

Evaluation of Contact Toxicity and Fumigant Effect of Some Medicinal Plant and Pirimiphos Methyl Powders against Cowpea Bruchid, *Callosobruchus maculatus* (Fab.) [Coleoptera: Chrysomelidae] in Stored Cowpea Seeds

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Abstract

The contact toxicity and fumigant effect of *Azadirachta indica* A. Juss, *Anacardium occidentale* (L), *Piper guineense* Schum and Thonn seeds powders and Pirimiphos methyl (Actellic) dust were evaluated against cowpea bruchid, *Callosobruchus maculatus* (Fab.). Contact toxicity assay show that *A. indica* and *P. guineense* powders have a comparative effect to synthetic insecticide, Pirimiphos methyl. Both were able to cause 100% mortality of *C. maculatus* at all tested concentrations (0.1, 0.2, 0.4 and 0.8g/20g of cowpea seeds) within 7 days of post treatment. However, in the fumigation assay, none of the plant powders was suitable as a fumigant since *A. indica* and *P. guineense* powders could only cause 23.3% and 20% mortality of adult cowpea bruchid respectively after 7 days of application. Pirimiphos methyl powder was good as a fumigant causing 100% mortality of *C. maculatus* after 7 days of application at all tested concentrations.

Keywords: Cowpea seed, *Callosobruchus maculatus*, *Azadirachta indica*, *Anacardium occidentale*, *Piper guineense*, Pirimiphos methyl, Biopesticide

1. Introduction

Food legumes occupy a prominent place in the diets of many people worldwide. Their edible seeds form a cheap source of high protein diets (Okosun and Adedire, 2010). The cowpea bruchid, *Callosobruchus maculatus*, a major insect pest of cowpea in the tropics (Ofuya, 1992) causes great loss of stored cowpea seeds in Nigeria (Caswell, 1970; Taylor, 1971).

Control of field-to-store insect pest like *C. maculatus* has centred mainly on the use of synthetic insecticides and fumigants (Oni and Ileke, 2008; Ashamo, 2010;). This has led to problems such as unavailability of pesticides at critical periods, development of resistant insect strains, and toxicity to the end users (Akinkulolere *et al.*, 2006; Oni and Ileke, 2008; Adedire *et al.*, 2011; Ileke and Oni, 2011; Oni, 2011). Research is ongoing for natural plant materials that are ecofriendly, biodegradable, and with medicinal values that may be used as grains protectant (Adedire and Lajide, 2003; Arannilewa *et al.*, 2006; Ileke and Oni, 2011). The use of plant powders to protect harvested grains against insect pests has been an ancient practice. Many plants, including spices had been reported to have insecticidal properties by many researchers (Ivbijaro and Agbaje, 1986; Adedire and Lajide, 2003; Ashamo, 2004; 2007; Oni and Ileke, 2008).

This present study evaluated the efficacy of three medicinal plant products compared with Pirimiphos methyl (Actellic) powders against cowpea bruchid, *C. maculatus*. This investigation also examined the method of application of these plant products powders.

2. Materials and Methods

This study was conducted at the Environmental Biology and Fisheries Research Laboratory, Adekunle Ajasin University, Akungba Akoko, Ondo State, Nigeria.

2.1 Insect Culture

Adult cowpea bruchid, *C. maculatus* was obtained from established laboratory culture reared on disinfested cowpea seeds, *Vigna unguiculata* (L.) Walp variety Ife brown at ambient temperature of $28\pm 2^{\circ}\text{C}$ and $75\pm 5\%$ relative humidity. The cowpea seeds used for bioassay was obtained from Agricultural Development Project, Ikole Ekiti, Ekiti State, Nigeria and these were firstly cleaned and disinfested by keeping at -5°C for 7 days to kill all hidden infestations. This is because all the life stages, particularly the eggs are very sensitive to cold (Koehler, 2003). The disinfested cowpea seeds were then placed inside a Gallenkamp oven (Model 250) at 40°C for 4 hours (Jambere *et al.*, 1995) and later air dried in the Laboratory to prevent mouldiness (Adedire *et al.*, 2011) before they were stored in plastic containers with tight lids disinfested by swabbing with 90% alcohol.

2.2 Plant Materials

The plant materials used in the present study include *Azadirachta indica*, *Anacardium occidentale* and *Piper guineense* seeds. These materials were sourced fresh from Akola farm at Igbara-Odo Ekiti, Ekiti State, Nigeria. The plants were sun dried for 3 to 7 days. The cleaned dried plants were pulverised into fine powders using a blender (Supermaster®, Model SMB 2977, Japan). The powders were further sieved to pass through 1mm² mesh. The powders were packed in plastic containers with tight lids and stored in a refrigerator at 4°C prior to use.

2.3 Effect of Contact Toxicity of Plants Products and Pirimiphos methyl Powders on Adult Mortality of *C. maculatus*

The plant powders were thoroughly mixed with 20g of cowpea seeds in 250ml plastic containers at 0.0 (control), 0.2, 0.4 and 0.8g concentrations. Similarly, 0.0 (control), 0.2, 0.4 and 0.8g of Pirimiphos methyl dust obtained from Agro Chemical store for comparison were mixed with 20g of cowpea seeds inside 250ml plastic containers. The experiments were set up in a Completely Randomized Design (CRD) and each treatment was replicated three times. Ten pairs of adult *C. maculatus* unsexed (2 to 3 days old) were introduced into the treated and control (Idoko and Adebayo, 2011; Udo, 2011). Bruchid mortality was assessed every 24 hours for three days, then at 7 days after treatment (Ashamo, 2007).

2.4 Fumigant Effect of Plants Products and Pirimiphos methyl Powders on Adult Mortality of *C. maculatus*

Ten grammes of the cowpea seeds were weighed into 50ml plastic film tubes that had been cut opened at the bottom and sealed with mushlin cloth. Plant powders weighing 0.0 (untreated), 0.2, 0.4 and 0.8g were put into another half-cut 50ml plastic film tubes. The 50ml tube and 25ml tube were then joined together with the aid of gum in corresponding orders. Ten pairs of unsexed adult *C. maculatus* (2 to 3 days old) were introduced to the tube containing 10g of cowpea seeds and tight sealed. The experiments were set up in a Completely Randomized Design (CRD) and each treatment was replicated three times. Bruchid mortality was assessed every 24 hours for three days, then at 7 days after treatment (Ashamo, 2007).

2.5 Statistical Analysis

Data were subjected to analysis of variance. Where significant differences existed, treatment means were separated using the New Duncan's Multiple Range Test (Zar, 1984).

3. Results

3.1 Effectiveness of Plants and Pirimiphos methyl Powders as Contact Insecticides

The contact toxicity of *A. indica*, *A. occidentale*, *P. guineense* and pirimiphos methyl powders against *C. maculatus* are show in Table 1. Neem, *A. indica* powder caused 50% mortality of adult cowpea bruchid, *C. maculatus* at rate of 0.8g/20g within 24 hours of post treatment. The corresponding value for *P. guineense* was 47.7% mortality. These were significantly lower ($P<0.05$) than that of pirimiphos methyl dust which caused 77.7% mortality of adult cowpea bruchid, *C. maculatus* at rate of 0.8g/20g of cowpea seeds within 24 hours of application. At three days post treatment, *A. indica*, *A. occidentale* and *P. guineense* caused 93.3%, 43.3% and 90% mortality of adult cowpea bruchid, *C. maculatus* at rate of 0.8g/20g of cowpea seeds compared with pirimiphos methyl who were able to evoked 100% adult mortality of *C. maculatus* at all the concentrations tested. However at seven days post application, *A. indica*, *P. guineense* and pirimiphos methyl powders caused 100% mortality of adult bruchid at all tested concentrations.

3.2 Fumigant Effect of Plant and Pirimiphos Methyl Powders

Table 2 shows the fumigant effect of *A. indica*, *A. occidentale*, *P. guineense* and pirimiphos methyl powders against *C. maculatus*. At three days of application, powders of *A. indica*, *A. occidentale* and *P. guineense* caused mortality of adult cowpea bruchid at all tested concentrations. However pirimiphos methyl powder caused 100% mortality of adult *C. maculatus* at all tested concentrations.

4. Discussion

Neem, *A. indica* powder when used as contact insecticides caused 100% mortality of adult bruchid within 7 days of treatment, an effect that have earlier been reported by Ofuya (1992); Onu and Baba (2003); Mainia and Lale (2004); Mbailao *et al.* (2006). The insecticidal potential of *A. indica* could be attributed to the presence of azadiratins, which is toxic to stored product insect pests (Onu and Baba, 2003; Mainia and Lale, 2004; Mbailao *et al.*, 2006).

Black pepper, *P. guineense* powder also had contact toxicity of comparable effect to that of pirimiphos methyl dust causing 100% adult mortality of bruchid within 7 days of application, an effect that is similar to the report of Adedire and Lajide (2003) that extract of *P. guineense* adversely affected the survival of *Sitophilus zeamais*. This result also validated the reports of Asawalam and Emosairue (2006); Ashamo (2007) that *P. guineense* and Pirimiphos methyl powders caused 100% mortality of adult maize weevil within 7 days of application. These authors established that the biological activity of *P. guineense* could be attributed to the presence of chavin and piperine, an unsaturated amide (Lale, 1992). Ofuya and Dawodu (2002) also reported the effectiveness of *P. guineense* powder against *C. maculatus*.

Cashew seed powder could be very effective against stored product insects if applied at high concentrations. Oparaeke and Bunmi (2006) reported that *A. occidentale* nut shell was highly toxic to *C. subinnotatus* and achieved 100% insect mortality within 48 hours at 7.5% and 100% mortality within 72 hours at 2.5% and 5.0% concentrations. Adedire *et al.* (2011) also reported that cashew kernel extracted with water, methanol, ethanol, acetone, pet- ether and n-hexane caused 85% to 100% adult mortality of *C. maculatus*. They attributed these insecticidal activities to the presence of secondary plant compounds such as anacardic acid and cardinal (Rehn and Espig, 1991). Others are quercetin and kampferol glycosides (Oliver- Bever, 1986).

In this study, the lethal effect of these powders on cowpea bruchid could be as a result of contact toxicity. Most insects breathe by means of trachea which usually opens at the surface of the body through spiracles (Adedire *et al.*, 2011). These spiracle might have been blocked by the powders thereby leading to suffocation of the insects.

The plants powders were not effective as fumigants with the highest mortality of 23.3% and 20% caused by *A. indica* and *P. guineense* powders respectively. This result validated the report of Ashamo (2007) who recorded that *P. guineense* powders tested for fumigant effect could only caused 28.4% adult mortality of maize weevil.

5. Conclusion and Recommendation

Results obtained from this study revealed the potentials of *A. indica* and *P. guineense* seeds powders as plant derived insecticides against cowpea bruchid, *C. maculatus*. The medicinal values, availability, biodegradable, low costs rate and potential as biopesticide make them candidates in upgrading traditional crop protection practices in sub-Saharan Africa.

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Table 1. Comparison of percentage mortality of adult *C. maculatus* in cowpea seeds treated with plants powders and Pirimiphos methyl dust for contact toxicity

Plant powder	Conc. g/20g Cowpea	Mean % Mortality at days Post treatment \pm S.E.			
		1	2	3	7
<i>A. indica</i>	0.1	10.0 \pm 0.0ab	30.0 \pm 0.0b	67.7 \pm 0.3d	100.0 \pm 0.0d
	0.2	14.7 \pm 0.6b	46.3 \pm 0.6c	70.0 \pm 0.0d	100.0 \pm 0.0d
	0.4	20.0 \pm 0.0b	54.7 \pm 0.3c	84.4 \pm 0.2e	100.0 \pm 0.0d
	0.8	50.0 \pm 0.0c	79.3 \pm 0.4d	93.3 \pm 0.6ef	100.0 \pm 0.0d
<i>A. occidentale</i>	0.1	0.0 \pm 0.0a	0.0 \pm 0.0a	20.0 \pm 0.0b	47.3 \pm 0.6b
	0.2	0.0 \pm 0.0a	0.0 \pm 0.0a	27.3 \pm 0.6bc	53.3 \pm 0.6bc
	0.4	0.0 \pm 0.0a	0.0 \pm 0.0a	33.7 \pm 0.3c	60.0 \pm 0.0c
	0.8	0.0 \pm 0.0a	0.0 \pm 0.0a	43.3 \pm 0.6c	63.3 \pm 0.6c
<i>P. guineense</i>	0.1	10.0 \pm 0.0ab	30.0 \pm 0.0b	63.3 \pm 0.6d	100.0 \pm 0.0d
	0.2	12.3 \pm 0.6b	44.2 \pm 0.2c	67.7 \pm 0.3d	100.0 \pm 0.0d
	0.4	19.7 \pm 0.3b	50.0 \pm 0.0c	83.3 \pm 0.6e	100.0 \pm 0.0d
	0.8	46.7 \pm 0.3c	76.7 \pm 0.3d	90.0 \pm 0.0ef	100.0 \pm 0.0d
Pirimiphos methyl	0.1	63.3 \pm 0.6d	80.0 \pm 0.0d	100.0 \pm 0.0f	100.0 \pm 0.0d
	0.2	67.7 \pm 0.3de	83.3 \pm 0.6de	100.0 \pm 0.0f	100.0 \pm 0.0d
	0.4	70.0 \pm 0.0de	87.7 \pm 0.3e	100.0 \pm 0.0f	100.0 \pm 0.0d
	0.8	77.7 \pm 0.3e	90.0 \pm 0.0e	100.0 \pm 0.0f	100.0 \pm 0.0d
Control	0.0	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a

Each value is a mean of \pm standard error of three replicates. Means within the same column followed by the same letter(s) are not significantly different at ($P>0.05$) from each other using New Duncan Multiple Range Test.

Table 2. Comparison of percentage mortality of adult *C. maculatus* in cowpea seeds treated with plants powders and Pirimiphos methyl dust for fumigant effect

Plant powder	Conc. g/20g Cowpea	Mean % Mortality at days Post treatment \pm S.E.			
		1	2	3	7
<i>A. indica</i>	0.1	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.2	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.4	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	10.0 \pm 0.0a
	0.8	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	23.3 \pm 0.6b
<i>A. occidentale</i>	0.1	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.2	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.4	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.8	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
<i>P. guineense</i>	0.1	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.2	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.4	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a
	0.8	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	20.0 \pm 0.0b
Pirimiphos methyl	0.1	10.0 \pm 0.0a	53.3 \pm 0.6b	100.0 \pm 0.0b	100.0 \pm 0.0c
	0.2	23.3 \pm 0.6b	60.0 \pm 0.0bc	100.0 \pm 0.0b	100.0 \pm 0.0c
	0.4	30.0 \pm 0.0bc	67.7 \pm 0.3cd	100.0 \pm 0.0b	100.0 \pm 0.0c
	0.8	37.7 \pm 0.3c	77.7 \pm 0.3d	100.0 \pm 0.0b	100.0 \pm 0.0c
Control	0.0	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a	0.0 \pm 0.0a

Each value is a mean of \pm standard error of three replicates. Means within the same column followed by the same letter(s) are not significantly different at ($P>0.05$) from each other using New Duncan Multiple Range Test.