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Potentiality of some botanical extracts as biopesticides against the maize weevil, *Sitophilus zeamais* Motsch. (Coleoptera: Curculionidae)

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Abstract
Laboratory studies were conducted to investigate the efficacy of some botanicals in the management of *Sitophilus zeamais* Motsch infesting stored sorghum. The selected plant species were *Euphorbia balsamifera* Aiton, *Lawsonia inermis* L. and *Mitracarpus hirtus* L. Chloroform leaf extracts of the botanicals applied at 6.25, 12.50, 25.00, 50.00 and 100.00 mg ml^{-1} were used to determine their effect on adult survival, the concentration of total body protein and inhibition of acetylcholinesterase enzyme (AChE) activity in the weevils at 30 ± 2 ºC and 70 ± 5% R.H. The botanicals resulted in adult mortalities ranging from 10.00 ± 0.00 to 90.00 ± 5.77%, reduced body protein and significantly inhibited the activity of AChE in *S. zeamais*. The botanicals tested could serve as biopesticides in the management of *S. zeamais* infesting sorghum in the storage.

Keywords: AChE, adult mortality, body protein, biopesticides, efficacy

1. Introduction
Insect pests are a key constraint to the effective production and utilization of cereal crops in sub-Saharan Africa (SSA), and post-harvest losses resulting from insects remain a huge challenge [1]. Insect infestations of stored products are one of the significant agricultural development problems in the tropics, which results in a substantial waste of farm produce and hence considerable loss to the economy [2, 3, 4].

The major insect pests that attack stored sorghum grains include the *Sitophilus* spp., *Rhyzopertha dominica* (F.), *Sitotroga cerealella* (Olivier.), *Trogoderma granarium* (Everts), *Oryzaephilus surinamensis* (L.) and *Tribolium* spp. [5, 6, 7].

S. zeamais is the most devastating storage pest of maize, causing serious management problems facing agriculture in developing countries [8]. The grain damage caused by *S. zeamais* to cereal crops prompted researchers to investigate more on controlling strategies of the weevils in maize, wheat, and even sorghum [7, 9, 10, 11].

Considering the dual necessity to achieve food security and safety, especially in the developing countries, there is a need for effective pest management approaches for smallholder farmers who form the bulk of producers in SSA [1]. For that, a number of approaches ranging from cultural to application of synthetic pesticides have been advanced for the management of post-harvest insect pests.

Despite effective pest control both on farm fields and storage provided by synthetic insecticides, there is growing concern about the potential hazards to the ecosystem [1, 11, 12]. Hill [13] among others, had earlier emphasized that, to reduce the hazards associated with chemical application in stored cereals, Integrated Pest Management (IPM) approach, involving all the suitable techniques and methods in maintaining pest population below economic injury level (EIL) is necessary.

To complement IPM strategies, botanicals have been used for many years by small scale farmers in many parts of Africa to protect stored products from insect infestations [14]. Plant materials have been applied to a commodity, usually at a rate of 1 to 5% (w/w), to protect stored grains from insect pest infestations [15]. Studies on some botanical products in Africa indicate their effectiveness against insect pests of stored products [4, 7, 16, 17. 18].

Plant materials could be toxic to insect pests of storage by causing adult mortality, reducing biochemical components (such as body protein, carbohydrate and lipid) and inhibiting acetylcholinesterase (AChE) activity. Previous findings proved that a vast number of botanicals are toxic against *S. zeamais* [19, 20, 21, 22].

In view of the promising effects of some botanicals against insect pests of stored products, *Euphorbia balsamifera* Aiton, *Lawsonia inermis* L. and *Mitracarpus hirtus* (L.) DC were selected for this study due to their reported bioactive compounds, which have been suggested to be of insecticidal importance [18, 23, 24].

2. Materials and Methods

2.1 Rearing of *S. zeamais*

Methods of Suleiman *et al*. [18] were adopted for raising the adult weevils. Fifty copulating pairs of *S. zeamais* were obtained from infested grain stores at Katsina Central Market, Nigeria. The weevils were introduced into each 500 ml rearing bottle containing 250 g of the disinfested sorghum grains, which served as parent stock. The bottles were covered with muslin cloth and kept in the incubator for oviposition at 30 ± 2 ºC and 70 ± 5% R.H. for 7 days, after which the adult weevils were removed. The bottles were maintained in the incubator under the same condition for adult emergence. Emerging weevils were sieved and released into well labeled separate bottles containing fresh disinfested sorghum grains and maintained in the incubator for 7 days. Progeny of 0 – 7 days old was sieved and used for the experiments.

2.2 Collection and preparation of chloroform leaf extracts

A sufficient amount of fresh leaves of *E. balsamifera*, *L. inermis* and *M. hirtus* (Plate I) were collected from the unfarmed area around some villages in Batagarawa Local Government Area of Katsina State, Nigeria. The leaves were rinsed with distilled water to remove any dust and unwanted particles. They were shade-dried in one Postgraduate Laboratory of the Biology Department, UMYUK at room temperature. The dried leaves were ground into powder using a laboratory blender and sieved using a laboratory sieve with a mesh size of 80 microns to obtain a fine powder. The powders were separately kept in black polythene bags to avoid photodegradation, discoloration and moisture uptake [25, 26].

E. balsamifera *L. inermis* *M. hirtus*

Plate I: Plant Species Selected for the Study

Extraction was done using the Soxhlet extraction apparatus. The resulting extracts were air-dried to remove traces of the solvent and stored in the refrigerator at 4 ^{0}C [27] prior to use for the laboratory experiments.

The concentrations of the extracts were made by diluting 0.125, 0.25, 0.50, 1.00 and 2.00 g of crude extracts of *E. balsamifera*, *L. inermis* and *M. hirtus* in 20 ml of chloroform to obtain 6.25, 12.50, 25.00, 50.00 and 100.00 mgml^{-1}, respectively.

2.4 Determination of adult mortality of *S. zeamais*

Crude extracts were diluted with chloroform to make different concentrations of 6.25, 12.50, 25.00, 50.00 and 100.00 mgml^{-1}. Twenty grams of sorghum grains was weighed and put in treatment bottles (250 ml) and mixed with 1 ml of chloroform extracts at the five concentrations, while the control contained the grains only. The grain mass was mixed thoroughly with the aid of glass rod and air-dried until complete solvent evaporation. All treatments were arranged in a completely randomized design (CRD) in the incubator with three replications. The set-ups were inspected daily to remove, count and record any dead weevils in each treatment for seven days.

Adult percent mortality was assessed as:

$$\% \text{ Mortality} = \left(\frac{\text{Number of Dead Weevils}}{\text{Total Number of Weevils}}\right) \text{X } 100$$

2.5 Determination of median lethal concentrations (LC_{50}) of chloroform leaf extracts

To evaluate LC_{50} values, methods of Ebadollahi and Mahboubi [28] were adopted. The number of dead weevils at the end of seven days exposure to chloroform extracts was used to determine LC_{50} of the botanicals. LC_{50} values were calculated by using probit analysis with SPSS (version 16.0) software package.

2.6 Determination of body protein in *S. zeamais* treated with LC_{50} of chloroform leaf extracts

Samples for protein assays were prepared following Askar *et*

al. [20]. Adult weevils exposed to LC_{50} of chloroform extracts of the botanicals for 7 days were freeze killed at – 20 ^{0}C. A sample of 0.5 g of the insects was taken from each bottle and homogenized in 5 mL volumes (w/v) of phosphate buffer (pH 7.2) using a glass homogenizer under ice. The total homogenates was centrifuged at 4000 rpm for 10 min at 4 ^{0}C using Sorvall ST 40R centrifuge machine (ThermoFisher SCIENTIFIC, Germany). The supernatant was transferred to new Eppendof's tubes and preserved at – 20 ^{0}C for spectrophotometry.

The body protein was estimated following the method of Lowry *et al*. [29]. Fifty µL of the supernatant was added to 3 ml of Lowry reagent (50 ml of 2% Na_2CO_3 in 0.1 N NaOH containing 0.5 g sodium tartrate plus 1 ml of 0.1 g $CuSO_4$ in a liter of distilled water). The mixture was vortexed and incubated for 10 minutes at room temperature. Then 0.3 ml of freshly prepared folin reagent (2.00 N Folin-Ciocalteu reagent diluted 1:1 with distilled water) was added to the mixture and incubated for 30 minutes. This was repeated three times.

Three replicates of a standard solution was also prepared by dissolving 100 mg of Bovine serum albumin (BSA) in 100 ml of 0.1 N NaOH. Thus 1 mg mL^{-1} of BSA was obtained and kept as stock solution. The stock solution was diluted into different concentrations of 200, 400, 600, 800 and 1000 µgml^{-1} by adding 490, 480, 470 460 and 450 of 0.1 N NaOH to 10, 20, 30, 40 and 50 µL of the stock solution, respectively.

The absorbance was measured spectrophotometrically after 30 minutes at 750 nm against the blank for both test samples and the standard solutions. The protein concentration was calculated and expressed as µgml^{-1} wet tissue using the formula:

$$\text{Concentration of body protein, } x = \frac{y}{m}$$

Where: x = the concentration;
y = absorbance of the solution; and
m = slope of the standard curve.

2.7 Determination of AChE activity inhibition in *S. zeamais* exposed to LC_{50} of chloroform leaf extracts

Adults of *S. zeamais* were fumigated with lethal concentrations of the chloroform extracts of *E. balsamifera*, *L. inermis* and *M. hirtus* as fumigant toxicity assay. After seven days of fumigation, the insects were removed, stored in Eppendorf tubes and used for determination of AChE activity. Fumigated weevils were homogenized in phosphate buffer saline (50 mM, pH 8) using a Teflon glass tissue homogenizer. The homogenate was centrifuged at 10,000 rpm for 20 min at 4 ºC. The supernatant was filtered and used as the AChE preparation.

A portion (0.1 mL) of acetylcholine iodine (ATChl) (0.5 mM) was added to 0.1 mL of enzyme source followed by addition of 0.05 mL of 5,5-Dithio-bis 2-nitrobenzoic acid (DTNB) (0.33 mM). A 1.45 mL of phosphate buffer (50 mM, pH 8) was then added. The mixture was incubated for 3 min at 25ºC. Absorbance was evaluated at 412 nm for 6 minutes and compared to that of the control. All measurements were made in triplicate.

AChE activity inhibition was calculated according to the following formula [30]:

$$\%\ \text{AChE activity inhibition} = \frac{A_{Control} - A_{Treatment}}{A_{Control}} \times 100$$

Where: $A_{Control}$ = the absorbance of the untreated control; and $A_{Treatment}$ = the absorbance of the treated samples

2.8 Data Analysis

Data generated were tested for normality using Shapiro-Wilk and Jacque-Bera normality tests and found to be non parametric. Therefore, data from adult mortality, concentration of body protein and AChE activity inhibition were subjected Kruskal Wallis test using GraphPad Instat (version 7.03). Probit analysis was employed to calculate LC_{50} of the extracts using SPSS (version 16.0). Significantly different means were separated by Dunn's multiple comparison test. All analyses were performed at $p<0.05$ level of significance.

3. Results

3.1 Adult mortality of *S. zeamais* exposed to sorghum grains treated with chloroform leaf extracts of selected botanicals

Chloroform leaf extracts of the selected botanicals applied at different concentrations of 6.25, 12.50, 25.00, 50.00 and 100.00 mgml^{-1} exhibited varying percentage mortalities of *S. zeamais* within 1, 3, 5 and 7 DAT. Treating the grains with *E. balsamifera* resulted in different adult mortalities of 3.33 ± 3.33 to 10.00 ± 0.00% at 6.25 mgml^{-1} within 7 DAT (Table 1). The adult mortalities in 12.50 mgml^{-1} of the botanical varied between 6.67 ± 3.33 and 20.00 ± 0.00% from 3 to 7 DAT. At 25.00 mgml^{-1}, the mortality was recorded as 6.67 ± 3.33 to 26.67 ± 3.33% from 1 to 7 DAT as shown in Table 1. When the concentration was increased to 50.00 mgml^{-1}, adult mortality of *S. zeamais* increased and ranged from 10.00 ± 0.00 to 36.67 ± 6.67% within the observation period. Further increase in the concentration to 100.00 mgml^{-1} resulted further increase in adult mortality of the weevils which varied between 13.33 ± 3.33 and 73.33± 3.33%.

The application of chloroform extracts of *L. inermis* to sorghum grains at 6.25 mgml^{-1} concentrations resulted in adult mortality of *S. zeamais* in the range of 3.33 ± 3.33 to 26.67 ± 3.33% within 1 to 7 DAT. Application rate of 12.50 mgml^{-1} of the botanical caused 6.67 ± 3.33, 16.67 ± 3.33, 33.33 ± 3.33 and 46.67 ± 3.33% after 1, 3, 5 and 7 DAT (Table 1). The mortalities recorded in 25.00 mgml^{-1} within 7 DAT ranged from 10.00 ± 0.00 to 53.33 ± 3.33%, while those in 50.00 mgml^{-1} varied between 13.33 ± 3.33 and 63.33 ± 3.33%. At 100.00 mgml^{-1} of chloroform extract of *L. inermis* the adult mortality of *S. zeamais* was 16.67 ± 3.33 to 90.00 ± 5.77% in the treated sorghum grains.

The adult mortality of *S. zeamais* in grains treated with chloroform extracts of *M. hirtus* varied over concentration and number of days after treatment. No adult mortality was caused by the botanical treatments at 6.25 mgml^{-1} within 1 DAT, but it was observed to be 20.00 ± 0.00% at 7 DAT (Table 1). After one day of treatments with 12.50 mgml^{-1}, the botanical led to the death of 6.67 ± 3.33% of the weevils which increased to 16.67 ± 3.33, 23.33 ± 3.33 and 26.67 ± 3.33% in 3, 5 and 7 DAT, respectively. At 25.00. 50.00 and 100.00 mgml^{-1} the adult mortality ranged from 10.00 ± 0.00 to 40.00 ± 0.00, 13.33 ± 3.33 to 50.00 ± 0.00 and 16.67 ± 3.33 to 83.33 ± 3.33%, respectively (Table 1). Application of permethrin at 0.025 mgml^{-1} caused total mortality of the weevils within 5 DAT. No mortality was recorded in the control.

Analysis of variance showed significant difference ($p<0.05$)

in adult mortality of *S. zeamais* exposed to sorghum grains treated with chloroform leaf extracts of *E. balsamifera*, *L. inermis* and *M. hirtus* applied at different concentrations.

3.2 Median lethal concentrations (LC_{50}) of chloroform leaf extracts of selected botanicals against *S. zeamais*

Chloroform extracts of *E. balsamifera* had the highest LC_{50} value as 65.90 $mgml^{-1}$ (42.64 – 136.77) followed by *M. hirtus* with 33.22 $mgml^{-1}$ (23.17 – 51.62) and the least was in *L. inermis* with 18.70 $mgml^{-1}$ (12.53 – 26.25) at 7 DAT (Table 2). The slopes ranged from 4.735 to 5.369.

3.3 Concentration of body protein of *S. zeamais* exposed to sorghum grains treated with chloroform leaf extracts of selected botanicals

Figure 1 shows that the body protein in *S. zeamais* exposed to chloroform extracts were 40.37 ± 0.38, 35.56 ± 0.54 and 42.22 ± 0.64 µg/ ml for *E. balsamifera*, *L. inermis* and *M. hirtus*, respectively. The control had 100.00 ± 0.00 µg/ ml protein. The protein concentration was significantly different (KW = 9.982, p < 0.0187) among the treatments. Dunn's multiple comparisons test showed that weevils exposed to selected botanicals had statistically similar protein content which was significantly lower than in those in the untreated grains.

3.4 Inhibition of AChE activity in *S. zeamais* by chloroform leaf extracts of selected botanicals

Inhibition of AChE activity in *S. zeamais* recorded in chloroform leaf extract treatments ranged from 61.96 ± 1.04 to 69.41 ± 0.68% with the highest in *L. inermis* and the least in *E. balsamifera* (Figure 2). There was a significant difference (KW = 10.569, p = 0.0143) in activity inhibition of the enzyme among weevils exposed to LC_{50} of chloroform extracts of the botanicals. The AChE activity inhibition in all treatments was statistically similar and differed from the control which was 0.00% (Dunn's multiple comparisons test).

Table 1: Adult mortality of *S. zeamais* exposed to chloroform leaf extracts of selected botanicals applied at varying concentrations

Treatment	Conc. ($mgml^{-1}$)	Mean Adult Mortality (% ± S.E.)			
		Exposure Period (Days)			
		1	3	5	7
E. balsamifera	6.25	0.00 ± 0.00^{a}	3.33 ± 3.33^{a}	6.67 ± 3.33^{a}	10.00 ± 0.00^{a}
	12.50	0.00 ± 0.00^{a}	6.67 ± 3.33^{a}	13.33 ± 3.33^{a}	20.00 ± 0.00^{b}
	25.00	6.67 ± 3.33^{a}	13.33 ± 3.33^{b}	20.00 ± 5.77^{b}	26.67 ± 3.33^{b}
	50.00	10.00 ± 0.00^{b}	23.33 ± 3.33^{b}	26.67 ± 3.33^{b}	36.67 ± 6.67^{b}
	100.00	13.33 ± 3.33^{b}	36.67 ± 6.67^{c}	60.00 ± 5.77^{c}	73.33± 3.33^{c}
L. inermis	6.25	3.33 ± 3.33^{a}	6.67 ± 3.33^{a}	23.33 ± 3.33^{b}	26.67 ± 3.33^{b}
	12.50	6.67 ± 3.33^{a}	16.67 ± 3.33^{b}	33.33 ± 3.33^{b}	46.67 ± 3.33^{b}
	25.00	10.00 ± 0.00^{b}	20.00 ± 0.00^{b}	36.67 ± 3.33^{b}	53.33 ± 3.33^{c}
	50.00	13.33 ± 3.33^{b}	26.67 ± 3.33^{b}	43.33 ± 5.77^{b}	63.33 ± 3.33^{c}
	100.00	16.67 ± 3.33^{b}	30.00 ± 0.00^{b}	50.00 ± 0.00^{c}	90.00 ± 5.77^{d}
M. hirtus	6.25	0.00 ± 0.00^{a}	10.00 ± 0.00^{a}	16.67 ± 3.33^{a}	20.00 ± 0.00^{a}
	12.50	6.67 ± 3.33^{a}	16.67 ± 3.33^{b}	23.33 ± 3.33^{b}	26.67 ± 3.33^{b}
	25.00	10.00 ± 0.00^{b}	26.67 ± 3.33^{b}	33.33 ± 3.33^{b}	40.00 ± 0.00^{b}
	50.00	13.33 ± 3.33^{b}	30.00 ± 0.00^{b}	46.67 ± 5.77^{b}	50.00 ± 0.00^{c}
	100.00	16.67 ± 3.33^{b}	43.33 ± 3.33^{c}	76.67 ± 3.33^{c}	83.33 ± 3.33^{d}
Permethrin	0.025	50.00 ± 5.77^{c}	90.00 ± 0.00^{d}	100.00 ± 0.00^{d}	100.00 ± 0.00^{d}
Control	0.00	0.00 ± 0.00^{a}	0.00 ± 0.00^{a}	0.00 ± 0.00^{a}	0.00 ± 0.00^{a}

Conc. = Concentration.

Means in the same column followed by different letter superscript are significantly different at p<0.05 by the Dunn's Multiple Comparisons Test.

Table 2: Median lethal concentration (LC_{50}) of chloroform leaf extracts of selected botanicals against *S. zeamais* at 7 DAT

Botanicals	LC_{50} ($mgml^{-1}$)	95% Confidence Limits		Slope ± S.E.
		Lower Bound	Upper Bound	
E. balsamifera	65.90	43.64	136.77	4.735 ± 0.281
L. inermis	18.70	12.53	26.25	5.369 ± 0.276
M. hirtus	33.22	23.17	51.62	5.142 ± 0.269

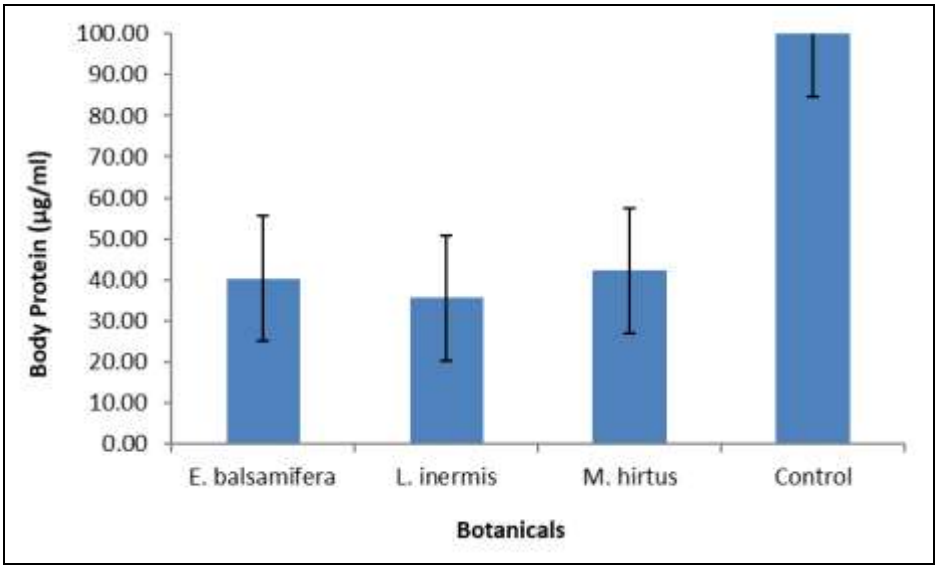

Fig 1: Concentration of body protein in *S. zeamais* exposed to LC_{50} of chloroform leaf extracts of selected botanicals

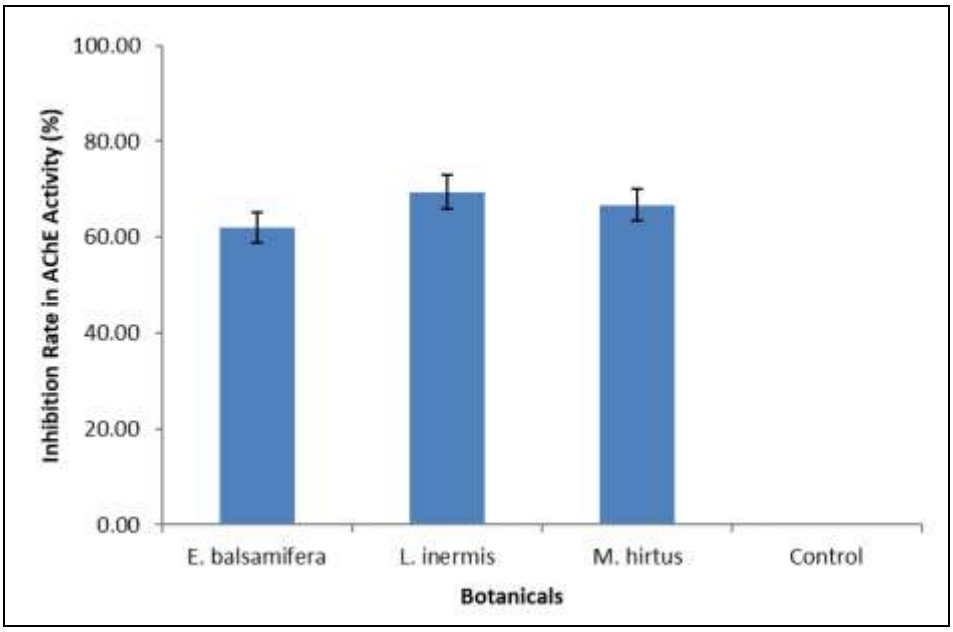

Fig 2: AChE Activity inhibition in *S. zeamais* exposed to LC_{50} of chloroform leaf extracts of selected botanicals

4. Discussion

4.1 Effect of chloroform leaf extracts of selected botanicals on adult mortality of *S. zeamais*

This study has revealed that leaves of *E. balsamifera*, *L. inermis* and *M. hirtus* caused significant mortalities of adult *S. zeamais*. The mortality effects of *E. balsamifera* against *S. zeamais* in this investigation conform the findings of Suleiman and Suleiman [31] who reported a high (90.00%) adult mortality of *C. maculatus* in cowpea seeds treated with leaf powder of *E. balsamifera* at 1.0/ 20 g (w/w) within 96 hours of exposure.

The high mortality of *S. zeamais* caused by chloroform leaf extract of *L. inermis* confirms the earlier reports that 77.40% mortality of adult *S. oryzae* was observed after 180 days of post treatments at 2% *L. inermis* in stored wheat [32]. Leaf powder of *L. inermis* was reported to have caused 33.33% adult mortality of *T. castaneum* in stored groundnuts when applied at 20/ 250 g (w/w) at 14 DAT [33].

Furthermore, this study revealed that chloroform leaf extracts of *M. hirtus* resulted in significant adult mortality of *S. zeamais* in sorghum grains. This finding concurs with a previous report that ethanolic leaf extract of *M. hirtus* caused total mortality of *S. zeamais* at 3 DAT when applied at 50.00 $mgml^{-1}$ [34]. Another report indicated that 100.00 $mgml^{-1}$ acetone extract of *M. hirtus* resulted in 63.33% adult mortality of *R. dominica* at 7 DAT [24].

This study has revealed that high mortality of insects exposed to plant extracts might have exhibited contact toxicity against the weevils as suggested by Hikal *et al*. [35]. The penetration of the botanicals could be possible as the active compounds might have diffused in through membraneous areas of sclerites of the exoskeleton. As the botanicals pass through the weevils' exoskeleton, they probably became abrasive to the epithelial layer, thereby poisoning the living tissues.

Chloroform extracts of *L. inermis* and *M. hirtus* applied at the concentration of 100.00 $mgml^{-1}$ resulted in adult mortality of *S. zeamais* which was not significantly different from what permethrin caused at 7 DAT. Permethrin as a synthetic pyrethroid, acts by interfering with the electrical signal passing down the axon of insect's nerve cells leading to loss of coordination, general breakdown of physiological homeostasis and eventual death of the insect [36].

All the selected botanicals were effective against the weevil. This was possible because plant species contain secondary metabolites which are vast storehouse of compounds such as the steroids, phenolic compounds and tannins with wide range of biological activity reported to have great impact on insecticidal activities [35, 37]. Other bioactive compounds such as terpenoids, flavonoids, alkaloids, saponins and glycosides were found in the leaf extracts of *E. balsamifera*, *L. inermis* and *M. hirtus* [24].

GC-MS analysis confirmed the presence of different compounds sucn as oileic acid Eicosyl ester, decanoic acid, hydrogen peroxides and N,N-diethyl nediamine in acetonic leaf extracts of *E. balsamifera*, *L. inermis* and *M. hirtus* and found to have some insecticidal properties [24].

4.2 Lethal concentration (LC_{50}) of selected botanicals against *S. zeamais*

The present study investigated LC_{50} of *E. balsamifera*, *L. inermis* and *M. hirtus* against *S. zeamais* in sorghum grains. Lethal concentrations of some botanicals used to control insect pests of storage have been determined [38, 39].

LC_{50} of the plants showed that the chloroform extracts had great efficiency by causing 50% adult mortality of *S. zeamais* in sorghum even at lowest concentration within 1 to 7 DAT. *L. inermis* was more effective than the other botanicals followed *M. hirtus* and then *E. balsamifera*. The effectiveness of the selected botanicals in killing adult weevils is in conformity with Biswas *et al.* [37] who reported high efficiency of *L. inermis* against *T. castaneum*.

Low LC_{50} values of chloroform extracts of the test botanicals concur with another finding which who reported that among methanolic extracts of the plants tested *Momordica charantia* L. was the most effective with LC_{50} of 2.82 mg / 20 g maize grains against *S. zeamais* [40].

4.3 Effect of chloroform leaf extracts on body protein of *S. zeamais*

There was a decrease in body protein of the maize weevils exposed to median lethal concentrations (LC_{50}) of all the botanicals compared to the control. The reduction in body protein agrees with Askar *et al.* [20] who reported that both clove oil and diatomaceous earth reduced body protein of *S. zeamais* and *S. oryzae* from 0.10 mg/ ml in the control to 0.09 and 0.06 mg/ ml, respectively. It was earlier reported that sub-lethal concentrations of spinosad resulted in a significant decrease in protein content of *Glyphodes pyloalis* Walker [41]. This decrease in body protein could be attributed to low feeding efficiency of the insects due to antifeedant effect of many other insecticides and hence a decrease in protein concentration [35]. Another reason might be as a result of protein break down into amino acids which help in energy supply to the insect [42].

It was noted that proteins are important for individual-level fitness-associated traits such as body size, growth rate and fecundity [41]. At high levels of organization, proteins have been linked to population dynamics, life cycles and even biological diversification. It was found that application of chloroform extracts of *E. balsamifera*, *L. inermis* and *M. hirtus* significantly affected body protein of *S. zeamais* which could probably affect the aforementioned biological aspects of the weevil. The reduction in concentration of the body protein might have contributed to decline in the insect's population and hence decreased infestations in the sorghum grains. This could also be attributed to decrease in vitellogenin, a precursor of yolk component, present of in the body protein of vitellogenic females, which its amount reflects the insect reproductive performance [34].

4.4 Effect of chloroform leaf extracts on activity of AChE in *S. zeamais*

AChE is a hydrolytic enzyme in insect body that influences the nervous system [43]. It is the key enzyme which terminates nerve impulses by catalyzing the hydrolysis of the neurotransmitter acetylcholine in the nervous system [44]. In the present study the LC_{50} of the extracts of *E. balsamifera*, *L. inermis* and *M. hirtus* significantly inhibited the activity of AChE in *S. zeamais*. This is in agreement with a finding that methyl alcohol, ethyl acetate and petroleum ether fruit extracts of *Illicium verum* Hook. F. inhibited the activity of AChE in *S. zeamais* [43]. Similarly, the LC_{50} of essential oils of *Pimpinella anisum* and *Ocimum basilicum* have demonstrated their interference with AChE activity in *S. granarius* [44]. Recently, methanolic extract of *Urginea maritime* at a concentration of 1000 µg/ ml was found to result in 73.37% inhibition of the activity of AChE in *S. oryzae* [30]. Also, another finding has confirmed the inhibitory effect of botanicals on the activity of AChE where it was reported that LC_{50} of *Cuminum cyminum* essential oil reduced AChE activity in *S. zeamais* to 66.90% compared to 31.59% of the control [22].

The inhibitory effect on AChE activity in *S. zeamais* exposed to lethal concentration of all the botanicals has shown their possible potentiality as biopesticides. This is because it has been reported that inhibition of AChE in cholinergic synapses of the nervous system is the primary mechanism of acute toxicity of insecticides [44]. Therefore, the botanicals probably have neurotoxic effects on *S. zeamais* by interfering with the passage of impulses in the insect nervous system leading to inability of AChE to hydrolyze acetylcholine which builds up in the synapse and results in excessive neuro excitation [45]. The mortality effects of *E. balsamifera*, *L. inermis* and *M. hirtus* against *S. zeamais* recorded in this study may also be attributed to their ability to inhibit the activity of AChE [45]. Thus, the neurotransmitting system of the weevils represents a target for insect control.

5. Conclusion

All the test botanicals were found toxic to *S. zeamais* by causing mortalities at all concentrations after 7 days of post treatment. The LC_{50} values of the botanicals were low and that of chloroform leaf extract of *L. inermis* was the lowest indicating its high level of efficacy. The study also revealed that the selected botanicals resulted in reducing body protein and inhibition of AChE activity in the weevils. This infers the botanicals' role in interfering with reproductive performance especially egg yolk production as well as blocking transmission of nerve impulses which leads to knock down and the insect death. The botanicals demonstrated potentialities as biopesticides against *S. zeamais* which may probably contribute to the control strategies of insect pests infesting sorghum.

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7. References

1. Midega CAO, Murage AW, Pittchar JO, Khan ZR. Managing storage pests of maize: Farmers' knowledge, perception and practices in western Kenya. Crop Protection. 2016;90:142-149. http://dx.doi.org/10.1016/ j.cropro.2016.08.033.
2. Abbas AB, Ndunguru G, Mamiro P, Alenkhe B, Mlingi N, Bekunda M. Post-harvest food losses in a maize-based farming system of semi-arid savannah area of Tanzania. Journal of Stored Products Research. 2014;57:49-57. http://dx.doi.org/10.1016/j.jspr.2013.12.004.
3. Sori W. Effect of selected botanicals and local seed storage practices on maize insect pests and health of maize seeds in Jimma zone. Singapore Journal of Scientific Research. 2014;4(2):19-28. DOI: http://dx.doi.org/10.3923/ sjsres.2014.19.28.
4. Suleiman M, Rugumamu CP. Management of insect pests of stored sorghum using botanicals in Nigerian traditional stores. Journal of Stored Products and Postharvest Research. 2017;8(9):93-102. DOI: http://dx.doi.org/ 10.5897/JSPPR2017.0247.
5. Utono IM. Assessment of grain loss due to insect pest during storage for small-scale farmers of Kebbi. IOSR Journal of Agriculture and Veterinary Science. 2013;3(5):38-50.
6. Mailafiya DM, Dauda Z, Degri MM, Kabir BGJ, Maina YT. An appraisal of insect pests associated with cereal grains traded in Maiduguri, Borno State, Nigeria. International Letters of Natural Science. 2014;14:76-83.
7. Suleiman M. Application of botanical powders for the management of stored sorghum insect pests in small-scale farmers' storage structures of Northern Nigeria. IOSR Journal of Agriculture and Veterinary Science. 2021;14(3)I:12-24. DOI: https://doi.org/10.9790/2380-1403011224.
8. Abulude FO, Ogunkoyal MO, Ogunleye RF, Akinola AO, Adeyemi AO. Effect of palm oil in protecting stored grain from *Sitophilus zeamais* and *Callosobruchus maculatus*. Journal of Entomology. 2007;4(5):393-396.
9. Goftishu M, Belete K. Susceptibility of sorghum varieties to the maize weevil *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae) African Journal of Agricultural Research. 2014;9(31):2419-2426. DOI: http://dx.doi.org/ 10.5897/AJAR2014.8634.
10. Khaliq A, Nawas A, Ahmad NH, Sagheer M. Assessment of insecticidal potential of medicinal plant extracts for control of maize weevil, *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). Basic Research Journal of Agricultural Science and Review. 2014;3(11):100-104.
11. Cosmas P, John CT, Agathar K, Ronald M, Kufa M, Betty C. Use of botanical pesticides in controlling *Sitophilus zeamais* (Maize Weevil) on stored *Zea mays* (Maize) grain. Modern Concepts and Developments in Agronomy. 2018;1(4):1-4.
12. Rotimi J, Evboumwan CO. Deterrent effects of citrus peel oils on oviposition and adult emergence of cowpea weevil, *Callosobruchus maculatus* (F) (Coleoptera: Bruhidae). Advances in Applied Science Research, 2012;3(6):3545-3550.
13. Hill DS. Agricultural Insect Pests of the Tropics and their Control. Cambridge University Press. New York, 1987, 746.
14. Tesema K, Kurabachew H, Tadesse TF. Evaluation of the efficacy of plant powders, cow dung ash and melathion dust against *Callosobruchus chinensis* L. (Coleoptera: Bruchidae) on chickpea in Jole Andegna: Southern Ethiopia. Journal of Agricultural Studies. 2015;3(2):129-144. http://dx.doi.org/10.5296/ jas.v3i2.7775.
15. Dales MJ. A Review of Plant Materials Used for Controlling Insect Pests of Stored Products (NRI Bulletin 65) [Working paper], 1996, 91.
16. Rugumamu CP. Insecticidal activity of botanical pesticides on *Callosobruchus chinensis* (L.) (Coleoptera: Bruchidae) in stored green gram, *Vigna radiate* (L). International Journal of Agricultural Research. 2015;4(1):013-020.
17. Ojo JA, Omoloye AA. Development and life history of *Sitophilus zeamais* (Coleoptera: Curculionidae) on cereal crops. Advances in Agriculture, 2016, 1-8.
18. Suleiman M, Rugumamu CP, Ibrahim ND. Repellency potential of some botanicals against the maize weevil, *Sitophilus zeamais* (Motschulsky, 1855) (Coleoptera: Curculionidae) in Stored Sorghum. Polish Journal of Entomology. 2018;87:85-99. DOI: http://dx.doi.org/ 10.2478/pjen-2018-0007.
19. Adedire CO, Akinkurolere RO, Obembe OM. Efficacy of cashew kernel extracts in the control of the maize weevil, *Sitophilus zeamais* (Coleoptera: Curculionidae). Archives of Phytopathology and Plant Protection. 2012;45(7):831-839. DOI: https://doi.org/10.1080/03235408.2011. 598259.
20. Askar SI, Al-Assaal MS, Nassar AMK. Efficiency of some essential oils and insecticides in the control of some *Sitophilus* insects (Coleoptera: Curculionidae). Egypt Journal of Plant Protection Research. 2016;4(2):39-55.
21. Ibrahim YJ, Abugri AD, Afun JVK. Efficacy of ethanolic leaf extract of *Chromolaena odurata* in controlling *Sitophilus zeamais* in stored maize. Journal of Experimental Agriculture International. 2016;14(5):1-10.
22. Chaubey MK. Evaluation of insecticidal properties of *Cuminum cyminum* and *Piper nigrum* essential oils against *Sitophilus zeamais*. Journal of Entomology. 2017;14(4):148-154. DOI: http://dx.doi.org/10.3923/ je.2017. 148.154.
23. Idris MM, Mudi SY, Datti Y. Phytochemical screening and mosquito repellent activity of the stem bark extracts of *Euphorbia balsamifera* (Ait). Chem Search Journal. 2014;5(2):46-51.
24. Abubakar R, Suleiman M, Wagini NH. Phytochemical and insecticidal properties of some botanical extracts against the lesser grain borer, *Rhyzopertha dominica* F. (Coleoptera: Bostrichidae). Journal of Entomology and Zoology Studies. 2021;9(2):06-13. DOI: https://doi.org/ 10.22271/j.ento.2021.v9.i2a.8452.
25. Chayengia B, Patgiri P, Rahman Z, Sarma S. Efficacy of different plant products against *Sitophilus oryzae* (Linn.) (Coleoptera: Curculionidae) infestation on stored rice. Journal of Biopesticides. 2010;3(3):604-609.
26. DFNPS Describe the different ways that food spoils. Food Safe Tvsit. Department of Food, Nutrition and Packaging Science, Clemson University, 2012, http://www.foodsafetysite.com/educators/competencies/general/microbiology/mic6.html. Accessed on 23rd April, 2020.
27. Abou-Elnaga ZS. Efficacy of extracts of some Egyptian plants against economically important stored grains pest *Sitophilus oryzae* L. Journal of Entomology and Zoology Studies. 2015;3(1):87-91.

28. Ebadollahi A, Mahboubi M. Insecticidal activity of the essential oil isolated from *Azilia eryngioides* (PAU) Hedge et Lamond against beetle pests. Chilean Journal of Agricultural Research. 2011;71(3):406-411.
29. Lowry OH, Passonneau JV, Rock MK. The stability of pyridine nucleotides. The Journal of Biological Chemistry. 1961;236(10):2756-2759.
30. Maazoun AM, Hlel TB, Hamdi SH, Belhadj F, Jemâa, JMB, Marzouki MN. Screening for insecticidal potential and acetylcholinesterase activity inhibition of *Urginea maritima* bulbs extract for the control of *Sitophilus oryzae* (L.). Journal of Asia-Pacific Entomology. 2017;20:752-760.
DOI: http://dx.doi.org/10.1016/j.aspen.2017 04.004.
31. Suleiman M, Suleiman HY. Control of *Callosobruchus maculatus* (F.) [Coleoptera: Bruchidae] using leaf powders of *Euphorbia balsamifera* L. and *Lawsonia inermis* L. International Journal of Science, Environment and Technology. 2014;3(1):100-109.
32. Arya M, Tiwari R. Efficacy of plant animal origin bioproducts against lesser grain borer, *Rhyzopertha dominica* (Fab.) in stored wheat. International Journal of Recent Scientific Research. 2013;4(5):649-653.
33. Joel OO. Efficacy of plant extracts against *Tribolium castaneum* Herbst in stored groundnut (*Arachis hypogaea* L.). African Journal of Plant Science. 2015;9(2):90-96.
34. Suleiman M, Sani I, Yusuf AM, Abdullahi BK. Entomocidal activity of some plant extracts against *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). African Journal of Agricultural Research. 2019;14(34):1862-1869.
DOI: 10.5897/AJAR2019.14271.
35. Hikal WM, Baeshen RS, Said-Al Ahl HAH. Botanical insecticides as simple extractives for pest control. Cogent Biology. 2017;3:1-9. DOI: https://doi.org/10.1080/ 23312025.2017.1404274.
36. Don-Pedro KN. Pesticide Science and Toxicology. University of Lagos Press, Lagos, 2010, 270.
37. Biswas KK, Sharmin N, Rabbi MA. Evaluation of the insecticidal activity of *Lawsonia inermis* Linn. against the red flour beetle, *Tribolium castaneum* (Herbst). Natural Products. 2016;12(1):8-11.
38. Souza AP, Marques MR, Mahmoud TS, Bolzani VS, Caputo BA, Canhete GM *et al*. Insecticidal effect of extracts from native plants to Mato Grosso do Sul, Brazil, on *Sitophilus zeamais* Mots. (Coleoptera: Curculionidae). Bio Assay. 2010;5(1):1-5.
39. Jayakumar M, Arivoli S, Raveen R, Tennyson S. Repellent activity and fumigant toxicity of a few plant oils against the adult rice weevil *Sitophilus oryzae* Linneaus 1783 (Coleoptera: Curculionidae). Journal of Entomology and Zoology Studies. 2017;5(2):324-335.
40. Rani PU, Devanand P. Efficiency of different plant foliar extracts on grain protection and seed germination in maize. Research Journal of Seed Science. 2011;4(1):1-14.
41. Piri F, Sahragard A, Ghadamyari M. Sublethal effects of spinosad on some biochemical and biological parameters of *Glyphodes pyloalis* Walker (Lepidoptera: Pyralidae). Plant Protection Science. 2014;50(3):135-144.
42. Nath BS, Suresh A, Mahendra VB, Kumar RP. Changes in metabolism in haemolymph and fat bodies of the silkworm, *Bombyx mori* L. in response to organophosphorus insecticides toxicity. Ecotoxicity and Environmental Safety. 1997;36:169-173.
43. Li J, Guo Q, Han S, Jiang L, Liang G. Types, morphologies and distribution of antennal sensilla of *Quadrastichus erythrinae* (Hymenoptera: Eulophidae). Florida Entomologist. 2013;96(4):1288-1297.
DOI: https://doi.org/10.1653/024.096.0407.
44. Abo El Makarem H, El Kholy SE, Abdel-Latif A, Seif AI. Physiological and biochemical effects of some essential oils on the granary weevil, *Sitophilus granartius* (L.) (Coleoptera: Curculionidae). Egypt Journal of Experimental Biology. 2015;11(2):117-123.
45. Rajashekar Y, Raghavendra A, Bakthavatsalam N. Acetylcholinesterase inhibition by biofumigant (Coumaran) from leaves of *Lantana camara* in stored grain and household insect pests. Biomed Research International, 2014, 1-6.
DOI: http://dx.doi.org/ 10.1155/2014/187019.