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# BIO-POTENCY EVALUATION OF MIXTURE OF *ACACIA POLYACANTHA* AND *HYMENOCARDIA ACIDA* WOOD ASH AGAINST *SITOPHILUS ZE*AMAIS (MAIZE WEEVIL) IN LAFIA, NASARAWA STATE-NIGERIA

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**ABSTRACT**
The maize weevil, *Sitophilus zeamais* is an insect pest, which has spread all over the world, and causes damage to maize grains and other species of stored cereal. The aim of this study was to investigate the insecticidal efficacy and repellency of mixture of *Hymenocardia acida and Acacia polyacantha* wood ash against the weevil on a 14 day exposure and trial. The study involved the use of three (3) replicates of the mixture of ashes at treatment doses of 5g, 10g and 20g while 0.5g, 1g, and 1.5g was used for diatomaceous earth (Positive Control). Results showed that mortality was dose dependent, and there was also a significant difference in mortality rate of the weevil across all treatments ($P < 0.05$). The study revealed 100% mortality within the study period similar to the positive control. The wood ash mixture of HA and AP recorded highest percentage progeny inhibition of 100% compared to control (44.44%). The study also demonstrated efficacy in terms of grain damage (1.33%) and weight loss (0.92%) which showed a significant difference compared to the control (2.67% damage and 9.09% weight loss). A highest repellency of 70cm of the mixture of ashes was observed in the treatment with 30% aqueous solution compared to control powder (60cm) which showed a significant difference in repellent properties between different treatments concentrations used. The study recommends that mixture of wood ashes from *H. acida and A. polyacantha* could be used to reduce grain infestation as stored grain protectants against *S. zeamais.*

**Keywords:** *Sitophilus zeamais*, mixture, wood ash, *Acacia polyacantha, Hymenocardia acida,* bio-potency, evaluation.

## Introduction

The primary cause of the major issues with maize storage is insect infestations (Kebede et al., 2024). The maize weevil, *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae), is a pest bug that has spread throughout the world and damages stored cereals, including maize grains. While an infestation may start in the field, most damage occurs in storage because both adults and larvae consume grains internally (Jean et al., 2015), Suleiman et al., 2015). Although losses are typically 4–5%, severe infestations of these pests in stored maize can result in weight losses of 15–30% in developing nations (Bergvinson, 2021). One of the most significant maize pests in Europe has been identified as *S. zeamais* (Trematerra et al., 1999), Asia (Peng, 1998) and USA (Sedlacek et al. 1998; Arbogast & Throne, 1997).

In the past, it was controlled with diclodiphenyltrichloroethane (DDT), malathion, pirimiphos-methyl, pyrethroids, and other chemicals (Corrŭa et al., 2011), which often led to resistance and other negative effects (Jean et al., 2015).

Research on alternative suppression techniques has focused on the use of various inert dusts, such as diatomaceous earth (Athanassiou et al., 2011) and various essential oils (Tofel et al., 2016). Inert dusts are any dry dusts that do not react chemically in the environment.

Liška et al. (2017) and Korunić (1998) divided them into a number of groups based on their level of activity, chemical composition, and physical makeup. These groups included non-silica dusts, groups that contained sand, kaolin, and wood ash, diatomaceous earths (or diatomite) primarily derived from diatomaceous earths and amorphous hydrated silica, with high-quality silica aerogels and synthetic silicates. These include wood ash and kaolin, which are commonly utilised locally (on small farms) and in less developed nations. According to Panagiotakopulu et al. (1995), the latter is among the oldest insecticides. Its significance has grown recently, especially in light of the issues surrounding synthetic pesticides (Guedes et al., 2017; Tofel et al., 2016).

According to a Slovenian study, common beech ash exerts a strong insecticidal effect on the granary weevil, *Sitophilus granarius* (L.) (Coleoptera: Curculinonidae)(Bohinc & Tradan, 2017). The study found that lower relative humidity levels had an adverse effect on both progeny production and death following exposure intervals. Applying wood ash alone and expanding its use with other items showed the high efficacy of non-chemical methods (Marina et al., 2023). Akob and Ewete (2007) and Jean et al. (2015) have already shown how to apply wood ash alone.

The aim of this study is to determine the effectiveness of wood ash from two native tree species, *Acacia polyacantha* (Mimosaceae) and *Hymenocardia acida* (Phyllanthaceae), against *S. zeamais* on maize in order to develop a sustainable weevil management approach. The efficacy was assessed using the following metrics: adult mortality, offspring inhibition, population suppression of *S. zeamais*, and the weevil's damage reductions on the treated grains as well as repellency.

## Materials and methods

### Study Area:

The experimental study was conducted into two sites. *In vitro* insecticidal efficacy test was conducted in the Zoology Department, Federal University of Lafia Nasarawa State Nigeria while the proximate analysis of wood ashes was conducted in the Chemistry Department Federal University of Lafia. The study was carried out between the months of July to September.

Lafia is Located within latitude 8.49$^0$N and 8.52$^0$E in Nasarawa State Lafia is bounded to the East by Nasarawa Egon and Kokona local government areas, on the West by Pleateau State, to the North by Wamba Local government area and to the South by Doma and Obi local government areas. The climatic condition of lafia is typical of the Tropical Guinea Savanna Belt of North Central Nigeria with an annual rainfall that varies between 130cm to 140cm and an average annual temperature of 28.4$^0$C.

**Study design:**

The study adopted adult immersion test *in vitro* which was conducted from July to September 2024.

**Collection and preparation of wood ash of** *Acacia polyacantha* **and** *Hymenocardia cida:*

From the Lafia town area, branches of *A. polyacantha* and *H. acida* were gathered respectively. To minimise contamination, latex gloves were worn during the collecting process. After two weeks of air drying in the sun, the stems were burned separately in a conventional kitchen to produce the necessary weight of ash ( 250g) was put in glass jars and kept at -40°C during the bioassay.

**Diatomaceous earth:**

The DE used as positive control was fossil field 90.0 fine brownish powder containing amorphous silica particles with size range between 5 and 30 mm, a content of 73% amorphous $Si0_2$, a water content of about 2% in addition to 3% aerosol and other mineral compounds (Fossilfield company, Bein GmbH, Ei terfield, Germany).

**Proximate analysis of wood ash collected from** *Acacia polyacantha* **and** *Hymenocardia acida* **trees and diatomaceous earth:**

After the trees' wood ash was gathered, an analysis was conducted. The main criteria evaluated for wood ash were its ash content, moisture content, and volatile matter; for diatomaceous earth, the percentage aerosol, water content, and silica were also determined

**Insect culture and treatment of maize grains:**

The first *S. zeamais* population was acquired from infected maize grains that were bought from Lafia Modern Market. Freshly harvested, un-infested maize varieties were acquired from the College of Agriculture, Lafia, and kept for three hours in ambient laboratory settings (20–290°C, 59–900°R) before being allowed to cool for an additional hour. After being added to the un-infested (sterilised) maize grains and given time to oviposit, the adults of the original population of *S. zeamais* were taken out. A second jar of recently collected maize seeds was filled with the emerging adults (F1).

This is to guarantee that the F1 adults used as the experiment's culturing stock are of the same age and size. According to Masuda et al. (2004), the insect culture was maintained at a temperature of 27 to 29 degrees Celsius and a relative humidity of 70 to 80%. All bioassays using maize as the substrate used live insects. To encourage the initial grain moisture content, the Shaba variety of maize was identified with the Department of Agronomy, Faculty of Agriculture, FULafia, and kept in a freezer at -4c for seven days.

**Insecticidal efficacy test:**

Each of the wood ash duplicates was weighed using 5 g, 10 g, and 20 g of wood ash from the samples, respectively. 50g of maize was placed in glass jars with 0.5g, 1g, and 1.5g of diatomaceous earth (Positive Control) added individually, and 50g of maize grains were mixed with a mixture of wood ashes of equal combinations, and the wood ashes' synergistic effectiveness was evaluated.

**Adult mortality test and F1 progeny production:**

In a completely randomised design, the treatments were placed on a shelf in the lab at room temperature (23.7+1.87°C, 59.0+7.63 r.h.). Within 24 hours and on days 3, 7, and 14 following treatment, insect mortality was noted. To guarantee that the dusts were evenly distributed throughout the grain mass, each jar was shook by hand for five minutes. Diatomaceous earth was used as the positive control, and maize grains devoid of wood ash were used as the negative control (Nukenine et al., 2007).

**Insect population and grain damage:**

The dosages of wood ash and diatomaceous earth used in the previously stated toxicity test were considered for 150g grains. Forty adult insects were placed in each jar of treated or untreated grains. Each procedure was performed three times. After being stored for two weeks, the number of live and dead insects in each jar was determined. Using Adams and Schullen's method, damage was assessed by counting and measuring the perforated and non-perforated grains as well as calculating the percentage of weight loss (Schullen et al., 1978) as follows:

$$\frac{(UNd)-(DNu)}{U\,(Nd+Nu)} \times 100$$

Where;

U= Weight of undamaged grains;

Nu= Number of undamaged grains;

D= Weight of damaged grains;

Nd= Number of damaged grains

**Persistence of wood ash on maize grains:**

Twenty (20) mature *S. zeamais* were exposed to the treated maize (same order as previously mentioned for bioassay above) which were stored for 7 days (Obeng-ofovi et al., 2005) and the persistence of dust on grain was evaluated.

**Repellency test of the wood ash:**

Weevil-infested maize seeds were placed in cotton wool soaked in aqueous solution of ashes at different concentrations at 2-hour intervals for a 12-hour period for seven days (one day for each concentration) (Adesina, 2019). The repellency activity of the aqueous solution of wood ash were determined using a free choice (area preference) bioassay model, which was carried out in the demonstration cage, e.g. 25cmx 25cmx 10cm, because it is easily applicable and dependable (Gambari, 2021). Concentration of aqueous ash solution was calculated as:

$$\frac{\text{Quantity of power (g)}}{\text{1000 mls of water}} \times 100\%$$

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**Statistical Analysis**

The analysis of variance (ANOVA) approach was applied to the converted data using the statistical analysis system (Finney et al. 1979), which comprised the following: Abbott's Formula (Abbot et al., 1925) was used to adjust for control mortality before ANOVA, Tukey's test (P=0.05) was used for mean separation, and Probit analysis (Zar et al., 1999) was used to determine the lethal content (LC) causing 50% (LC50) and 95% (LC95) mortality of *S. zeamais* following treatment application.

**Results**

**Table 4.1: Showing proximate analysis of wood ashes and diatomaceous earth (synthetic)**

| **Parameter wt (%)** | *Acacia polyacantha* | *Hymenocardia acida* | **Diatomaceous earth** |
|---|---|---|---|
| Ash content | 3.90 | 0.13 | ---- |
| Moisture content | 146 | 0.75 | 2.00 |
| Fixed carbon | 15.59 | 11.12 | ---- |
| Volatile matter | 76.51 | 81.25 | ---- |
| Amorphous silica (SiO2) | --- | --- | 73.00 |
| Aerosol | --- | --- | 3.00 |

**Table 4.2: Showing percentage mortality, progeny suppression, damage reduction, weight loss and persistence of wood ash in synergy**

| **Treatment groups** | **%Mortality** | **%Progeny suppression** | **%Damage** | **%Weight loss** | **%Persistence** |
|---|---|---|---|---|---|
| Control | 0±0 | 32.41±8.83 | 97.67±0.33 | 93.26±1.68 | 0±0 |
| 5g | 97.5±1.44 | 31.67±9.28 | 3±0.58 | 2.82±0.98 | 86.67±3.33 |
| 10g | 98.33±0.83 | 39.44±10.56 | 2±0.58 | 3.95±0.52 | 95±2.89 |
| 20g | 100±0 | 36.11±7.35 | 1.33±0.33 | 0.92±0.35 | 96.67±1.67 |
| DE 0.5g | 100±0 | 44.44±5.56 | 2.33±0.33 | 4.34±2.12 | 96.67±1.67 |
| DE 1g | 100±0 | 33.33±33.33 | 2.67±0.67 | 9.09±3.97 | 100±0 |
| DE 1.5g | 100±0 | 16.67±16.67 | 0.33±0.33 | 2.66±2.52 | 100±0 |

*The Mean difference is significant at 0.05 level (Tukey's test)

**Table 4.3: Lethal contents resulting in 50% ($LC_{50}$) and 95% ($LC_{95}$) mortality of** *Sitophilus zeamais*

| **Treatment** | **$LC_{50}$** | **$LC_{95}$** |
|---|---|---|
| BIN | 0.23 | 3.32 |

*The Mean difference is significant at 0.05 level (Tukey's test)

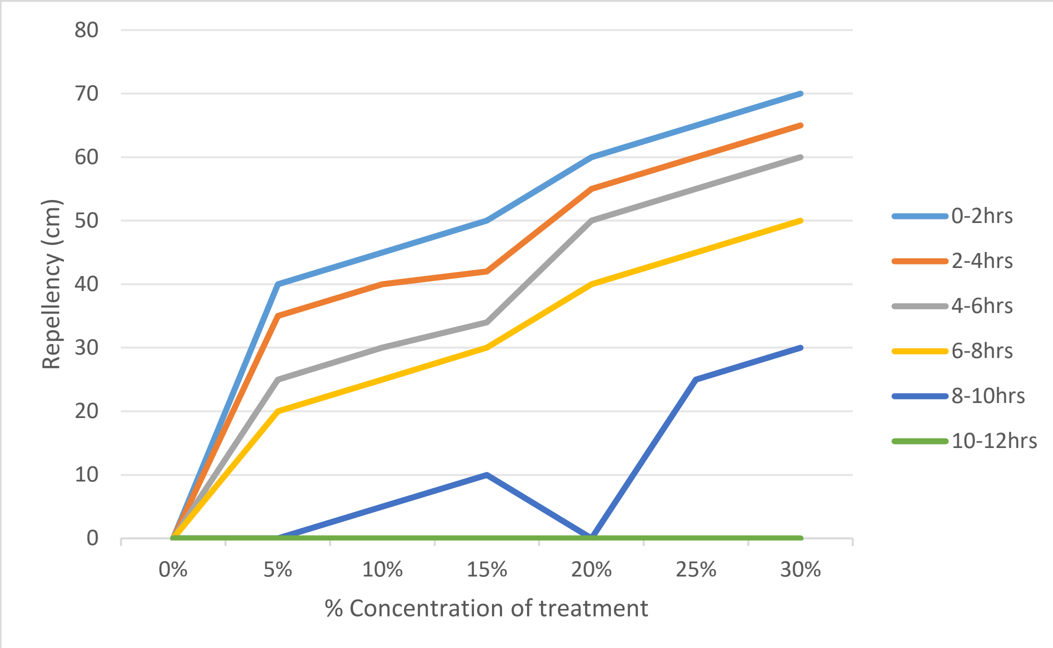


**Figure 4.1**: **Concentration of Wood ash from** *H. acida* **and** *A. polyacantha* **Combination**

**Discussion**

Two plant species- *Acacia polyacantha* and *Hymenocardia acida* whose branches were gathered and burnt, and the ash of wood was subjected to proximate analysis as shown in Table 4.1. Ash content, moisture content, fixed carbon and volatile matter were found in either of the trees as major parameters for the analysis. Moisture content, fixed content and volatile matter were found in high concentration, but ash content in either of the tree branches was low in concentration. The chemical composition of DE is indicated by the commercial label as shown in Table 4.2. The combination of wood ash from *Hymenocardia acida* and *Acacia polyacantha* has significantly raised the death rate of adult *S. zeamais*. As exposure time and dose increased, so did this mortality (Table 4.3). There was little difference in the tested powders' effectiveness. Over the course of 14 days, this fluctuation generally became very significant ($P< 0.05$). Within 14 days of exposure, the highest dose (20g) produced the highest fatality rate (100%) of any dose. At 5g, the mixed ashes showed a low death rate. However, as exposure time increased, the lowest dose (5g) resulted in a significant mortality rate of 97.50%.

A comparable findings to that of Mewis and Ulrich (2001), who found that after seven days of exposure, Fossil Shield dramatically decreased the survival of adult *Tribolium castaneum* and *Tribolium molitor*. Because insects' mobility enhances the cuticle's interaction with dust particles, exposure duration is therefore critical to DE's effectiveness (Athanassiou et al., 2005). With the exception of 0.5g, DE caused complete mortality starting on the seventh day after treatment. Demissie et al. (2008) observed the same pattern against *S. zeamais* using a different commercial DE formulation (Silicosec). These authors claim that at 1% and 2% exposure rates, Silico Sec eradicated 100% of *S. zeamais* in a 7-day exposure period. Subramanyam and Roesli suggest that insect mortality from diatomaceous earth may be due to dehydration caused by the abrasiveness of the small particles of this inert dust and the adsorption of oils in the insect's body (Field et al., 2000). This causes the epicuticle's wax covering to disintegrate, which worsens the fatal water loss (Subramanyam et al., 2000).

The production of *S. zeamais* offspring was significantly suppressed by the combination of *A. polyacantha* and *H. acida* wood ashes. The combinations of these products affect the development of *S. zeamais* in addition to raising mortality. Adult larval development may be disrupted or delayed by mixtures. Similar findings were made by Karso and Al Mallah (2014), who found that the combination of soya oil and Acetamprid insecticide resulted in the highest average mortality of *Trogoderma granarium* larvae, with the amount varying. According to Abrahim (2012), DE decreased the number of offspring by lowering oviposition, ovicidal, and larvicidal activities, as well as raising adult mortality. Arthur and Throne (2003) demonstrated that while exposure to DE kills adult weevils, some oviposition may still take place and suppression of progeny may not be successful. According to this study, this could account for the emergence of progeny with the lowest DE content (Table 4.2). Wood ash may limit the number of offspring by preventing emergence. Kitchen ash sprayed at a rate of 20% inhibited bruchid reproduction for a 39-week storage period, according to Baier & Webster (1992).

Weight losses, the proportion of damaged grains, and insect population increase were all reduced for the three treatments. A few of the variables that affected the effectiveness of the insecticidal substance were dosage and storage duration. However, each plant material's insecticidal properties would be determined by its active ingredients (Mewis, 2007).

This study also showed that pesticide persistence is correlated with exposure duration and diminishes with storage duration. Seven days after treatment, wood ash of *A. polyacantha* and *H. acida* showed a lower persistence rate. There was no discernible reduction of efficacy with regard to DE persistence. Compared to wood ashes, diatomaceous earth is more effective over an extended period of time. The results of Khakame et al. (2012), who found that Dryacide dust, a diatomaceous earth formulation, successfully shielded stored maize grain from *S. zeamais* for 15 days, are supported by this investigation. This suggested that determining the length of action of each insecticidal substance employed for grain storage, followed by the insecticide's persistence, was essential. This is comparable to the findings of Ntonifor et al. (2011), who found that when 1 g of Chenopodium ambrosioides leaf powder was applied to 50 g of maize, the mortality rate was 100% in the first week, but dropped to 87.5% in the second week for the same concentration, indicating that C. ambrosioides had lost its potency and required retreatment.

According to this study, grains that have been stored must be closely watched to avoid insect pest re-infestation, especially when the insecticidal material's long-term effectiveness is diminished. This study demonstrated that the combination of AP and HA wood ash generated an antagonistic impact after 14 days, that *S. zeamais* mortality produced a synergistic effect, and that the efficacy of mixes made of various insecticidal materials generally increased. The HA and AP mixtures of wood ashes also revealed significant mortality of *S. zeamais*. On the one hand, the combination of insecticidal materials can increase efficacy by lowering the doses of each product and complementing its bio-efficacy; on the other hand, it can broaden its spectrum of activity and reduce the likelihood of resistance development (Das, 2014). The additive effect, which asserts that the combined effect of two compounds is equal to the sum of each component delivered alone (1+2=3), was also demonstrated by the combination of AP and HA during the 14 days of exposure in the current investigation.

The treatment with 30% aqueous solution of ash mixture showed the maximum repellency of 70 cm in the first two hours. This was the pattern in the other treatments, with the treatment containing 5% concentration of the mixture exhibiting the least repellency of 40 cm as seen in figure 4.1. Over the next few hours, the effect progressively faded until the tenth or twelfth hours, when the weevils (0 cm) were unaffected. This is in line with previous research studies by Chaeib et al. (2009) and Marimuthu (2011), which showed that certain plant extracts distilled from *C. citrates, C. zeylanicum*, and *Z. officianale* had promising dose-dependent repellent properties against *Culex tritaeniorhynchus* and *Anophilus subpictus*, there is, therefore, an indirect relationship between exposure time and repellency as well as a direct relationship between concentration and repellency.

**Conclusion**

The study showed that mixture of wood ashes are hazardous to maize grains that have been kept. After stopping population increase, the ashes shielded the grains that had been preserved. Likewise, DE powder, which was employed as a positive control, demonstrated its ability to effectively prevent maize weevil infection in stored maize grains. There was a positive correlation between the ashes' reduction of damage and suppression of *S. zeamais* population growth. By reducing the quantity of perforated grains and weight loss while simultaneously preventing population growth, combinations of HA and AP wood ashes significantly decreased damage. Ash combination's repellent properties were noted, albeit they waned with time and lost their effectiveness after ten hour. The most significant discovery here is that wood ash's repellency declined over time, but it was strongly correlated with the amount of wood ash solution used.

**Recommendations**

Combining *A. polyacantha* and *H. acida* wood ash is also recommended because, while they increase the insecticidal efficacy by improving the bio-efficacy of the individual products and simultaneously reducing their dosages, they also broaden the range of activity and reduce the possibility of resistance development. More research is required to determine whether using these materials for grain storage poses a risk of toxicity to mammals.

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