in village stores (Albeek, 1996). The magnitude of damage that can occur will depend on many factors such as the kind of crop and insect storage duration and the prevailing environmental conditions (Hill and Waller, 1990). Delima (1987) reported that insect pest problems in the tropical storage environment are greater than those in the temperate climates, presumably because of optimal conditions of temperature and humidity for pest development found in the former.

Many synthetic insecticides have been reported as effective in the control of field and storage insect pests (Hill and Waller, 1990; Obeng-Ofori and Dankwah, 2004; Ogunleye, 2001; 2006; Anyim, 2003; NFMAWR and ODABG, 1996), but their adoption and use by most tropical farmers are hampered by several factors including prohibitive costs, inconsistent supplies of the chemical and safety of workers and consumers (Wolfson *et al*., 1991; Golob

et al., 1999). Insecticides can also lead to pest resurgence.

Current research focus in stored products protection includes the development of non-chemical technologies which may eliminate the use of insecticides and have economic and health benefits for the applicators, consumers and the environment (Murdock *et al*., 1997; Talukder and Howse, 2000; Elhag, 2000). The use of more natural and sustainable methods of protecting harvested crops from insect damage is most favored (Golob *et al*., 1999). A lot of successes have been recorded in this area (Ogunleye, 2000; 2003; 2006; Ogunleye *et al*., 2003; Omotoso and Ogunleye, 2006; Obeng-Ofori and Dankwah, 2004; Elhag, 2000; Kéita *et al*., 2001).

This research work evaluates the possible usage of *Garcinia kolae* extracts and dust for the control of *Sitophilus zeamais* and *Callosobruchus maculatus*.

MATERIALS AND METHODS

Insects culture

A pure culture of the two species of insects, *Sitophilus zeamais* and *Callosobruchus maculatus*, was maintained in the laboratory. Clean uninfested seeds of cowpea and maize were kept separately in Kilner jar-like containers. Twenty randomly selected species of *C. maculatus* and *S. zeamais* were placed in cowpea and maize containers, respectively. These were then left on the laboratory table under ambient environmental conditions of 32 °C and 60% RH. The insects multiplied in the containers within six weeks. All the insects used for these experiments were taken from these containers.

Preparation of plant dust

Five kilograms of fresh, clean seeds of *Garcinia kolae* were purchased at the central market, Igede-Ekiti, a town located in Ekiti State, Nigeria. The seeds were then spread on the laboratory table to air dry under ambient environmental conditions of 32 °C and 50% RH. This was done for a period of two months to ensure proper drying. The seeds were thereafter pulverized using a milling machine, kept in clean plastic containers and preserved in the refrigerator to maintain its freshness.

Application of plant dust

Twenty grams of uninfested cowpea were weighed into Petri-dishes. Into each of these dishes placed were different doses of the plant dusts, 2.5 g, 2.0 g, 1.5 g and 1.0 g, respectively. Twenty species of *S. zeamais* and *C. maculatus* were introduced into each of the cages. They were them shaken vigorously to ensure even distribution of the dust on the insects. Each of the cages was replicated three times. The control had no dust.

Preparation of plant extract

One hundred grams of pulverised plant powder was soaked in 250 ml of methanol in a 500 ml beaker for 5 d. The mixture was shaken vigorously at 12-h intervals to ensure proper soaking of the plant product. After 5 d, decanting of the extract was done and the liquid extract was filtered using a fine cotton cloth. The filtrate was left on the laboratory table for 2 d for total evaporation of the solvent; a pure solid extract was left in the container. This weighed 10 g. It was then kept in the refrigerator.

Different concentrations of the extract were prepared as follows: (1) 1 g of the extract+2 ml of methanol; (2) 1 g of the extract+3 ml of methanol; (3) 1 g of the extract+5 ml of methanol; (4) 1 g of the extract+10 ml of methanol; (5) 1 g of the extract+15 ml of methanol.

Application of plant extract

The insect species were put in the refrigerator for 3 min to immobilize, being prevented from flying away. Twenty species of *C. maculatus* and *S. zeamais* were placed separately in Petri-dishes. Each of the prepared concentrations of the extract was applied topically at the rate of one drop on each insect. A 5 ml hypodermic syringe was used for the application. For the control, only methanol was applied on the insects. The experiment was replicated 3 times. The containers were covered with nets and left on the laboratory table. Percentage mortality of insects was taken on hourly basis.

Statistical analysis

All data collected were subjected to analysis of variance and the means were separated, using Fisher's least significant difference.

RESULTS AND DISCUSSION

The results of the effects of different doses of the dust of *G. kolae* on *C. maculatus* are presented in Table 1. The plant dust had no lethal effect on the insects as none of the insects died in all the treatments and the control even at 3 d post treatment. There was also a gradual increase in all the treatments as well as the control from the 4th day to the 5th day in which the experiment was terminated. The reduction in insect population at this time could be attributed to old ages. Analysis of variance showed no significant difference in all the treatment and the control. From this result, it can be deduced that *G. kolae* is not effective in its powder form for the control of *C. maculatus*. However, the highest dose, 2.5 g of the dust, recorded significantly high percentage mortality. This is a pointer to the fact that, if the dose is increased, further, it is likely to show some potency.

Table 1 Mean percentage mortality of *Callosobruchus maculatus* treated with different rates of the dust of *Garcinia kolae*

Treatment	Mean percentage mortality (%)				
	Day 1	Day 2	Day 3	Day 4	Day 5
2.5 g	0±0	0±0	0±0	30.0±5.2^a	52.2±9.7^a
2.0 g	0±0	0±0	0±0	25.0±5.0^a	42.5±8.3^b
1.5 g	0±0	0±0	0±0	12.5±5.4^b	37.5±4.1^b
1.0 g	0±0	0±0	0±0	30.0±0.0^a	35.0±8.7^b
Control	0±0	0±0	0±0	27.5±6.0^a	35.0±8.7^b

Means followed by the same letter are not significantly different at 5% level of probability using Fisher's least significant difference (LSD)

Table 2 shows the result of the application of plant dust on *Sitophilus zeamais* for 5 d. Little or no mortality was observed on the insects in all the treatments and control for the 5 d period.

Table 2 Mean percentage mortality of *Sitophilus zeamais* treated with different rates of the dust of *Garcinia kolae*

Treatment	Mean percentage mortality (%)				
	Day 1	Day 2	Day 3	Day 4	Day 5
2.5 g	0±0	0±0	0±0^a	2.1±2.1^b	2.5±2.1^b
2.0 g	0±0	0±0	0±0^a	0±0^a	0±0^a
1.5 g	0±0	0±0	2.5±1.8^b	2.5±1.8^b	2.5±1.8^b
1.0 g	0±0	0±0	0±0^a	0±0^a	0±0^a
Control	0±0	0±0	0±0^a	0±0^a	2.5±1.8^b

Means followed by the same letter are not significantly different at 5% level of probability using Fisher's least significant difference (LSD)

Mortality ranged from 0% to 2.5% for all the treatments. This is a clear indication that the population of *S. zeamais* cannot be controlled by the dust of *G. kolae*. The effect of methanol extract of *G. kolae* on *C. maculatus* is presented in Table 3. It was generally observed that the insects died soon after application of the extract, except the lowest concentration in which 95% mortality was recorded. All the treatments had 100% mortality after 2 h of application. It was reported that the active principles of an ideal insecticidal plant should not be poisonous to human. The plant should also be easy to cultivate through agricultural procedures. The mortality rates in the control experiment remained low with 2.5% after 2 h when the experiment was terminated. It can be deduced from this result that there are some insecticidal properties in the test plant which are not available or potent in the powder form. It can however be extracted and made available by methanol. The insecticidal plant powders may be more potent through extraction using appropriate solvent (Makanjuola, 1989; Ogunleye, 2000). Methanol has been used by some researchers to extract some active principles from plant materials. Methanol extracts of *G. kolae*, *A. leiocarpus* and *V. donianas* were tested and found to have anti-microbial effects against methicilin resistant *Staphylococcus aureus*, vancomycin resistant *Enterocollus* and mut-tidung-resistants *Burkholderia capacia* and *Pseudomonas aeriginosa* (Oluronke *et al*., 1999).

Table 3 Mean percentage mortality of *Callosobruchus maculatus* treated with different concentrations of *Garcinia kolae* extracts at 2 h after treatment

Treatment	Mean percentage mortality (%)	
	1 h	2 h
1 g extract+2 ml methanol	100±0^a	100±0^a
1 g extract+3 ml methanol	100±0^a	100±0^a
1 g extract+5 ml methanol	100±0^a	100±0^a
1 g extract+10 ml methanol	100±0^a	100±0^a
1 g extract+15 ml methanol	95.0±5.0^a	100±0^a
Control	2.5±4.3^b	2.5±4.3^b

Means followed by the same letter are not significantly different at 5% level of probability using Fisher's least significant difference (LSD)

Table 4 shows the mean percentage mortality of *S. zeamais* treated with different concentrations of *G. kolae* extracts after 3 d. The highest dose of 2 ml methanol+1 g extract gave 100% mortality after 24 h.

Table 4 Mean percentage mortality of *Sitophilus zeamais* treated with different concentrations of *Garcinia kolae* extracts for 3 d

Treatments	Mean percentage mortality (%)		
	Day 1	Day 2	Day 3
1 g extract+ 2 ml methanol	100±0[a]	100±0[a]	100±0[a]
1 g extract+ 3 ml methanol	87.5±4.3[a]	100±0[a]	100±0[a]
1 g extract+ 5 ml methanol	70.0±7.1[a]	100±0[a]	100±0[a]
1 g extract+ 10 ml methanol	0±0[b]	22.5±4.3[a]	97.5±4.3[a]
1 g extract+ 15 ml methanol	0±0[b]	0±0[b]	15.0±5.0[b]
Control	0±0[b]	0±0[b]	0±0[b]

Means followed by the same letter are not significantly different at 5% level of probability using Fisher's least significant difference (LSD)

For the experiments with 1 g extract+3 ml methanol and 1 g extract+5 ml methanol, mortality ranged from 87.5%~100% and 70%~100%, respectively, from 24 h to 48 h; in the case of 1 g extract+10 ml methanol and 1 g extract+15 ml methanol, mortality of *S. zeamais* ranged from 0% to 100% in the two cases from the 1st day to the 4th day. The result of the control experiment measured 0% throughout the period of study. The potency of *Garcinia kolae* has been attributed to the presence of terpenes, steroids, coumarines, flavonoids, phenolic acid, lignins, xanthones and anthraquinones (Tona *et al*., 2004).

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