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Evaluation of botanical powders and extracts from Nigerian plants as protectants of maize grains against maize weevil, *Sitophilus zeamais* (Motschulsky) [Coleoptera: Curculionidae]

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**Abstract**

Toxicities of leaf powders and extracts of *Acanthus montanus*, *Acanthospermum hispidum*, *Alchornea laxiflora* and *Argyreia nervosa* against maize weevil (*Sitophilus zeamais*) were evaluated. Powders were tested at dose 3.0g/20g while extracts were tested at concentration 3%/20g of maize grains. Mortality, oviposition, and adult emergence rates as well as weight loss, seeds damage and weevil perforation index (WPI) were evaluated. Phytochemical constituents of the experimental plants were also carried out. The results showed that *Acanthus montanus* powder was the most potent with 65% adult mortality after 24 h of treatment. This is followed by *Argyreia nervosa* powder that evoked 52.5% weevil mortality. The least toxic to *S. zeamais* was *Acanthospermum hispidum* powder with 32.5% adult mortality. Extracts were more toxic than the powders of the tested plants. *Acanthus montanus* extract was the most toxic since it promoted 80% adult mortality after 24 h of treatment. *Acanthus montanus*, *Alchornea laxiflora* and *Argyreia nervosa* leaf powders and extracts completely prevented oviposition by adult insect, adult emergence, weight loss and seeds damaged. The phytochemicals present in *Acanthus montanus* were alkaloids (3.67 mg/g), saponin (3.33 mg/g), tannin (3.00 mg/g) and flavonoid (2.67 mg/g) contents. *Acanthospermum hispidum* had the least alkaloid (2.67 mg/g), saponin (1.67 mg/g), tannin (1.33 mg/g) and flavonoid (1.00 mg/g) contents. *Acanthus montanus*, *Argyreia nervosa*, *Alchornea laxiflora* and *Acanthospermum hispidum* were efficacious against *S. zeamais* instead of synthetic chemical insecticides that have environmental health hazards and they can be used in integrated pest management by farmers and foods merchants.

**1. Introduction**

Maize is one of the major cereal grains cultivated in abundance during the raining season in West Africa most especially in Nigeria. It ranked fourth most edible grain after sorghum, millet and rice (FAO, 2019). Maize accounted for 19.5% calorie being the world’s highest supplier of calorie for body growth, followed by rice (16.5%) and wheat which accounted for 15.0% (FAO, 2019). Peasant farmers produce huge tonnes of maize annually which is usually more than enough for sale in the markets. This has resulted into wastage due to inadequate storage structures and insect pest attack such as *Sitophilus* species.

The maize weevil (*S. zeamais*) is a field-to-store pest of maize grains in the world (Adedire, 2001). Post-harvest losses to *S. zeamais* have been acknowledged as an increasingly important problem to food security in Africa (Abebe et al., 2009; Markham et al., 1994; Tefera et al., 2011a). Generally, postharvest losses in maize grains due to maize weevil range between 20 and 30% weight losses during storage for three months on farm in Kenya (Boxall, 2002). It has been reported to cause both qualitative and quantitative damages to stored products which could account for grain weight loss of about 20–90% for untreated stored maize in Cameroon (Nukenine et al., 2002; Muzemu et al., 2013). Losses of 45–50% in maize grains were recorded during storage in an attempt to increase the supply of the grains in rural and urban household (Makundi, 2006; Taylor-Davis and Stone, 2007). Maize weevil caused 60% weight losses and quality in terms of nutritional values in maize within 3–6
months in storage which directly affect food security in developing countries such as Nigeria (Adesina, 2012; Ileke et al., 2016). Often times, these damages result to reduced nutritional value and weight loss, low seed germination and ultimately low market value (Tefera et al., 2011b; Napoleao et al., 2013). The larvae and the adult stages of maize weevils are notorious for causing serious damages just like other food storage insect pests that belong to the order Coleoptera (Adedire et al., 2011).

Synthetic chemical insecticides have been used for many years to curtail the menace caused by S. zeamais and other stored product pests most especially under large scale production (Gbeye and Holloway, 2011). But the effectiveness of these insecticides is limited, due to high cost of procurement of the chemicals, toxic residue buildup in foods, and development of resistance by the pests, destruction of natural enemies and also harmful to non-targeted organisms (Oni and Ileke, 2008). A modern trend aimed at alleviating the problems associated with the use of synthetic chemical insecticide is focusing research in the area of the efficacy of plant materials, such as plant powders, plant extracts and plant oils to ascertain their insecticidal properties (Adedire et al., 2011; Ileke et al., 2016). This is because findings have shown that the use of botanicals have little or no effects as compared to the problems posed to the plant using synthetic chemical insecticides. Some plants acts as natural cereals and legumes protectants against insect pests by inhibiting reproduction while others acts as growth or development inhibitors, toxicants, repellent and antifeedants (Trivedi et al., 2018).

Also, the use of botanical products in form of powders and extracts to control stored product weevils and beetles is more convenient by farmers, the powders and extracts are easy to apply by peasant farmers in the fields. A daily measure of reducing the infestations of stored product pests especially the maize weevil (Ileke and Oni, 2011; Karunakaran and Arulnandhy, 2016). But the effectiveness of these insecticides is limited, due to high cost of procurement of the chemicals, toxic residue buildup in foods, and development of resistance by the pests, destruction of natural enemies and also harmful to non-targeted organisms (Oni and Ileke, 2008). A modern trend aimed at alleviating the problems associated with the use of synthetic chemical insecticide is focusing research in the area of the efficacy of plant materials, such as plant powders, plant extracts and plant oils to ascertain their insecticidal properties (Adedire et al., 2011; Ileke et al., 2016). This is because findings have shown that the use of botanicals have little or no effects as compared to the problems posed to the plant using synthetic chemical insecticides. Some plants acts as natural cereals and legumes protectants against insect pests by inhibiting reproduction while others acts as growth or development inhibitors, toxicants, repellent and antifeedants (Trivedi et al., 2018).

Also, the use of botanical products in form of powders and extracts to control stored product weevils and beetles is more convenient by farmers, the powders and extracts are easy to apply by peasant farmers and the produces remain fresh, clean and attractive to buyers after the treatment (Ojo and Ogunleye, 2013). Natural plant products have been found to be cheap, humanly safe and ecologically tolerant to control measures of reducing the infestations of stored product pests especially in the tropics (Lale, 1992; Adedire and Ajayi, 1996). It has been discovered that many of the botanicals used as crop protectants in the control are safe for human consumption (Omotoso, 2015). Plants such as Azadirachta indica, Alstonia boonei, Garcinia kola, Morinda oleifera, Nicotiana tabacum, Lantana camera, Annona squamosa, Justicia adhatoda and Ocimum tenuiflorum have been studied as potential alternatives to control maize weevil (Ileke and Oni, 2011; Karunakaran and Arulnandhy, 2016).

Acanthus montanus, a quick-growing evergreen herb is referred to as mountain thistle or alligator plant by many people. It is a perennial herbaceous plant belonging to the family Acanthaceae. Gboghe (2010) and Nnamani et al. (2015) reported that the plant is one of the threatened and underutilized (in terms of insecticidal activities) vegetables species in Africa which may be due to the fact that it is highly perishable. Acanthospermum hispidum is an annual herb that can grow up to 90 cm high and has a tap root that is branched and shallow. A plant that is very treasured for its ethnobotanical importance. It is used for treating of yellow fever, malaria and stomach disorder (Denis, 2002; Mann et al., 2003; Gafon et al., 2012). Argyreia nervosa plant starts out as a bush in the first one to two years before growing into a vine of up to 10 m in length (Ashutosh et al., 2011). The stems woody at the base which is densely white pubescent when young, and glabrescent (CABI, 2018). Argyreia nervosa is quite popular in India as it is a very important medicinal plant, which has long been used in their traditional Ayurvedic medicine to treat and manage several ailments. Virtually all the plant parts are considered to be useful which includes the roots, leaves, flowers and seeds (Galani et al., 2010; Ashutosh et al., 2011; PIER, 2016). Alchornea laxiflora is a deciduous shrub or small tree that has a smooth bark which is pale grey and flaking. The leaves are alternate, elliptic-lanceolate to oblong-ob lanceolate measuring up to 17 cm in length and 8 cm in width possessing 3-veins from the base that are thinly textured (Hyde et al., 2019).

The insecticidal properties of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora as protectants of maize against maize weevil, S. zeamais is scarce literatures. Therefore, the specific objectives of this research study are to evaluate bioinsecticidal properties of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves powders and extracts as an ecofriendly protectants against adult maize weevil, S. zeamais in stored maize, and to evaluate the phytochemical compositions of the experimental plants extracts.

2. Materials and methods

2.1. Insect rearing

Adults maize weevil, S. zeamais were supplied by Storage Entomology Research Laboratory, Department of Biology, Federal University of Technology, Akure (FUTA), Nigeria. Fifty pairs of the weevils were introduced into 1 L glass kilner jar containing 600 g of maize grains obtained from a grain Merchant shop within Akure metropolis, Akure, Ondo State, Nigeria. The weevil colony was maintained under a constant insectarium condition of 28±2°C and 75 ± 5% relative humidity.

2.2. Identification and sexing of adult of S. zeamais

The identification and sexing of S. zeamais were carried out in the Entomology Research laboratory, Department of Biology, FUTA. The weevils were identified to species according to features of genital morphology (Halstead, 1963; Appert, 1987; Odeyemi and Daramola, 2000), antennae Odeyemi and Daramola, 2000), reddish-brown or orange-brown oval markings on the elytra, and circular punctures also present on the prothorax (Odeyemi and Daramola, 2000). Then, adults were sexed according to the length of the rostrum (the female has a comparatively longer rostrum than the male).

2.3. Collection and preparation of plant powders

The leaves of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora were sourced fresh from a farmland at Iresi, Akure South Local Government Area of Ondo State, Nigeria. These leaves were first of all air dried naturally in the laboratory. The dried leaves were later pulv erized separately into fine powder with the aid of an electric blender, JTC Omni Blender V (Model TM-800). The fine powders were allowed to pass through a nylon mesh of 1 mm dimension. The powders were then packed into an air tight containers and put in a refrigerator at 4°C to retain its good quality before application.

2.4. Collection of maize grains

The maize grains used for this research work were obtained from newly harvested stock of maize grains free of insecticides at the Ministry of Agriculture, Agricultural Development Programme, Akure, Ondo State, Nigeria. The clean grains were first of all sterilized by putting them in a deep freezer and maintained at 5 °C for 72 h to ensure that all existing insect eggs and larvae are killed. This process is carried out because all the life stages of insects such as eggs and larvae are sensitive to low temperature (Koebler, 2003). The disinfested maize grains were later air dried naturally in the laboratory for 72 h to ensure that the
grains do not become moldy (Adedire et al., 2011).

2.5. Preparation of ethanolic extracts

The leaves of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves were extracted using absolute ethanol as solvent. About 300g of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaf powders were soaked separately in an extraction bottle containing 600 ml of absolute ethanol. The mixture was stirred at an interval of 6 h with a glass rod and extraction was terminated after 3 days. The resulting mixture was filtered using a double layer of Whatman No. 1 filter paper and the solvent was regained by redistilling in a rotary evaporator at 30-40 °C with rotary speed of 3-6 rpm for 8 h (Udo et al., 2011). The resulting materials were air dried in order to remove traces of solvents (ethanol). The crude extracts were then kept in a dark bottle and labeled separately inside a refrigerator to maintain its quality. From this stock solution, 3% concentration was prepared by diluting 0.3 ml of extract in 9.7 ml of solvent (Ashamo and Akinnawonu, 2012; Ileke et al., 2013).

2.6. Phytochemical screening of the plants

Chemical tests were carried out on the ethanolic extracts of the leaves of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora for the qualitative and quantitative determination of phytochemical constituents using standard procedures as described by Harborne (1973); Trease and Evans (1985); Sofowora (1993), Ejikeme et al. (2014), Ezeonu and Ejikeme (2016).

2.7. Qualitative analysis

2.7.1. Saponin determination

Analytical qualitative determination of saponin was carried out using the methods described by Harborne (1973); Ejikeme et al. (2014); Ezeonu and Ejikeme (2016). Fifty centimeter cube (50 cm³) of distilled water was added to 0.5 g each of the experimental plant powders in a beaker on a steam water bath for 10 min before filtration using Whatman No 1 filter paper. 5 cm³ of a mixture of distilled water and 10 cm³ of filtrate was agitated vigorously to form a stable persistent lather. Three drops of olive oil formed an emulsion was taken as presence of saponin.

2.7.2. Tannin determination

Tannin qualitative determination was carried out using the methods described by Sofowora (1993); Ezeonu and Ejikeme (2016). Fifty centimeter cube (50 cm³) of distilled water was added to 0.5 g each of the experimental plant powders in a beaker on a steam water bath for 10 min before filtration using Whatman No 1 filter paper. To the filtrate (5 cm³), 3 drops of 0.1% ferric chloride was added, and a brownish green colouration was formed confirming the presence of tannin.

2.7.3. Alkaloid determination

Alkaloid qualitative determination was carried out using the methods reported by Harborn (1973); Trease and Evans (1985); Hijikado et al. (1984). Each of the experimental plant extract (0.5g) was added to 1% aqueous hydrochloric acid (5 ml) on a steam water bath and stirred regularly before filtration. To the filtrate, few drops of Dragendorf reagent was added to 1 ml of the filtrate. A blue black turbidity was observed and this was taken as maiden confirmation for the presence of alkaloid.

2.7.4. Phlobatannin determination

Phlobatannin qualitative determination methodology used in this research was that by Ejikeme et al. (2014) as reported by Ezeonu and Ejikeme (2016). Fifty centimeter cube (50 cm³) of distilled water was added to 0.5 g each of the experimental plant powders in a beaker and the mixture was allowed to extract for 24 h. This was followed by boiling 10 cm³ of each extract with 5 cm³ of 1% aqueous hydrochloric acid and deposit of red precipitate showed the presence of Phlobatannin.

2.6.4. Anthraquinone determination

Analytical method was according to Trease and Evans (1985); Sofowora (1993); Amadi et al. (2004). Bornträger’s test was adopted for the detection of Anthraquinone. About 0.5 g of each of the experimental plant extracts was measured into a test tube and 10 ml of benzene was added, shake gently before filtration. To the filtrate, 5 ml of 10% ammonia (NH₄OH) solution was added to 10 cm³ of the filtrate. The mixture was thoroughly shaken and the presence of pink red or violet colour in the ammonia layer shows the presence of free anthraquinone.

2.7.5. Flavonoid determination

Flavonoid qualitative determination was by the method reported by Sofowara (1993); Amadi et al. (2004); Ejikeme et al. (2014); Ezeonu and Ejikeme (2016). Fifty centimeter cube (50 cm³) of distilled water was added to 0.5 g each of the experimental plant powders in a beaker and the mixture was allowed to extract for 2 h before filtration with Whatman No 1 filter paper. To the filtrate, 10 cm³ of each experimental plant extract was added to 5 cm³ of 1.0 M dilute ammonia (NH₄OH) solution. This was followed by the addition of 5 cm³ of concentrated tetraoxosulphate (VI) acid (H₂SO₄) that formed a yellow colouration that disappeared on standing indicate the presence of flavonoid (Ejikeme et al., 2014; Ezeonu and Ejikeme, 2016).

2.8. Quantitative analysis

2.8.1. Determination of saponin

Saponin quantitative determination was by methods described by Sofowara (1993); Obadoni and Ochuko (2002); Ejikeme et al. (2014). Five grams (5g) of each experimental plant was put into 250 cm³ conical flask that contain 20% ethanol. The content was heated with continuous stirring over a hot water at a temperature of 55 °C for 4 h. The residue obtained was re-extracted after filtration and heated with continuous stirring over a hot water at a constant temperature for 4 h. Combined extract was evaporated to 40 cm³ over water bath at 90°C. To the concentrate, 20 cm³ of Diethyl ether was added in a separator funnel of about 250 cm³, strongly agitated to recover the aqueous layer and ether layer was discarded. The purification procedure was repeated two times. N-butanol (60 cm³) was added to 5% sodium chloride (10 cm³) and extracted two times. The sodium chloride layer was discarded while the remaining solution was heated in a water bath for 30 min. The solution was transferred into crucible before oven dried to a constant. The saponin content was expressed in percentage as follows:

\[
\text{% Saponin} = \frac{\text{Weight of saponin}}{\text{Weight of sample}} \times 100
\]

2.8.2. Determination of alkaloid

Analytical quantitative determination of alkaloid was by methods described by Harborne (1973); Ezeonu and Ejikeme (2016). Five grams (2.5 g) of each of the experimental plant powders was weighed into an extraction bottle, followed by addition of 200 ml of 10% acetic acid in ethanol and the mixture was allowed to stand for 4 h before filtration with Whatman No 1 filter paper and extract was concentrated to one quarter of its original volume on a water bath. 15 drops of concentrated ammonium hydroxide was added dropwise to the extract until the precipitation was completed after filtration process. The mixture was allowed to settle for 3 h and the supernatant was discarded and precipitate was washed with 20 cm³ of 0.1M of ammonium hydroxide and before filtration. The residue was oven dried and weighed (Harborne, 1973) and % alkaloid is expressed as

\[
\text{%Alkaloid} = \frac{\text{Weight of alkaloid}}{\text{Weight of sample}} \times 100
\]
2.8.3. Flavonoid determination

Analytical determination of flavonoid was by the methods reported by Ejikeme et al. (2014) and Boham and Kocipai (1994). Fifty (50 cm²) of ethanol was added to 2.5 g of the sample in a 250 cm³ beaker with lid and allowed to stand for 24 h. The supernatant was discarded followed by the re-extraction of the residue for three times. No 1 Whatman was used in the filtration of all the experimental plants. Each of the experimental plants filtrate was transferred into a crucible and allowed to evaporate to dryness over a water bath. The remaining content in the crucible was allowed to cool in a desiccator and weighed until constant weight was achieved (Ezeonu and Ejikeme, 2016).

\[
\text{%Flavonoid} = \frac{\text{Weight of flavonoid}}{\text{Weight of sample}} \times 100
\]  

(3)

2.8.4. Determination of tannin

Tannin quantitative determination was by the method reported by Sofowora, Amadi et al. (2004); and Ejikeme et al. (2014). An insoluble polyvinyl-polypyrrolidone (PVPP) that binds tannins was used for the determination of tannin content. 1 mg/ml of each of the experimental plants was prepared in ethanol. This followed the determination of total phenolics through the mixing of 1 ml of each experimental plant extract with 100 mg of polyvinyl-polypyrrolidone, vortexed then centrifuged at 3000 rpm for 10 min. A pure supernatant non-tannin phenolics were determined using the methods of finding total phenolics. Tannin content was evaluated as a difference between total phenolic and non-tannin content (Ezeonu and Ejikeme, 2016).

2.9. Insect bioassay

2.9.1. Toxicities of plants powders on adult mortality and adult emergence of S. zeamais

Twenty grams (20 g) of clean uninfested maize grains was weighed into 250 ml of plastic cups. Then, an aliquot of 1.0 ml of 3% extracts of the leaf Acanthus montanus was measured with the aid of an electronic weighing balance (Model JTC 2101N) in the laboratory and put inside plastic cups (250 ml). Thereafter, 0.3 g dosage of the leaf powder of Acanthus montanus was carefully measured and admixed with 20 g of the clean uninfested maize grains. The plastic cups containing the powder and the maize grains were thoroughly shaken to ensure adequate mixing. Then, ten copulating pairs (10 males: 10 females) of newly emerged (less than four days old) adults of S. zeamais were introduced into each of the plastic cups containing the treated maize grains and covered with muslin cloths. The same procedure was used in determining the contact toxicity of other plant extracts using the same concentration of 3%. The control experiment had only 20 g of maize grains and ten copulating pairs of adult S. zeamais (no plant powder was included in the control). Insect mortality was assessed every day for 5 days (120 h). Dead weevils were those that did not move and did not respond to pin probing (response to sharp pin). At the end of 120 h post treatment, data on percentage adult mortality was calculated using Abbott (1925) formula as described above. The total number of eggs laid by the female S. zeamais was also determined after 120 h of exposure. This was achieved by identifying the egg plugs of S. zeamais after staining with acid fuchsin dye solution (Frankenfeld, 1948). The experimental setup was kept inside the insect rearing cage for further 35 days to allow the new adults to emerge. Percentage Adult emergence, weight loss, seeds damage and Weevil Perforation Index were calculated as described above.

3. Data analysis

Data were subjected to analysis of variance (ANOVA) and treatment means were separated using New Duncan’s Multiple Range Test (NDMRT). The ANOVA was performed with SPSS 25.0 software (SPSS, 2017).

4. Results

4.1. Phytochemicals screening of ethanolic extracts of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves

Phytochemical screening of ethanolic extracts of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves is presented in Table 1. The phytochemicals present in the ethanolic extracts of Acanthus montanus, Acanthospermum hispidum, Argyreia
least toxic to trend of results were also recorded after day 2, day 3, day 4 and day 5 of caused 32.5% mortality of adult weevil after 24 h of treatment. Similar Argyreia nervosa mortality of adult leaf powder was the most potent at the test rate which caused 65%
The weevil mortality ranges from 32.5% to 100%. 0.05) reduced adult maize weevils compare to untreated maize grains. Argyreia nervosa obtained showed that of maize grains on adult Argyreia nervosa and Alchornea laxiflora leaves. {Alchornea laxiflora}

4.2. Toxicity of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves Powders to adult mortality of S. zeamais

Toxicity of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaf powders at concentration 3.0g/20g of maize grains on adult S. zeamais is presented in Fig. 2. The results obtained showed that Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaf powders significantly (p < 0.05) reduced adult maize weevils compare to untreated maize grains. The weevil mortality ranges from 32.5% to 100%. Acanthus montanus leaf powder was the most potent at the test rate which caused 65% mortality of adult S. zeamais after 24 h of treatment. This is followed by Argyreia nervosa leaf powder that evoked 52.5% weevil mortality. The least toxic to S. zeamais was Acanthospermum hispidum powder that caused 32.5% mortality of adult weevil after 24 h of treatment. Similar trend of results were also recorded after day 2, day 3, day 4 and day 5 of treatment. It was only Acanthus montanus leaf powder that caused 100% mortality of weevil after 5 days of treatment. The results showed that adult weevil mortality increased with an increase in exposure period and concentration dependent. There was no significant different (p > 0.05) in the grains treated with Acanthus montanus leaf powder compare with Argyreia nervosa powder but Acanthus montanus was higher in term of percentage.

4.3. Effect of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves Powders on oviposition and Emergence of S. zeamais

The values of eggs laid are shown in Table 2. Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora powders successfully inhibited egg laying potential of maize weevil, S. zeamais. The number of eggs laid by S. zeamais on treated maize grains was significantly lower (p < 0.05) than untreated seeds. There was no significant difference (p > 0.05) in the mean number of eggs laid on the treated seeds with Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora. On Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora powders, the numbers of egg laid were 0.00, 6.00, 1.00 and 3.00, respectively compared to untreated that recorded 42.50 eggs. There was no sign of egg laid and adult emergence in the maize grains treated with Acanthus montanus powders. The percentage adult emergence in the untreated cowpea seeds was significantly different (p < 0.05) from oviposition and emergence in the treated cowpea seeds. There was no sign of progeny development in maize grains treated with Acanthus montanus, Argyreia nervosa and Alchornea laxiflora leaf powders. It was only Acanthospermum hispidum powder that had 15.4% adult emergence which significantly different from untreated that had 70% adult emergence.

4.4. Grain damage, Weight loss and Weevil Perforation index assessment of Maize seeds Treated with plant leaves powders

Acanthus montanus, Argyreia nervosa and Alchornea laxiflora completely prevented infestation and damage of the treated maize grains (Table 3). There was neither seed damage nor weight loss recorded in the treated maize grains and Weevil Perforation Index was zero for the concentration tested except in the treated seeds with 3g of Acanthus montanus, Argyreia nervosa and Alchornea laxiflora after 5 days of application. However, the WPI of 3.50 obtained for 3g of Acanthospermum hispidum, was significantly different from WPI of the untreated. In the untreated cowpea seeds, 65% damage occurred as revealed by

| Phytochemicals | Acanthus montanus extract | Acanthospermum hispidum extract | Argyreia nervosa extract | Alchornea laxiflora extract |
|----------------|---------------------------|-------------------------------|------------------------|---------------------------|
| Alkaloids      | +                         | +                             | -                      | +                         |
| Saponins       | +                         | +                             | +                      | -                         |
| Tannins        | +                         | -                             | +                      | +                         |
| Flavonoids     | -                         | -                             | -                      | -                         |

Keys: - Absent; + Present.

![Fig. 1. Quantitative analysis of Phytochemicals in ethanolic extracts of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves. (Means with different alphabets are significantly different (p > 0.05) using NDMRT).](image-url)
Each value is the mean ± standard error, n = 4 replicates. Letters as in Table 2, mean followed by the same letters within the same column are not significantly different (p > 0.05) using NDMRT.

4.5. Toxicity of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves Extracts to adult mortality of S. zeamais

Fig. 3 presented the contact toxicity of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaf extracts at concentration 3%/20g of maize grains on adult S. zeamais. The results obtained showed that Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaf extract significantly (p < 0.05) reduced adult maize weevils compare to untreated maize grains. The weevil mortality ranges from 50% to 100%. Acanthus montanus leaf extract was the most potent at the test rate which caused 80% mortality of adult S. zeamais after 24 h of treatment. This followed by Argyreia nervosa leaf extract that evoked 67.5% weevil mortality. The least toxic to S. zeamais was Acanthospermum hispidum extract that caused 50% mortality of adult weevil after 24 h of treatment. Similar trend of results were also recorded after day 2, day 3, day 4 and day 5 of treatment. It was only Acanthus montanus leaf extract that caused 100% mortality of weevil after 2 days of application. The results showed that adult weevil mortality increased with an increase in exposure period and concentration dependent. There was no significant different (p > 0.05) in the grains treated with Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaf powders after 4 days of application.

4.6. Effect of Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves Extracts on oviposition and Emergence of S. zeamais

The values of eggs laid are shown in Table 4. Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora extracts successfully inhibited egg laying potential of maize weevil, S. zeamais. The number of eggs laid by S. zeamais on treated maize grains was significantly lower (p < 0.05) than untreated seeds. There was no emergent holes of the weevils. There was neither seed damage nor weight loss recorded in the cowpea seeds treated at rate 3.0g Acanthus montanus, Argyreia nervosa and Alchornea laxiflora.
significant difference (p > 0.05) in the mean number of eggs laid on the treated seeds with Acanthus montanus, Argyreia nervosa and Alchornea laxiflora extracts, the numbers of egg laid were 0.00, 2.00, 0.00 and 0.00, respectively compared to untreated that recorded 40 eggs. There was no sign of egg laid and adult emergence in the maize grains treated with Acanthus montanus powder. The percentage adult emergence in the untreated cowpea seeds was significantly different (p < 0.05) from oviposition and emergence in the treated cowpea seeds. There was no sign of progeny development in maize grains treated with Acanthus montanus, Argyreia nervosa and Alchornea laxiflora, Argyreia nervosa and Alchornea laxiflora leaf powders compared with untreated that had 77.5% adult emergence.

### 4.7. Grain damage, Weight loss and Weevil Perforation index assessment of Maize seeds Treated with plant leaves Extracts

Acanthus montanus, Acanthospermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves extracts completely prevented infestation and damage of the treated maize grains (Table 5). There was neither seed damage nor weight loss recorded in the treated maize grains and Weevil Perforation Index was zero for the concentration tested after 5 days of application. In the untreated cowpea seeds, 62.5% damage occurred as revealed by emergent holes of the weevils. There was neither seed damage nor weight loss recorded in the cowpea seeds treated at rate 3.0g Acanthus montanus, Argyreia nervosa and Alchornea laxiflora.

### 5. Discussion

Plants powders, extracts or essential oils consists of complex mixtures of monoterpenes, sesquiterpenes and biogenetically related phenols (Trivedi et al., 2018). The mode of action of these complex mixtures present in plants powders, extracts or oils against insect pests of stored...
produces is through neurotoxic mode of action (Trivedi et al., 2018). Researches have revealed that the complex mixtures in plant powders, extracts or oils inhibited acetyl cholinesterase enzyme (AChE) action (Houghton et al., 2006). The inhibited acetyl cholinesterase enzyme (AChE) activity interferes with the neuromodulator octopamine (Kostyukovsky et al., 2002; Trivedi et al., 2018), it can also block GABA-gated chloride channels of the insect pest resulting to their death (Priestley et al., 2003). Monoterpenes can act on other susceptible sites such as cytochrome P450-dependent mono-oxygenases which may also lead to the destruction of pests (Lee et al., 2001).

The insecticidal screening test carried out in this research work has revealed that all the plant powders and extracts tested for their insecticidal properties were highly toxic to S. zeamais when compared with the control experiment. Plant powders and extracts have been used to suppress the population of storage insect pests (Ogunleye et al., 2004; Ojo and Ogunleye, 2013). The present study has shown that the leaves of Acanthus montanus (powders and extracts) were highly toxic to S. zeamais. This level of effectiveness was followed by the leaf powder and extracts of Argyreia nervosa. All the other powders and extracts proved to be moderately toxic to S. zeamais. The toxicities of these powders and extracts in this present study varied with plant species and period of exposure. The present study also revealed that the leaf extracts were generally more toxic to the insects than the leaf powders as used in contact toxicities. This difference in effectiveness could be linked to the fact that the active components of the plant materials have been concentrated via the extraction process. The potency of insecticidal plant powders can be increased through extraction using appropriate solvent as suggested by Makanjuola (1989) and Ogunleye (2000). Lale (1995) reported that plant extracts are highly lipotropic and have the ability of penetrating into the cuticle of insects. According to Asawalam et al. (2007), insecticidal activity of any plant extract depends on the active constituents of the plant extract. The active compounds form a complex mixtures in plant powders, extracts or oils such as monoterpenes, sesquiterpenes and biogenetically related phenols which inhibit acetyl cholinesterase enzyme (AChE) activity (Houghton et al., 2006) that interferes with the neuromodulator octopamine leading to the death of insect pests (Kostyukovsky et al., 2002).

The high mortality could be linked to the feeding habits of the insect. The powders and extracts covered the tests of the treated grains thereby serving as food poison to the adult weevil (Ogunleye, 2011). Also, the insects must have been deprived from obtaining enough nourishment from the maize grains which would have supported its normal growth and development as reported by Ogunleye (2011) and this eventually led to insect mortality. The results obtained in this research work agreed with the findings of Mbara and Ekpendu (1992) who reported that 0.4 g/5.0 g of powdered seeds of P. guineense when admixed with maize resulted in 50% mortality of adult S. zeamais. Also, Ileke and Oni (2011) observed that the plant powders of Azadirachta indica and Alstonia boonei applied at 2.5% w/w, 5.0% w/w, 12.5% w/w and 25.0% w/w on wheat grains gave high percentage mortality on S. zeamais after 72 h of exposure. Similarly, the seed powder and oil of black pepper, Piper nigrum, Piper guineense, Piper umbellatum and Capsicum frutescens are known to adversely affect the biology of the maize weevil, S. zeamais and also cause high percentage mortality (Lajide et al., 1998). The high percentage mortality of S. zeamais exposed to the leaf powders and extracts of tested plants could also be due to contact toxicity. Most insects breathe by means of trachea which usually opens at the surface of the body through spiracles. These openings or air chambers might have been hindered from receiving enough oxygen into the body of the insects which eventually led to their asphyxiation and death as suggested by Adedire et al. (2011).

The present research has also shown that all the plant powders and extracts used as crop protectants were capable of preventing oviposition, adult emergence, seed damage and weight loss except the leaf powder and extracts of Acanthusperum hispidum where only few eggs were laid. The ability of some plant powders and extracts to reduce or prevent oviposition by female Coleopteran pests and mortality of the developmental stages have been studied by a number of authors and well documented (Adedire et al., 2011; Ojo and Ogunleye, 2013). The effects of the plant powders and extracts on oviposition by S. zeamais could be due to metabolic alteration and consequently other systems of the body of the insects (Ileke, 2014). The number of adult emergence observed in the treated maize grains could be due to high insect mortality. Also, the few eggs that were laid on maize grains exposed to the leaf powder of Argyreia nervosa and Alchornea laxiflora could not develop into adults as the leaf powders must have passed through the chorion of the eggs and thereby disrupt normal developmental stages from eggs to adults (Ileke and Olotua, 2012). All the leaf powders and extracts evaluated for insecticidal properties in this research work significantly reduced seed damage and weight loss caused by S. zeamais. In this regard, the leaf extracts appeared to be more toxic than leaf powders since there was no seed damage and weight loss recorded in the maize grains treated with the leaf extracts and the weevil perforation index (WPI) was zero for all the extracts at the concentration tested. The ability of the plant powders and extracts to completely prevented seed damage and weight loss could be due to high insect mortality. It could also be due to the fact that the insects could not lay eggs on the treated grains which could have led to larval feeding and consequently prevented seed damage and weight loss as suggested by Alabi and Adewole (2017).

The result obtained on the phytochemical constituents of the ethanolic extracts of the tested plants revealed the presence of alkaloids, saponins, tannins, flavonoids, and cardiac glycosides. Secondary metabolites such as phenolic compounds, saponins, alkaloids, flavonoids, and terpenoids have been identified to exhibit strong activities against several pathogens and insect pests (De Geyter et al., 2007). Acanthus montanus was the most toxic to maize weevil, it had the highest composition of alkaloid, saponin, tannin and flavonoid compared to other experimental plants. Saponin can acts as an anti-inflammatory, anti-viral, antifungal, insecticidal, molluscicidal, piscidical and anti-bacterial activity (Ngoci et al., 2011). The toxicity and antifeedant effect of alkaloids towards stored products insect pest has been reported (Yang et al., 2006). The insecticidal activity of the saponin has been studied by Chaieb (2010). The observed insecticidal activities of the tested plants could be due to the presence of these active ingredients present in the extracts of the tested plant materials which might have interferes with the neuromodulator octopamine leading to the death of insect pests (Kostyukovsky et al., 2002; Trivedi et al., 2018).

6. Conclusion

Acanthus montanus, Acanthuspermum hispidum, Argyreia nervosa and Alchornea laxiflora leaves species are found to have potent insecticidal activity toward maize weevil instead of synthetic chemical insecticides that causes environmental health hazards and lethal dose to the users. They present antifeedant characteristics, oviposition deterrent and affected significantly larval growth of S. zeamais. The use of Acanthus montanus, Argyreia nervosa and Alchornea laxiflora leaves as bio-insecticides in the control of maize weevil in stored maize seeds among poor resource farmers and food merchants should be advocated since the plant is ecofriendly and readily available and used among rural peoples for its ethno medical importance.

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Kayode David Ileke: Conceptualization, Data curation, Methodology, Formal analysis, Writing - review & editing. Joy Ejemen Iloko: Methodology, Formal analysis, Writing - original draft. Durojaye Olanrewaju Ojo: Data curation, Investigation. Bukola Christianah Adesina: Data curation, Investigation.

Declaration of competing interest
The authors declare that they have no competing interests.

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Appendix A. Supplementary data
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