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## Efficacy of diatomaceous earth and wood ash for the control of *Sitophilus zeamais* in stored maize

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### Abstract

The effectiveness of diatomaceous earth (FossilShield) and wood ash from *Acacia polyacantha* and *Hymenocardia acida* were assessed on *Sitophilus zeamais*, regarding adult mortality, progeny production, persistence, population increase, grain damage and seed viability. Maize grains were admixed with the dusts at the rates 0, 0.5, 1, 1.5 and 2 g/kg for FossilShield and 0, 5, 10, 20 and 40 g/kg for ash. FossilShield at 0.5 g/kg achieved 100% mortality of *S. zeamais* within 7 days of exposure and was more efficacious than the wood ash from *H. acida* (87.11% mortality) and *A. polyacantha* (4.82% mortality) at the rate of 40 g/kg for the same time-point. The three dusts caused significant reduction of progeny emergence, damaged grains and weight loss, but showed no adverse effect on the viability of the protected seeds. FossilShield and wood ash from *A. polyacantha* and *H. acida* could be of value as green stored grain protectants against *S. zeamais*.

**Keywords:** maize, *Sitophilus zeamais*, wood ash, diatomaceous earth, efficacy, persistence

### Introduction

Insect pests of stored products are responsible for considerable economic losses in the storage sector. The cosmopolitan maize weevil, *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae), is a very serious pest of maize grains and other cereals during storage<sup>[1,2]</sup>. The weevil together with other insects cause an estimated 24.5% loss of maize, and damaged grains have reduced nutritional value and weight, low frequency of germination, and low market value<sup>[3,4,5]</sup>. In Cameroon, where maize is the most important food crop and it is grown in all the 10 regions of the country, Nukenine *et al.*<sup>[6]</sup> recorded 33% of damaged grains caused by *S. zeamais* after five months of storage in the Adamawa Region.

In order to protect agricultural crops against insect pests, both in the field and storage, high amounts of synthetic residual insecticides are used around the world. These chemicals, although effective, have caused many environmental problems such as pollution, human health hazards and resistance in pests, among others<sup>[7,8]</sup>. Furthermore, the majority of farmers in Africa are resource-poor and do not have the means nor the skills to obtain and handle pesticides appropriately. These drawbacks of synthetic insecticides have stimulated intensive search for alternative measures that are safer and environment friendly<sup>[9,10]</sup>. Among potential natural reduced-risk pesticides are inert dusts and botanical insecticides. The use of chemically inert or less active materials, such as wood ashes, sand or other minerals, powders or plant products in large quantities to fill up the interstitial space in grain bulks, is quite widespread<sup>[11]</sup>. This means that newly hatched insect pests are denied of their activities. They experience more difficulty in finding partners and as result are forced to deposit their entire stock of eggs on relatively few maize kernels.

Many authors have reported the efficacy of wood ash as a grain protectant<sup>[12,13]</sup>. Golob *et al.*<sup>[13]</sup> evaluated the effectiveness of admixing locally available powders at the rate of 1%, 5%, 15% and 30% (w/w) with maize and found that wood ash, restricted infestation; wood ash at 30% being almost as effective as pirimiphos-methyl. The large quantities of wood ash needed to effectively protect stored grains against insect infestations, render its use less practical. Reducing the quantities of wood ash in stored grains is thus imperative. Since insecticidal properties of ashes vary according to the plant species<sup>[14]</sup>, the testing of wood ash from different plant species may lead to the identification of more active ash, which may be more practical in the storage sector. Another solution would be the use of other types of inert dust, which are far more active than wood ash. Silica-based inert materials such as silica aerogels and diatomaceous earths (DE) have proven to be very effective in smaller quantities.

Diatomaceous earth is almost pure amorphous silicon dioxide, made up of fossilised diatoms; it acts as an insecticide by absorption of epicuticular lipids and fatty acids, leading to desiccation in arthropods. Numerous DE formulations have been attempted for the management of stored-product pests with good results [15, 16]. DE is persistent in its action, poses few or no pest resistance problems, and it leaves no residue [17]. The efficacy of DE is affected by factors such as: its provenance, temperature, humidity and characteristics of target pests and substrate [17]. There are no studies reporting on DE and insects in the Western highlands of Cameroon (Bamenda), where the present study was carried out.

Thus, the present study was proposed to determine the efficacy of FossilShield (a diatomaceous earth) and wood ash from two local tree species, *Acacia polyacantha* (Mimosaceae) and *Hymenocardia acida* (Phyllanthaceae) against *S. zeamais* on maize, in view of establishing a sustainable management strategy for the weevil. Adult mortality, progeny inhibition and population suppression of *S. zeamais* as well as the weevil's damage reductions on the treated grains were among the parameters used to assess efficacy.

## 2. Materials and methods

### 2.1 Test insects and maize grain

*S. zeamais* was reared on maize grain in the Crop Protection laboratory of the Institute of Agricultural Research and Development (IRAD), Bambui, North-West region, Cameroon under ambient laboratory conditions (20 – 29 °C, 59 – 90% r.h.) (Data logger Model EL-USB-2, LASCAR, China). Adults of *S. zeamais* were obtained from a colony kept since 2005 in the Applied Zoology Laboratory of the University of Ngaoundere, Adamaoua region, Cameroon. Insects aged between 7 to 14 days were used for all bioassays with maize as substrate. The maize variety Shaba was obtained from IRAD Wakwa, Ngaoundere, Cameroon and the initial grain moisture content was 12.65%, measured using an Insto Moisture Tester (Insto, Auburn, IL, U.S.A.). The study was carried out during the period of January to May 2014.

### 2.2 Collection and preparation of wood ash of *Acacia polyacantha* and *Hymenocardia acida*

Stems of *A. polyacantha* and *H. acida* were collected respectively from Kousseri (latitude 12°04' N, longitude 15°02' E, altitude 283 m.a.s.l.), Far North region and Ngaoundere Cameroon (latitude 7°22' N, longitude 13°34' E, altitude 1.100 m.a.s.l.), Adamaoua region, Cameroon. The identity of the plants was confirmed at the Cameroon National Herbarium in Yaounde, where voucher samples were deposited. *A. polyacantha* is registered on number 36699/HNC and *H. acida* on number 50114/HNC. The stems were air-dried under the sun for two weeks, and then burnt separately in a traditional kitchen until at least 1 kg of ash was produced. The wood ash (1 kg) from each plant species was packaged in glass jars and kept in a refrigerator (-4 °C) until needed for bioassay.

### 2.3 Diatomaceous earth

The diatomaceous earth dust used was fossilShield 90.0, fine brownish powder containing amorphous silica particles with size ranging between 5 and 30 µm, a content of 73 % amorphous SiO<sub>2</sub>, a water content of about 2 %, in addition to 3% aerosol and other mineral compounds. It was obtained from the Fossil Shield Company, Bein GmbH, Eiterfeld, Germany.

### 2.4 Adult toxicity test and F<sub>1</sub> progeny production

The weights 0.25, 0.5, 1 and 2 g for *A. polyacantha* and *H. acida* wood ash and 0.025, 0.05, 0.075 and 0.1 g of

FossilShield were separately introduced into 50 g of maize in glass jars to give the contents 5, 10, 20 and 40 g/kg for wood ash and 0.5, 1, 1.5 and 2 g/kg for FossilShield. Controls consisted of maize grains without wood ash and FossilShield. Each jar was hand-shaken for 5 min to ensure uniform distribution of the dusts to the entire grain mass. Groups of 20 *S. zeamais* were separately added to glass jars containing the treated maize. Control glass jars also separately received twenty insects. All treatments were arranged in a completely randomized design on shelves in the laboratory under ambient conditions (23.7 ± 1.87 °C, 59.0 ± 7.63 r.h.), and each treatment had four replications. Mortality was recorded 1, 3, 7 and 14 days after treatment. Insects were considered dead when no movement was observed after touching them carefully with entomological forceps.

After the 14-day mortality recordings, all the insects and dusts were separated from the grains and discarded. The grains were left inside the jars and all the F<sub>1</sub> progeny were counted [18] (24.1 ± 2.07 °C, 62.1 ± 11.24 r.h.).

### 2.5 Persistence of wood ash and FossilShield on grains

Maize grains were treated with wood ash and FossilShield dusts in the same order as described previously for toxicity bioassay. Twenty adult *S. zeamais* were exposed to the treated grains which had been stored for 0, 15, 30 and 90 days [19] (23.9 ± 2.04 °C, 69.4 ± 4.95 r.h.). Mortality counts were carried out 7 days after exposure. All treatments were replicated four times.

### 2.6 Population increase and grain damage

Similar dosages of wood ash and FossilShield as for the toxicity bioassay described above were considered for 150 g grains. A group of 50 adult insects, of mixed sex were introduced into each jar containing treated or untreated grains. All treatment was replicated four times. After three months of storage, the number of live and dead insects was determined for each jar. Damage assessment was performed by counting and weighing the number of perforated and non-perforated grains using the method of Adams and Schulten [20]. Percent weight loss was determined as follows: [(initial weight-final weight)/100] × 100.

### 2.7 Seed germination

In order to assess the viability of seeds, seed germination was tested using 30 randomly picked seeds from undamaged grains after separation of damaged and undamaged grains in each jar from section 2.6 above. The seeds were placed on moistened paper in 9 cm glass Petri dish, which was humidified every two days, and the number of germinated and non-germinated seeds was recorded after 10 days [21].

### 2.8 Data analysis

Data on % cumulative corrected mortality, % reduction in F<sub>1</sub> progeny, % grain damage, % weight loss and % germination were arcsine-transformed [(square root(x/100))] and the number of F<sub>1</sub> progeny produced was log-transformed (x + 1). The transformed data were subjected to the analysis of variance (ANOVA) procedure using the Statistical Analysis System [22, 23]. Tukey's test (P = 0.05) was applied for mean separation. Probit analysis [22, 24] was conducted to determine lethal content (LC) causing 50% (LC<sub>50</sub>) and 95% (LC<sub>95</sub>) mortality of *S. zeamais* at 1, 3, 7 and 14 days after treatment application. Abbott's formula [25] was applied to correct for control mortality before ANOVA.

**3. Results**

**3.1 Toxicity tests**

As expected, compared to the control, all the three dusts generally caused significant dose-dependent mortality to adult *S. zeamais* (Table 1). However, the diatomaceous earth was more potent than the wood ash, and *H. acida* wood ash more active than that of *A. polyacantha*. Complete mortality (100%) of *S. zeamais* was achieved by FossilShield at the contents of 1, 1.5 and 2 g/kg, within seven days of exposure. Maximum mortality caused to the weevil by the highest tested content of the wood ash 40 g/kg was 94.66 for *H. acida* and 75.51 for *A. polyacantha*, 14 days after treatment. At 14 days post-treatment, the lowest tested contents for FossilShield, *H. acida* ash and *A. polyacantha* ash caused 79.24%, 69.08% and 20.73% mortality to *S. zeamais*, respectively.

The 7-day LC<sub>50</sub> values were 0.473 g/kg (FossilShield), 1.946 g/kg (*H. acida*) and 77.469 g/kg (*A. polyacantha*), while the 14-day LC<sub>50</sub> values in the same order were 0.456 g/kg, 1.936 g/kg and 12.209 g/kg (Table 2). The R<sup>2</sup> values ranged between 0.692 and 0.998. The slopes were higher for FossilShield (2.922 – 21.667) compared to wood ash (0.433 – 1.667).

**3.2 F<sub>1</sub> progeny production**

All the products caused significant reduction in progeny emergence relative to the control, which was dose-dependent (Table 3). FossilShield completely (100%) suppressed F<sub>1</sub> progeny emergence at all content level, except for the lowest content level 0.5 g/kg, which provided 95.50% of progeny inhibition. The two ashes almost totally suppressed F<sub>1</sub> progeny emergence at their highest tested content level (97.98% and 99.28%, respectively for *A. polyacantha* and *H. acida*). Even the lowest tested contents of the dusts caused ≥ 85% of progeny inhibition.

**3.3 Persistence on grains**

In general, the efficacy of FossilShield, but not wood ash, decreased with storage time (Table 4). The dust content

appeared to influence insecticidal efficacy because at the highest tested dust rate, the potency of FossilShield remained constant up to 90 days of storage, while with *H. acida* ash a higher *S. zeamais* mortality of 87.37% was registered at 0 day compared with 55.66% at 90 days.

**3.4 Population increase and damage reduction**

The rate of increase of the population of *S. zeamais* was significantly reduced by the three dusts regardless of content (Table 5). FossilShield at contents above 0.5 g/kg completely suppressed the populations of the weevil. No live insects were recovered after three months. At the lowest tested content 5 g/kg, the rate of population growth of the weevil was reduced roughly 10-fold and 61-fold respectively by ash of *H. polyacantha* and *H. acida*.

As with population increase, all the powders significantly reduced grain damage from the *S. zeamais* infestation, with the treated samples recording a smaller number of damaged grains than the controls (Table 5). At contents above 1 g/kg, FossilShield averted weevil damage. The samples treated with this dust showed no grain damage, and correspondingly no weight was lost. The lowest tested contents of ash 5 g/kg reduced grain damage and weight loss by at least 4-fold, while FossilShield at the lowest content 0.5 g/kg reduced these parameters by at least 22-fold.

**3.5 Germination**

All the three dusts greatly reduced germination losses resulting from *S. zeamais* infestation of maize grains (Table 5). Only 17.5% of the grains from the control jars germinated compared to the greater than 75% for wood ash and greater than 90% for FossilShield even at the lowest tested contents. The number of grains germinating increased with ascending contents for wood ash, but appeared to decline with increasing contents for FossilShield.

**Table 1 :** Corrected cumulative mortality of *Sitophilus zeamais* exposed to diatomaceous earth (FossilShield) and two wood ashes (*Acacia polyacantha* and *Hymenocardia acida*) under fluctuating laboratory conditions (t = 20 – 29 °C; r.h. = 59–90%)

Products and contents (g/kg)	1	3	7	14	F <sub>(3; 12)</sub>
<b><i>A. polyacantha</i></b>					
0	0.00 ± 0.00 <sup>ab</sup>	0.00 ± 0.00 <sup>ba</sup>	0.00 ± 0.00 <sup>ba</sup>	0.00 ± 0.00 <sup>ca</sup>	–
5	1.25 ± 1.25 <sup>ab</sup>	15.26 ± 5.49 <sup>ab</sup>	19.96 ± 9.37 <sup>ab</sup>	20.73 ± 5.38 <sup>ab</sup>	3.81*
10	5.00 ± 2.04 <sup>ac</sup>	16.65 ± 5.41 <sup>bc</sup>	25.94 ± 5.90 <sup>ab</sup>	50.00 ± 3.94 <sup>ab</sup>	14.29*
20	6.25 ± 2.39 <sup>ac</sup>	16.65 ± 5.41 <sup>bc</sup>	35.03 ± 8.47 <sup>ab</sup>	67.40 ± 6.19 <sup>ab</sup>	17.13***
40	7.50 ± 2.50 <sup>ac</sup>	27.06 ± 6.41 <sup>bc</sup>	41.82 ± 5.64 <sup>ab</sup>	75.51 ± 5.85 <sup>ab</sup>	22.79***
F <sub>(4;15)</sub>	2.84 <sup>ns</sup>	6.40*	16.76**	40.00***	
<b><i>H. acida</i></b>					
0	0.00 ± 0.00 <sup>ca</sup>	0.00 ± 0.00 <sup>ca</sup>	0.00 ± 0.00 <sup>ba</sup>	0.00 ± 0.00 <sup>ba</sup>	–
5	2.50 ± 1.44 <sup>bc</sup>	21.08 ± 3.73 <sup>bb</sup>	63.55 ± 12.84 <sup>ab</sup>	69.08 ± 10.78 <sup>ab</sup>	15.55**
10	5.00 ± 8.75 <sup>ab</sup>	24.57 ± 1.99 <sup>ab</sup>	72.69 ± 8.58 <sup>ab</sup>	82.60 ± 7.91 <sup>ab</sup>	26.11***
20	8.75 ± 1.25 <sup>ab</sup>	33.66 ± 2.01 <sup>ab</sup>	77.96 ± 8.65 <sup>ab</sup>	90.64 ± 6.23 <sup>ab</sup>	22.40***
40	12.50 ± 1.44 <sup>ab</sup>	37.41 ± 4.63 <sup>ab</sup>	87.11 ± 4.91 <sup>ab</sup>	94.66 ± 3.72 <sup>ab</sup>	39.12***
F <sub>(4;15)</sub>	19.66**	66.26***	18.27***	26.03***	
<b>FossilShield</b>					
0	0.00 ± 0.00 <sup>ca</sup>	0.00 ± 0.00 <sup>da</sup>	0.00 ± 0.00 <sup>ca</sup>	0.00 ± 0.00 <sup>ca</sup>	–
0.5	3.75 ± 1.25 <sup>bc</sup>	48.16 ± 2.79 <sup>ca</sup>	69.47 ± 9.73 <sup>ba</sup>	79.24 ± 10.76 <sup>ba</sup>	17.34***
1	10.00 ± 2.89 <sup>bc</sup>	76.12 ± 6.80 <sup>bb</sup>	100.00 ± 0.00 <sup>ab</sup>	100.00 ± 0.00 <sup>ab</sup>	155.41***
1.5	16.25 ± 2.39 <sup>ac</sup>	88.62 ± 3.13 <sup>abb</sup>	100.00 ± 0.00 <sup>ab</sup>	100.00 ± 0.00 <sup>ab</sup>	338.60***
2	21.25 ± 4.27 <sup>ab</sup>	97.44 ± 1.48 <sup>ab</sup>	100.00 ± 0.00 <sup>ab</sup>	100.00 ± 0.00 <sup>ab</sup>	159.04***
F <sub>(4;15)</sub>	18.46***	68.98***	195.99***	80.84***	

Means ± S.E. followed by the same capital letter in lines and the same lower letter in the column do not differ significantly at P < 0.05 (Tukey's test)

Each datum represents the mean of four replicates of 20 insects each.

<sup>ns</sup> P > 0.05, \* P < 0.05, \*\* P < 0.001, \*\*\* P < 0.0001

**Table 2:** Lethal contents resulting in 50% (LC<sub>50</sub>) and 95% (LC<sub>95</sub>) mortality of *Sitophilus zeamais* adults in grains treated with wood ashes (*Acacia polyacantha*, *Hymenocardia acida*) and diatomaceous earth (fossil shield) in ambient conditions (t = 20–29 °C; r.h. = 59–90%)

Products	Slope ± SE	R <sup>2</sup>	LC <sub>50</sub> (95%FL) (g/kg)	LC <sub>95</sub> (95%FL) (g/kg)	χ <sup>2</sup>
<b>3 days</b>					
<i>A. polyacantha</i>	0.433 ± 0.216	0.712	—	—	1.455 <sup>ns</sup>
<i>H. acida</i>	0.569 ± 0.199	0.966	135.326 (50.483; 18401) <sup>#</sup>	—	0.289 <sup>ns</sup>
FossilShield	2.922 ± 0.345	0.971	0.536 (0.436; 0.622)	1.963 (1.638; 2.567)	1.880 <sup>ns</sup>
<b>7 days</b>					
<i>A. polyacantha</i>	0.694 ± 0.198	0.992	77.469 (39.066; 688.413) <sup>#</sup>	—	0.085 <sup>ns</sup>
<i>H. acida</i>	0.826 ± 0.210	0.988	1.946 (0.280; 3.910)	190.821 (73.336; 3007) <sup>#</sup>	0.323 <sup>ns</sup>
FossilShield	21.665 ± 4.618	0.692	0.473 <sup>‡</sup>	0.564 <sup>‡</sup>	
<b>14 days</b>					
<i>A. polyacantha</i>	1.667 ± 0.330	0.922	12.509 (2.657; 33.058)	121.239 (40.215; 34433197) <sup>#</sup>	5.199 <sup>ns</sup>
<i>H. acida</i>	1.268 ± 0.249	0.929	1.936 (0.618; 3.316)	38.341 (25.041; 91.006)	0.195 <sup>ns</sup>
FossilShield	20.652 ± 4.740	0.692	0.456 <sup>‡</sup>	0.548 <sup>‡</sup>	

ns: P &gt; 0.05 ;

#. The LC values were obtained by extrapolation;

—: Estimated LC values are too large or estimation impossible due to inadequate mortality

‡: The Fudicial limit values for these LC could not be computed due to very low variations in mortality among the different contents of insecticidal powder

**Table 3:** Progeny production of *Sitophilus zeamais* in grains treated with FossilShield, *Acacia polyacantha* and *Hymenocardia acida* wood ashes under fluctuating laboratory conditions (t = 20–29 °C; r.h. = 59–90%)

Products and contents (g/kg)	F <sub>1</sub> progeny	% reduction of F <sub>1</sub> relative to control	EC <sub>50</sub> (g/kg)
<b><i>A. polyacantha</i></b>			
0	43.00 ± 9.32 <sup>a</sup>	0.00 ± 0.00 <sup>d</sup>	0.708
5	6.75 ± 1.89 <sup>b</sup>	84.66 ± 2.12 <sup>c</sup>	
10	5.00 ± 1.78 <sup>b</sup>	88.96 ± 3.24 <sup>bc</sup>	
20	2.25 ± 1.60 <sup>b</sup>	95.77 ± 2.13 <sup>ab</sup>	
40	1.25 ± 0.95 <sup>b</sup>	97.98 ± 1.37 <sup>a</sup>	
F <sub>(4; 15)</sub>	22.18***	161.14***	
<b><i>H. acida</i></b>			
0	43.00 ± 9.32 <sup>a</sup>	0.00 ± 0.00 <sup>d</sup>	0.010
5	1.75 ± 0.48 <sup>b</sup>	95.60 ± 2.44 <sup>a</sup>	
10	2.00 ± 1.08 <sup>b</sup>	95.88 ± 0.91 <sup>a</sup>	
20	1.25 ± 0.63 <sup>b</sup>	97.10 ± 1.47 <sup>a</sup>	
40	0.50 ± 0.50 <sup>b</sup>	99.28 ± 0.73 <sup>a</sup>	
F <sub>(4; 15)</sub>	38.82***	200.07***	
<b>FossilShield</b>			
0	43.00 ± 9.32 <sup>a</sup>	0.00 ± 0.00 <sup>b</sup>	0.403
0.5	2.25 ± 1.44 <sup>b</sup>	95.50 ± 3.22 <sup>a</sup>	
1	0.00 ± 0.00 <sup>b</sup>	100.00 ± 0.00 <sup>a</sup>	
1.5	0.00 ± 0.00 <sup>b</sup>	100.00 ± 0.00 <sup>a</sup>	
2	0.00 ± 0.00 <sup>b</sup>	100.00 ± 0.00 <sup>a</sup>	
F <sub>(4; 15)</sub>	47.82***	282.86***	

Means ± S.E. in the same column for the same category of insecticide, followed by the same letter do not differ significantly at P &lt; 0.05 (Tukey's test)

ns P &gt; 0.05, \* P &lt; 0.05, \*\* P &lt; 0.001, \*\*\* P &lt; 0.0001.

**Table 4:** Corrective cumulative mortality of *Sitophilus zeamais* after 7 days exposure treated with insecticidal powders stored for 0; 15; 30 and 90 days before infestation (t = 20–29 °C; r.h. = 59–90%)

Products and contents (g/kg)	Storage periods (days) before infestation by <i>Sitophilus zeamais</i>				F <sub>(3; 12)</sub>
	0	15	30	90	
<b><i>A. polyacantha</i></b>					
0	0.00 ± 0.00 <sup>bA</sup>	0.00 ± 0.00 <sup>bA</sup>	0.00 ± 0.00 <sup>bA</sup>	0.00 ± 0.00 <sup>bA</sup>	—
5	22.89 ± 4.63 <sup>aA</sup>	36.25 ± 11.97 <sup>aA</sup>	29.21 ± 3.47 <sup>aA</sup>	34.34 ± 4.08 <sup>aA</sup>	3.06 <sup>ns</sup>
10	28.03 ± 5.00 <sup>aA</sup>	32.50 ± 5.95 <sup>aA</sup>	32.70 ± 6.44 <sup>aA</sup>	39.61 ± 10.44 <sup>aA</sup>	0.61 <sup>ns</sup>
20	36.84 ± 7.94 <sup>aA</sup>	41.25 ± 4.73 <sup>aA</sup>	41.51 ± 9.01 <sup>aA</sup>	40.46 ± 1.69 <sup>aA</sup>	0.39 <sup>ns</sup>
40	47.89 ± 7.48 <sup>aA</sup>	43.55 ± 4.94 <sup>aA</sup>	42.50 ± 5.95 <sup>aA</sup>	40.79 ± 8.08 <sup>aA</sup>	0.51 <sup>ns</sup>
F <sub>(4; 15)</sub>	25.10***	13.90***	21.57***	20.83***	
<b><i>H. acida</i></b>					
0	0.00 ± 0.00 <sup>bA</sup>	0.00 ± 0.00 <sup>bA</sup>	0.00 ± 0.00 <sup>bA</sup>	0.00 ± 0.00 <sup>bA</sup>	—
5	64.47 ± 12.00 <sup>aA</sup>	53.09 ± 4.02 <sup>aA</sup>	47.04 ± 5.50 <sup>aA</sup>	42.50 ± 8.29 <sup>aA</sup>	1.41 <sup>ns</sup>
10	73.36 ± 8.19 <sup>aA</sup>	56.25 ± 4.73 <sup>aA</sup>	55.86 ± 10.62 <sup>aA</sup>	48.22 ± 5.20 <sup>aA</sup>	1.06 <sup>ns</sup>
20	78.49 ± 8.25 <sup>aA</sup>	57.50 ± 7.77 <sup>aB</sup>	56.25 ± 10.08 <sup>aA</sup>	49.34 ± 10.82 <sup>aA</sup>	2.59 <sup>ns</sup>
40	87.37 ± 4.77 <sup>aA</sup>	60.72 ± 4.35 <sup>aA</sup>	59.41 ± 5.23 <sup>aB</sup>	55.66 ± 9.26 <sup>aB</sup>	5.70*
F <sub>(4; 15)</sub>	19.42***	25.40***	35.29***	24.06***	
<b>FossilShield</b>					

0	0.00 ± 0.00 <sup>cA</sup>	0.00 ± 0.00 <sup>cA</sup>	0.00 ± 0.00 <sup>dA</sup>	0.00 ± 0.00 <sup>cA</sup>	–
0.5	70.33 ± 9.21 <sup>bA</sup>	71.25 ± 11.43 <sup>bA</sup>	64.34 ± 6.01 <sup>cA</sup>	60.86 ± 4.94 <sup>bA</sup>	0.50*
1	100.00 ± 0.00 <sup>aA</sup>	98.75 ± 1.25 <sup>aA</sup>	89.80 ± 2.21 <sup>bB</sup>	79.54 ± 6.82 <sup>bB</sup>	14.21**
1.5	100.00 ± 0.00 <sup>aA</sup>	100.00 ± 0.00 <sup>aA</sup>	96.18 ± 1.27 <sup>bA</sup>	93.62 ± 2.46 <sup>aAB</sup>	5.71*
2	100.00 ± 0.00 <sup>aA</sup>	100.00 ± 0.00 <sup>aA</sup>	100.00 ± 0.00 <sup>aA</sup>	100.00 ± 0.00 <sup>aA</sup>	–
F(4;15)	212.12***	65.00***	192.19***	122.02***	

Means ± S.E. followed by the same capital letter in lines and the same lower letter in the column do not differ significantly at  $P < 0.05$  (Tukey's test)

Each datum represents the mean of four replicates of 20 insects each.

<sup>ns</sup>  $P > 0.05$ , \*  $P < 0.05$ , \*\*  $P < 0.001$ , \*\*\*  $P < 0.0001$

**Table 5:** Population increase (mean number of progeny for four jars ± SE), damage parameters of *Sitophilus zeamais* and germination of maize seeds admixed with different contents of FossilShield, *Acacia polyacantha* and *Hymenocardia acida* wood ashes<sup>a</sup> and stored for three months under fluctuating laboratory conditions (t = 20–29 °C; r.h. = 59–90%)

Content (g/kg) and Products	Live insects	Percentage of seed damage	Percentage of weight loss	Percentage of germination
<b>A. polyacantha</b>				
0	184.25 ± 9.34 <sup>a</sup>	27.88 ± 3.11 <sup>a</sup>	7.71 ± 1.06 <sup>a</sup>	17.50 ± 3.16 <sup>b</sup>
5	19.25 ± 5.68 <sup>b</sup>	6.89 ± 1.46 <sup>b</sup>	1.85 ± 0.35 <sup>b</sup>	76.69 ± 7.58 <sup>a</sup>
10	8.50 ± 3.50 <sup>b</sup>	5.77 ± 0.80 <sup>b</sup>	1.37 ± 0.24 <sup>b</sup>	85.67 ± 0.84 <sup>a</sup>
20	5.50 ± 2.72 <sup>b</sup>	4.76 ± 0.89 <sup>b</sup>	0.92 ± 0.14 <sup>b</sup>	90.00 ± 2.72 <sup>a</sup>
40	9.75 ± 2.69 <sup>b</sup>	5.00 ± 1.10 <sup>b</sup>	1.30 ± 0.27 <sup>b</sup>	93.33 ± 1.36 <sup>a</sup>
F(4;15)	206.59***	34.54***	29.48***	64.00***
<b>H. acida</b>				
0	184.25 ± 9.34 <sup>a</sup>	27.88 ± 3.11 <sup>a</sup>	7.71 ± 1.06 <sup>a</sup>	17.50 ± 3.16 <sup>c</sup>
5	3.00 ± 1.00 <sup>b</sup>	4.03 ± 0.78 <sup>b</sup>	0.83 ± 0.33 <sup>b</sup>	76.67 ± 4.30 <sup>b</sup>
10	7.00 ± 3.19 <sup>b</sup>	3.90 ± 0.95 <sup>b</sup>	1.04 ± 0.30 <sup>b</sup>	86.67 ± 3.60 <sup>ab</sup>
20	5.75 ± 2.14 <sup>b</sup>	3.56 ± 0.74 <sup>b</sup>	0.68 ± 0.17 <sup>b</sup>	90.00 ± 1.36 <sup>a</sup>
40	2.25 ± 0.48 <sup>b</sup>	1.97 ± 0.79 <sup>b</sup>	0.52 ± 0.22 <sup>b</sup>	92.50 ± 1.60 <sup>a</sup>
F(4;15)	313.30**	48.96***	34.71***	107.71***
<b>FossilShield</b>				
0	184.25 ± 9.34 <sup>a</sup>	27.3 ± 3.11 <sup>a</sup>	7.71 ± 1.06 <sup>a</sup>	17.50 ± 3.16 <sup>b</sup>
0.5	1.75 ± 0.85 <sup>b</sup>	1.22 ± 0.46 <sup>b</sup>	0.24 ± 0.14 <sup>b</sup>	93.33 ± 1.36 <sup>a</sup>
1	0.00 ± 0.00 <sup>b</sup>	0.27 ± 0.27 <sup>b</sup>	0.05 ± 0.05 <sup>b</sup>	95.83 ± 1.60 <sup>a</sup>
1.5	0.00 ± 0.00 <sup>b</sup>	0.00 ± 0.00 <sup>b</sup>	0.00 ± 0.00 <sup>b</sup>	90.00 ± 2.36 <sup>a</sup>
2	0.00 ± 0.00 <sup>b</sup>	0.00 ± 0.00 <sup>b</sup>	0.00 ± 0.00 <sup>b</sup>	91.67 ± 2.89 <sup>a</sup>
F(4;15)	384.16***	76.03***	51.16***	200.97***

Means ± S.E. in the same column for the same category of insecticide, followed by the same letter do not differ significantly at  $P < 0.05$  (Tukey's test)

<sup>ns</sup>  $P > 0.05$ , \*  $P < 0.05$ , \*\*  $P < 0.001$ , \*\*\*  $P < 0.0001$ .

#### 4. Discussion

Our investigations showed that the diatomaceous earth (DE), FossilShield and the wood ash from *A. polyacantha* and *H. Acida* were toxic to adult *S. zeamais*, causing significant mortality to the weevil. However, the toxic action of the wood ash was far slower than that of the DE. FossilShield at the rate 1 g/kg caused 100% to *S. zeamais* within 7 days, while, for the same time-point; the wood ash caused 87% mortality to the weevil at the rate 40 g/kg. The potency of the of *H. acida* wood as was superior to that of *A. polyacantha* since within 7 days and at the rate 40 g/kg, the former and the later caused respectively 87.11% and 41.82% mortality to *S. zeamais*. The toxic action increased with ascending exposure period for all the three dusts. This observation was well elucidated by the decreasing LC<sub>50</sub> values from the 3<sup>rd</sup> to the 14<sup>th</sup> day after treatment. The increase of adult mortality according to ascending exposure period and contents was due to the increase of the quantity of active ingredients contained in the insecticidal materials.

Mewis and Ulrich [26] reported that FossilShield significantly reduced the survival of adult *Tribolium castaneum* and *Tribolium molitor* after seven days of exposure; this is similar to the present finding. The exposure time is crucial for the effectiveness of DE, because insects' movement increases the contact of the cuticle with dust particles [27]. FossilShield induced total mortality from the 7<sup>th</sup> day post-treatment for all the content levels, except 0.5 g/kg. The same tendency was

recorded by Demissie *et al.* [11] using another commercial DE formulation (Silicosec) against *S. zeamais*. These authors reported that SilicoSec caused 100% mortality of *S. zeamais* within 7 days exposure period, at the rates 1% and 2%. The death of insects caused by diatomaceous earth could be attributed to the dehydration provoked by the abrasiveness of the small particles of this inert dust and by adsorption of oils in the body of the insect [28, 29, 30], which breaks the layer of wax on the epicuticle, exacerbating the fatal loss of water as reported by Subramanyam and Roesli [15]. Wood ash as inert dusts induces insect mortality by their physical properties; it desiccates the insect cuticle and provokes suffocation in insect. Abraham *et al.* [31] observed that DE reduced the production of progeny by increasing adult mortality, reducing oviposition, ovidical and larvicidal activities. Arthur and Throne [32], showed that adult weevils are killed by exposure to DE, some oviposition could still occur and progeny suppression may not be effective. That may explain the emergence of progeny at lowest content of FossilShield. The two wood ashes were highly toxic and this toxicity was different according to the plant species. The wood ash of *H. acida* was more effective than those of *A. polyacantha*. Gwinner *et al.* [33] reported that the effect of ash varies according to the type of ash. Wood ash may control the production of progeny by inhibiting emergence. Baier and Webster [34] found kitchen ash applied at the rate of 20% w/w controlled bruchid reproduction throughout 39 weeks storage period.

The efficacy of insecticidal is related to time of exposure, it decreases when the storage period goes on. This revealed important to determine the action duration of each insecticidal material used for grain storage then the persistence of insecticide. In our experiment, the efficacy of three insecticidal products slightly decreased when the storage period increased. Ntonifor *et al.* [35] reported that 1 g of leaf powder of *Chenopodium ambrosioides* applied to 50 g of maize caused 100% mortality during the first week. But at the second week for the same concentration, the mortality rate decreased to 87.5%. Thus *C. ambrosioides* lost its potency and needed to be retreated. This report makes obligatory the monitoring of stored grains to avoid re-infestation by insect pests, especially when the persistent efficacy of used insecticidal material is not longer as the storage time. The similar findings were reported for *A. polyacantha* and *H. acida* ashes; the mortality rate decreased within 0 day to 90 days. Concerning the persistence of FossilShield, the loss of efficacy was not really observed. Diatomaceous earth remains effective for long time compared to wood ashes. Khakame *et al.* [36] reported that Dryacide dust, a formulation of diatomaceous earth provided effective protection to stored maize grain against *S. zeamais* for 9 months. Thus, this inert powder remained effective for long time compared to the wood ashes.

Reductions in insect population growth, percentage of grains damaged and weight losses were observed for the three treatments. The efficacy of insecticidal material was influenced by several factors such as content, storage period, and season. The efficacy of insecticidal powder decreases when the relative humidity increases. Because the water contained in relative humidity dilutes the powder reducing the concentration of active compounds. However, the insecticidal property of any plant material would depend on the active constituents of the plant material [37]. Diatomaceous earth use is one of the well-studied alternatives to traditional neurotoxic grain protectants. Kavallieratos *et al.* [29] reported that DE was effective against *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae) in: maize, wheat, barley, oats and rice. Many studies reported the ability of wood ashes to protect stored grains against infestation by insect pests. Gemu *et al.* [38] found that the wood ash at content of 10% reduced weight (0.24%) and seed damage (15 /500) compared to the control (weight loss: 4.21%; seed damage: 165.67/500). Leaf ashes of *Eucalyptus grandis* at the rate of 0.25g/25g grains significantly reduced the number of emerged weevils. *A. polyacantha* and *H. acida* wood ashes showed the similar efficacy to protect stored maize grains. The two ashes prevented population growth then provided protection of stored grains. Wood ash can effectively protect grain stored in small lots by traditional Africa farmers [13].

In many African countries, the storage grains provide not only the grains for meal but also the seeds for crop. Thus, the conservation of seed viability by protectant is necessary. The seeds treated with inert materials (ashes, FossilShield) conserved significantly their viability and no difference in terms of germination was observed among these three products. But the seeds treated with the lowest contents of these products recorded the lowest germination rate; this was related to the development of weevils, which emerged at these level contents. The seeds used for germination were non-perforated but they would contain the immature stages of weevil that could destroy the parts of seed embryo. The viability of bambara groundnut seeds treated with diatomaceous earth and plant powder against *C. subinnotatus* demonstrated that the inert materials have no visible effect on

the germination ability of the treated seeds except that some of the treatments were infected by moulds which resulted in reduced germination percentage [31]. Stathers *et al.* [39] also reported that diatomaceous earth did not have any negative effects on seed germination. Akob and Ewete [14] observed that *E. grandis* ash at 2 g/2kg of maize protected grains for 6 months without any adverse affect on seeds germination. According to Couturon [40], when environmental conditions are not well controlled, germination rate decreases quickly. That could partly explain the loss of viability.

Diatomaceous earths (FossilShield), *A. polyacantha* and *H. acida* wood ashes showed their potency to protect stored maize grains against maize weevil infestation. In developing countries, especially in Sub-saharan Africa where the populations are resource poor, the use of insecticidal plant materials can greatly contribute to fight food insecurity by reducing post harvest losses as the plants are locally available. However, further work would be required inter alia to estimate the duration of efficacy and consumer safety, before promoting their use in stored product protection.

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