

Insecticidal Evaluation of Some Plant Materials as Grain Protectants Against the Maize Weevil, *Sitophilus zeamais* (Mots) (Coleoptera: Curculionidae)

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Abstract: Laboratory experiments were carried out to investigate the efficacy of ethanolic extract of four plant materials, namely: *Zanthoxylum xanthoxyloides* (Lam.) Waterm., *Aristolochia ringens* (Vahl), *Colocasia esculenta* (L.) Schott and *Morinda lucida* (L.) on the maize weevil *Sitophilus zeamais* (Mots). Parameters assessed were adults toxicity, adult emergence, percentage of reduction of plants and damage effect of weevils to the grains at 0.0, 1.0, 1.5 and 2.0% (w/v) concentrations. *Zanthoxylum xanthoxyloides* evoked 100% adult mortality within 24 h after treatment; and *A. ringens* also significantly caused 100% mortality within 48 h. *Aristolochia ringens* at all concentrations provided good protection to the grains stored for 49 days and also gave 100% reduction value as well as 0.00% damage and 0.00 Weevil Perforation Index. This study therefore revealed the grain protectant ability of *A. ringens* and *Z. xanthoxyloides* on the maize weevil, *Sitophilus zeamais*. The bioassay procedure used and results obtained are reported.

Key words: Grain protectant, *aristolochia ringens*, *sitophilus zeamais*, percentage of reduction, weevil perforation index

INTRODUCTION

A major problem facing agriculture in developing countries like Nigeria, is post-harvest losses caused by storage pests. The losses of grain in storage either directly through consumption of the grain, or indirectly by producing "hot-spots" (thereby causing migration of moisture and as a result making the grains more suitable for other pests) are some of the inevitable losses encountered^[1].

Control of these insect pests relies heavily on the use of synthetic insecticides. Although synthetic insecticides can control the pests, the adoption and use of any of the compounds in the traditional African storage system is limited, due largely to unavailability and prohibitive cost of pesticides, lack of pesticide application skills, environmental residual effects of pesticides and, lately, problems with pesticide adulteration^[2]. These factors have therefore necessitated a shift into searching for naturally-occurring plant-derived insecticides as protectants. It is expected that such protectants must be easy to use, cheap, accessible, biodegradable with greater selectivity^[3].

The plants evaluated, except *C. esculenta*, have been confirmed to be medicinal. *Zanthoxylum xanthoxyloides* is known to be active in reverting sickle-cell

anaemia *in vitro*^[4], *A. ringens* is efficacious in treating stomach disorders and peptic-ulcer while *M. lucida* is used to treat yellow fever^[5]. *Colocasia esculenta* is a stout rhizomatous herb with peltate leaves and it is cultivated for its tuber^[6].

The research work was therefore designed to investigate the insecticidal activities of ethanolic extracts of the plants on *Sitophilus zeamais* within a short exposure period.

MATERIALS AND METHODS

Insect cultures: Clean, undamaged and uninfested maize grains were purchased from Oja-Oba (King's market) in Akure, Nigeria. Whole and undamaged grains were hand-picked in the laboratory, packed in a polythene bag and disinfested in a deep freezer for 96 h. The grains were later air-dried in the laboratory to prevent mouldiness^[7]. *Sitophilus zeamais* was collected from an established culture reared on disinfested maize grains at ambient temperatures of 28±2°C and 75±5% relative humidity in a grain storage research laboratory, Federal University of Technology, Akure, Nigeria. *Sitophilus zeamais* was then transferred onto the grains in 1litre Kilner jars and from this, an established culture for the experiment was maintained as new generations emerged.

Table 1: Plants evaluated for insecticidal activities against *S. zeamais*

Scientific name	Family	Parts used	Common name
<i>Zanthoxylum xanthoxyloides</i>	Rutaceae	Root bark	Sichuan/Chinese pepper
<i>Aristolochia ringens</i>	Aristolochiaceae	Root bark	Gaping Dutchman's pipe
<i>Colocasia esculenta</i>	Araceae	Leaves	Cocoyam
<i>Morinda lucida</i>	Rubiaceae	Stem bark	Indian Morus

Plant materials: The selected plants and parts used for this experiment are presented in Table 1. *Colocasia esculenta* was collected fresh from a farm within the University, while the other plants were purchased from Isinkan market in Akure. They were all brought to the Laboratory, further air-dried and pulverized into fine powders with the aid of a ball-mill machine. The powders were sieved and transferred into airtight containers.

Plant extract formulation: Twenty-five grammes of each plant material was soaked in 200 mL of ethanol (96%)/distilled water solvent mixture (9:1v/v) for 72 h. They were later filtered using the porcelain filter and fine-pored muslin cloth. The filterates were concentrated *in vacuo* and the concentrates obtained were further air-dried to remove traces of solvent. They were later transferred into sample bottles and kept in refrigerator and from them the concentrations applied were prepared when required.

Toxicity bioassay: The toxicity assay of the plants on adult *S.zeamais* was accomplished in Petri-dishes (9 cm diameter) containing 25 g maize grains with concentrations of 1.0, 1.5 and 2.0% (w/v) ethanolic plant extracts. The dishes were left open for 1 h to allow likely traces of ethanol to evaporate; after which 20 teneral adults (0-48 h old) were introduced into the dishes and mortality was observed for 96 h (i.e., 4 days). Solvent-treated grains served as controls and all experiments were carried out in 3 replicates. Adults were assumed dead when probed with forceps and there was no response.

Effect of extracts on adult emergence and reduction: Having removed both dead and live adults weevils from the treated grains, they were stored for 49 days (i.e., 7 weeks). At the end of the 49-day observation period, the number of adults that emerged (F_1 generation) were counted and the values obtained were used as criteria for evaluation of the percentage of reduction. The F_1 adult emergence in the treated sample compared to the F_1 adult emergence in the control is an index of effectiveness of the treated material in reducing infestation. The percentage of reduction of the extracts was then calculated using the formular^[8]:

$$\% \text{Red} = 100 - \left[\frac{E_T}{E_C} \right] \times 100$$

where E_T = No of emerged adults from treated sample and E_C = No of emerged adults from control.

Effect of extracts on grain damaged by weevils: The extent of grain damage by *S. zeamias* was determined at the end of experiment by counting the insect-damaged kernels in the treated and control experiments. The Percentage Damage (PD)^[9] and Weevil Perforation Index (WPI)^[10] caused by the weevils to the grains were calculated

$$PD = \frac{\text{Total number of treated grains perforated}}{\text{Total number of grains}} \times 100$$

$$WPI = \frac{\% \text{age of treated grains perforated}}{\left(\frac{\% \text{age of control}}{\text{grains perforated}} \right) + \left(\frac{\% \text{age of treated}}{\text{grains perforated}} \right)} \times 100$$

Data analysis: Data obtained were subjected to one-way analysis of variance and where significant differences ($p < 0.05$) existed, means were separated by New Duncan's Multiple Range Test.

RESULTS AND DISCUSSION

Toxicity bioassay of extracts: The toxic effect of plant extracts on adult *S. zeamais* within 96 h is presented in Table 2. Results showed that the percentage mortality of *S. zeamias* significantly varied within the plants extracts and the concentrations of a particular plant extract. Adult mortality also significantly varied with the exposure period. *Zanthoxylum xanthoxyloides* evoked 100% adult mortality within 24 h, while *A. ringens* also produced 95% mortality within the same hours. At 48 h post-treatment *A. ringens* evoked 100% mortality at 1.50 and 2.00% concentrations while it was 98.35% mortality at 1.00% of same plant treatment. At 72 h, all the 3 concentrations of *A. ringens* has evoked 100% mortality. At 96 h, *Z. xanthoxyloides* evoked 100% mortality in all the concentrations. The results obtained have indicated *Aristolochia ringens* to be very active even at lower concentrations within 48 h. However *Z. xanthoxyloides*

Table 2: Toxicity effect of Plant extracts on adult mortality of *S. zeamais*

Plants	Concentration (% w/v)	Mean % mortality (\pm S.E) 96 h Post-treatments			
		24 h	48 h	72 h	96 h
<i>Z. xanthoxyloides</i>	1.00	40.00 \pm 5.77 ^c	76.65 \pm 6.67 ^d	83.35 \pm 3.33 ^e	100.00 \pm 0.00 ^f
	1.50	63.35 \pm 3.33 ^e	83.35 \pm 6.67 ^{de}	93.35 \pm 3.33 ^f	100.00 \pm 0.00 ^f
	2.00	100.00 \pm 0.00 ^f	100.00 \pm 0.00 ^f	100.00 \pm 0.00 ^g	100.00 \pm 0.00 ^f
<i>A. ringens</i>	1.00	76.66 \pm 1.67 ^e	98.35 \pm 1.67 ^f	100.00 \pm 0.00 ^g	100.00 \pm 0.00 ^f
	1.50	88.33 \pm 1.67 ^h	100.00 \pm 0.00 ^f	100.00 \pm 0.00 ^g	100.00 \pm 0.00 ^f
	2.00	95.00 \pm 2.88 ^{hi}	100.00 \pm 0.00 ^f	100.00 \pm 0.00 ^g	100.00 \pm 0.00 ^f
<i>C. esculenta</i>	1.00	26.65 \pm 3.33 ^b	46.65 \pm 3.33 ^b	50.00 \pm 0.00 ^b	55.00 \pm 2.88 ^b
	1.50	53.35 \pm 3.33 ^d	60.00 \pm 0.00 ^c	63.35 \pm 1.67 ^c	80.00 \pm 2.88 ^c
	2.00	96.65 \pm 3.33 ^{hi}	98.35 \pm 1.67 ^f	100.00 \pm 0.00 ^g	100.00 \pm 0.00 ^f
<i>M. lucida</i>	1.00	65.00 \pm 2.88 ^{ef}	75.00 \pm 2.88 ^d	78.35 \pm 1.67 ^d	85.00 \pm 0.00 ^d
	1.50	73.35 \pm 1.67 ^{fg}	86.66 \pm 1.67 ^e	90.00 \pm 2.88 ^f	91.65 \pm 1.67 ^e
	2.00	76.65 \pm 3.33 ^g	88.35 \pm 1.67 ^e	95.00 \pm 0.00 ^g	98.35 \pm 1.67 ^f
Control (Solvent-treated)	0.00	0.00 \pm 0.00 ^a	1.65 \pm 1.67 ^a	5.00 \pm 0.00 ^a	6.65 \pm 1.67 ^a

Each value is the mean of three replicates. Means followed by the same letter are not significantly different ($p < 0.05$) from each other, using New Duncan's Multiple Range Test

Table 3: Emergence of adult *S. zeamais* (after 49 days of storage) and Percentage of reduction

Plants	Concentration (%w/v)	Mean No of emerged adults counted (\pm S.E)	% reduction (\pm S.E)
<i>Z. xanthoxyloides</i>	1.00	14.00 \pm 0.67 ^b	50.58 \pm 2.03 ^d
	1.50	4.33 \pm 0.33 ^a	84.72 \pm 1.17 ^e
	2.00	0.00 \pm 0.00 ^a	100.00 \pm 0.00 ^f
<i>A. ringens</i>	1.00	0.00 \pm 0.00 ^a	100.00 \pm 0.00 ^f
	1.50	0.00 \pm 0.00 ^a	100.00 \pm 0.00 ^f
	2.00	0.00 \pm 0.00 ^a	100.00 \pm 0.00 ^f
<i>C. esculenta</i>	1.00	18.67 \pm 0.33 ^b	34.10 \pm 1.17 ^b
	1.50	14.00 \pm 0.67 ^b	50.58 \pm 2.03 ^d
	2.00	4.67 \pm 0.33 ^a	83.52 \pm 1.17 ^e
<i>M. lucida</i>	1.00	16.67 \pm 0.67 ^b	41.16 \pm 2.35 ^c
	1.50	14.67 \pm 0.33 ^b	48.22 \pm 1.17 ^d
	2.00	5.00 \pm 0.67 ^a	82.35 \pm 2.03 ^e
Control (Solvent-treated)	0.00	28.33 \pm 0.33 ^c	0.00 \pm 0.00 ^a

Means followed by the same letter are not significantly different ($p < 0.05$) from each other using New Duncans Multiple range test

was active within 24 h; but results have shown that higher concentrations may be active as from 24 h while the lower concentrations may take up to 96 h post-treatment before they could cause 100% mortality. *Aristolochia ringens* possesses a strong choky odour, which may be a factor for its potency. The odour may have exerted a toxic effect by disrupting normal respiratory activity of the weevil, thereby resulting in asphyxiation and subsequent death^[9]. A related plant species, *Aristolochia albida* was discovered to have acidic metabolites like *Aristolic acid*, *Aristolochic acid*, *Aristolone* and *Aristolotctam*. These metabolites may also be present in *A. ringens* and may have caused its high potency against the adult weevil; and it may have acted as contact or food poison. There are eleven known species of *Zanthoxylum* in Nigeria^[11]. The information on metabolites of the test plant (*Z. xanthoxyloides*) is very scanty. However, the insecticidal activity of a Hawaiian species of *Zanthoxylum* has been described to have metabolites like Isobutylamides, aliphatic ketones, mono-and sesquiterpenes^[12].

Presently, an investigation is going on to identify the metabolites in the Nigerian species: *A. ringens* and *Z. xanthoxyloides*.

Effect of extracts on adult emergence, reduction and grain damage:

The effect of plant extracts on development and adult emergence of F_1 generation and reduction values after 49 days of storage are presented in Table 3. Adult emergence decreased with increase in concentrations. No emergence was observed in the treatment with *A. ringens* at all concentrations, while 2.00% of *Z. xanthoxyloides* also have no emergence.

It has been reported that adult weevils lay their eggs inside the grains; and upon hatching, the larvae begins to feed inside the grains, excavating some tunnels as they develop^[13]. Treatment of grains with the extracts may probably have killed the larvae or may possibly have rendered the grain tissues unedible for them. This will then result to death and subsequently, no adult emergence.

Aristolochia ringens gave 100% reduction in the adult emergence in all the concentrations followed by *Z. xanthoxyloides*. Plants that have reduced number of adult emergence always indicate high percentage of reduction. This therefore shows further, the potency of *A. ringens* in its activities against the maize weevil. Some level of significance were also shown on the values.

Table 4: Damage by *S. zeamias* to the grains (after 49 days of Storage)

Plants	Concentration (%w/v)	Total no. of grains	No of grains damaged	Grains undamaged	% damaged	*WPI
<i>Z. xanthoxyloides</i>	1.0	232	12	220	5.17	12.68
	1.5	234	4	230	1.70	4.56
	2.0	235	0	235	0.00	0.00
<i>A. ringens</i>	1.0	232	0	232	0.00	0.00
	1.5	229	0	229	0.00	0.00
	2.0	239	0	239	0.00	0.00
<i>C. esculenta</i>	1.0	235	24	211	10.21	22.30
	1.5	227	12	215	5.28	12.92
	2.0	227	5	222	2.20	5.82
<i>M. lucida</i>	1.0	232	17	215	7.32	17.06
	1.5	237	14	223	5.90	14.22
	2.0	232	6	226	2.58	6.76
Control (solvent treated)	0.0	236	84	152	35.59	50.00

*Weevil Perforation Index (WPI). A value above 50 is an index of negative grain protectant or enhancement of infestation by weevil

Zanthoxylum xanthoxyloides was less active than *A. ringens* by the values it gave. It's activity might be as a contact poison on the adult weevils, but there was less activity on its development and emergence. In a related work, it was observed that the root bark powder of *Z. xanthoxyloides* was less inhibitory to development of cowpea seed bruchid, *Callosobruchus maculatus* larva. The extracted constituents may not be absorbed into the internal tissue where larvae feed and thus not enough titer is consumed as the 1st stage larvae eat into the seed^[14].

The values obtained for the grain damage by weevils and Weevil Perforation Index (WPI) are presented in Table 4. A similar trend of activities of the plant extracts was observed. *Aristolochia ringens* was observed to be most active by preventing any grain damage and WPI. Next to it was *Z. xanthoxyloides* which also gave some higher values of damage and WPI at 1.00 and 1.50%, but no damage at 2.00% concentration. Others gave some relatively higher values. However, none of the values were up to 17.8% percentage damage and 25.00 Weevil Perforation Index (representing half of control values obtained). These values therefore suggest that the four plant extracts have displayed some degree of potency as control agents; but *A. ringens* and *Z. xanthoxyloides* were more potent than the two others.

CONCLUSION

This research work has shown some two other potent plant materials, *Aristolochia ringens* and *Zanthoxylum xanthoxyloides* as likely grain protectants against the maize weevil, *Sitophilus zeamias*; which can be active within 1-4 days of treatment. The plant materials are medicinal; therefore less toxic to humans. Their protectant ability qualifies their use especially among low-resources farmers who store small amounts of grain for consumption and planting. *Colocasia esculenta* and *Morinda lucida* may be more active if applied at higher concentrations.

An investigation into the active metabolites in the two plants and a study of their modes of action on some selected pests is presently going.

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