

Full Length Research Paper

Entomocidal activity of some plant extracts against *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae)

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Entomocidal activity of ethanolic leaf extracts of four plant species namely *Euphorbia balsamifera* Aiton, *Lawsonia inermis* L., *Mitracarpus hirtus* (L.) DC. and *Senna obtusifolia* L. were evaluated in order to assess their protectant ability to stored sorghum against *Sitophilus zeamais* Motsch. The botanicals were applied to 20 g of sorghum grains at the rate of 25, 50 and 100 mgml⁻¹, while no extracts was added to the control. Percentage repellency was recorded at 24 h after exposure (HAE) while adult mortalities of the weevils were obtained at 3 days after treatment (DAT). Adult emergence and grain perforations were recorded at 49 and 84 DAT, respectively. Repellency levels of the botanicals against *S. zeamais* ranged from 87.05 ± 0.45 to 100.00 ± 0.00%. Also, the ethanolic extracts resulted in high adult mortalities of the weevils in the treated sorghum grains. No adult emergence was recorded in grains treated with the botanical extracts. Highest (10.00 ± 2.04%) grain perforations among the treatments were observed in 25 mgml⁻¹ of *S. obtusifolia*, while 2.50 ± 1.25% was the least in the highest concentration of *E. balsamifera*. Findings of the study have revealed that the selected botanicals could serve as stored sorghum protectants against *S. zeamais* infestations.

Key words: Adult mortality, adult emergence, grain perforations, plant extracts, repellency levels, *Sitophilus zeamais*.

INTRODUCTION

Sorghum (*Sorghum bicolor* L. Moench) is an important cereal crop grown worldwide for food and animal feed purposes (Dahlberg et al., 2011). Sorghum belongs to the grass family Gramineae and one of the most important cereal crops grown in the tropics and sub-tropics (Mofokeng, 2016). The crop is mostly cultivated in semi-arid tropics where water is scarce and drought is frequent (ICRISAT, 2015).

Insect pests are a key constraint to effective production

and utilization of cereal crops in sub-Saharan Africa (SSA), and post-harvest losses resulting from insect remain a huge challenge (Tefera et al., 2010; Midega et al., 2016). Insect infestations to stored products are one of the major agricultural development problems in the tropics which results in substantial waste of farm produce and hence considerable loss to the economy (Adejumo and Raji, 2007; Abbas et al., 2014; Sori, 2014). Insect pests damage to stored grains results in reduction of

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quantity as well as quality deterioration of stored cereals like sorghum, maize, wheat and rice (Okonkwo, 1998).

Sitophilus zeamais is the most devastating storage pest of maize, causing serious management problem facing agriculture in developing countries (Abulude et al., 2007). The grain damage caused by *S. zeamais* to cereal crops prompted researchers to investigate more on controlling strategies of the weevils in maize and wheat (Rugumamu, 2012; Khaliq et al., 2014; Cosmas et al., 2018). Although most of the researchers investigated the level of damages caused by *S. zeamais* to stored maize, some have found it necessary to involve stored sorghum considering its position as the second cereal crop in Africa which is also infested and damaged by the maize weevil (Goftishu and Belete, 2014; Suleiman, 2014; Suleiman and Rugumamu, 2017; Suleiman et al., 2018a). Despite effective pest control both on farm fields and storage provided by synthetic insecticides, there is growing concern about the potential hazards to the ecosystem (Midega et al., 2016; Cosmas et al., 2018). Hill (1997) among others had earlier emphasized that, to reduce the hazards associated with chemical application in stored cereals, Integrated Pest Management (IPM) approach, involving all the suitable techniques and methods in maintaining pest population below economic injury level (EIL) is necessary.

To complement IPM strategies, botanicals have been used for many years by small scale farmers in many parts of Africa to protect stored products from insect infestations (Tesema et al., 2015). Plant materials have been applied to a commodity, usually at a rate of 1 to 5% (w/w) to protect stored grains from insect pest infestations (Dales, 1996). Laboratory and field evaluations of some botanical products in Africa indicate their effectiveness against insect pests of stored products (Ajayi, 2013; Rugumamu, 2014, 2015; Ojo and Omoloye, 2016; Suleiman et al., 2018b).

In view of the promising effects of some botanicals against insect pests of stored products, *Euphorbia balsamifera* Aiton, *Lawsonia inermis* L., *Mitracarpus hirtus* (L.) DC and *Senna obtusifolia* L. were selected for this study due to their local availability, and reported efficacies of some of them against other insect pests such as *Callosobruchus maculatus* F. and *Trogoderma granarium* Everts (Al-Moajel, 2004; Chudasama et al., 2015). This study was therefore aimed at evaluating effect of ethanolic extracts of the aforementioned botanicals on the control of *S. zeamais* infesting stored sorghum grains.

MATERIALS AND METHODS

Rearing of *S. zeamais*

Fifty pairs of *S. zeamais* were obtained from infested grain stores and then introduced into each of 500 ml rearing bottles containing 250 g of the disinfested sorghum grains which served as parent stock. The bottles were covered with muslin cloth and kept in the

incubator for oviposition at 30°C and 70% R.H. for 14 days, after which the parents were removed. The bottles were maintained in the incubator under the same condition for adult emergence. Progeny of 1-7 days old were sieved and used for the laboratory experiments.

Preparation of the botanical extracts

Sufficient amount of fresh leaves of *E. balsamifera*, *L. inermis*, *M. hirtus* and *S. obtusifolia* was collected and rinsed with distilled water to remove any dust and unwanted particles. They were then shade-dried at room temperature. The dried leaves were ground into powder using a laboratory blender and sieved using a laboratory sieve with mesh size of 80 microns.

Method of Khaliq et al. (2014) was followed for extraction of the botanicals. One hundred grams of each of the plant powders was dissolved in 400 ml of ethanol in conical flasks. Mouths of the conical flasks were properly corked and kept in a refrigerator for 48 h. The extract was separated using muslin cloth and filtered with Whatman No.1 filter papers using vacuum pump. The filtrate was separately concentrated by evaporating excess solvents using rotary evaporator with rotary speed of 3 to 6 rpm for 8 h. The aliquot was poured into crucibles and placed on water bath to evaporate the remaining excess solvents. The resulting extracts were air-dried and stored in refrigerator at 4°C.

Determination of repellency levels of ethanolic extracts against *S. zeamais*

Each of the four botanicals was diluted to three different concentrations of 25, 50 and 100 mgml⁻¹ each of ethanol. Two milliliters of each of the extracts was mixed with 20 g of sorghum grains in bottle A and allowed to completely air-dried. Another 20 g of the grains without any botanical extract was placed in bottle C serving as a control. Ten adult *S. zeamais* were introduced in bottle B from where their direction of movement to either bottle A or C was observed at 32 ± 2°C and 65 ± 10% R.H. The number of weevils moving from (B) to either (A) or (C) was recorded at 24 h after exposure (HAE). Percentage repellency was calculated according to the methods of Sakuma and Funkami (1985) using the following formula:

$$PR = \left[1 - \frac{NT}{NT + NC} \right] \times 100$$

Where: PR = Percentage Repellency.

NT = Number of insects in the chemical-treated test chamber.

NC = Number of insects in the control test chamber.

Assessment of adult mortality of *S. zeamais* in sorghum treated with ethanolic leaf extracts

Method of de Oliveira et al. (2012) was adopted to assess per cent adult mortality of *S. zeamais* in sorghum grains treated with different leaf extracts of the test plants. Crude extracts were diluted with ethanol to make different concentrations of 25, 50 and 100 mgml⁻¹ as previously described. Twenty grams of sorghum grains were weighed and placed in treatment bottles (250 ml). The grains were impregnated with 2 ml of the diluted extracts at the three concentrations, while the control contained the grains only. The grain mass was mixed thoroughly with the aid of glass rod and air-dried for 4 h. Five pairs of adult weevils were introduced into the bottles containing the treated grains, and covered with muslin cloth secured by rubber bands. All treatments were arranged in a completely randomized design (CRD) in the incubator with four

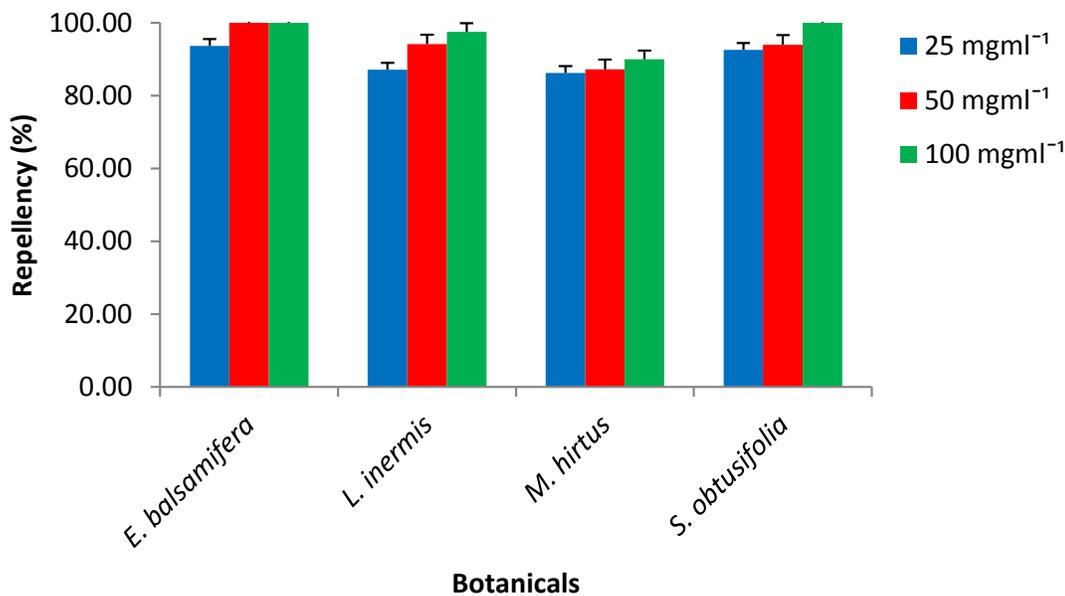


Figure 1. Mean percentage repellency of ethanolic extracts of different botanicals applied at different concentrations against *S. zeamais* at 24 HAE.

replicates. The set-ups were inspected daily and dead weevils in each treatment were removed and recorded daily for three days (when the first total mortality was achieved).

Data were recorded, organized, analyzed and presented as mortalities of *S. zeamais* within 3 DAT. Weevils in untreated containers were allowed to remain in the grains until they reached 14 days for oviposition before they were removed leaving the grains only until F_1 emerged. Percent adult mortality was assessed as number of dead weevils divided by total number of weevils per each bottle, multiplied by one hundred.

Examination of adult emergence of *S. zeamais*

Grains were inspected daily after removal of the earlier introduced individuals for adult emergence of the weevils. The emerging progenies (F_1) from each bottle were removed daily, counted and recorded for 49 days (84 days after infestation) after which observations stopped to avoid overlapping of generations. Inhibition rate (IR) in adult emergence was calculated using the following formula (Tapondju et al., 2002):

$$IR = \frac{C_n - T_n}{C_n} \times 100$$

Where:

IR = Inhibition rate (%)

C_n = Number of insects that emerged in the control; and

T_n = Number of insects that emerged in the treated grains.

Assessment of grain perforations in treated sorghum infested by *S. zeamais*

To assess grains perforations made by *S. zeamais*, 20 grains from the extract treatments were sampled and examined after 49 days of emergence of F_1 . Percent grain perforations were assessed as

follows:

$$\% \text{ Grain Perforations} = \left(\frac{\text{Number of Perforated Grains}}{\text{Total Number of Grains Sampled}} \right) \times 100$$

Data analysis

GraphPad Prism (version 7.03) was the statistical package used to analyze all the data obtained from this study. Shapiro-Wilk normality test was used for normality test of the data. Two-way ANOVA was employed for level of significance in percent repellencies of the extracts against *S. zeamais*, adult mortality and percent grain perforations. Significantly different means were separated by using Bonferroni's multiple comparisons test at 5% level of significance.

RESULTS

Repellency of ethanolic botanical extracts against *S. zeamais*

E. balsamifera had the highest repellency which ranged from 93.65 ± 3.72% to 100.00 ± 0.00%, while *M. hirtus* had the least (87.22 ± 2.42 to 90.00 ± 4.08%) at 24 HAE (Figure 1). Percentage repellencies of *L. inermis* varied between 87.05 ± 0.45 and 97.50 ± 2.50%. The repellency of *S. obtusifolia* at 25, 50 and 100 mgml⁻¹ was 92.50 ± 2.50, 93.93 ± 3.61 and 100.00 ± 0.00%, respectively at 24 HAE (Figure 1).

The varying concentrations of ethanolic extracts showed different degrees of repellency against the maize weevils within 24 h of exposure. At 25 mgml⁻¹, *E. balsamifera* had the highest repellency, followed by *S. obtusifolia*, *L. inermis* and finally *M. hirtus*. At 50 mgml⁻¹

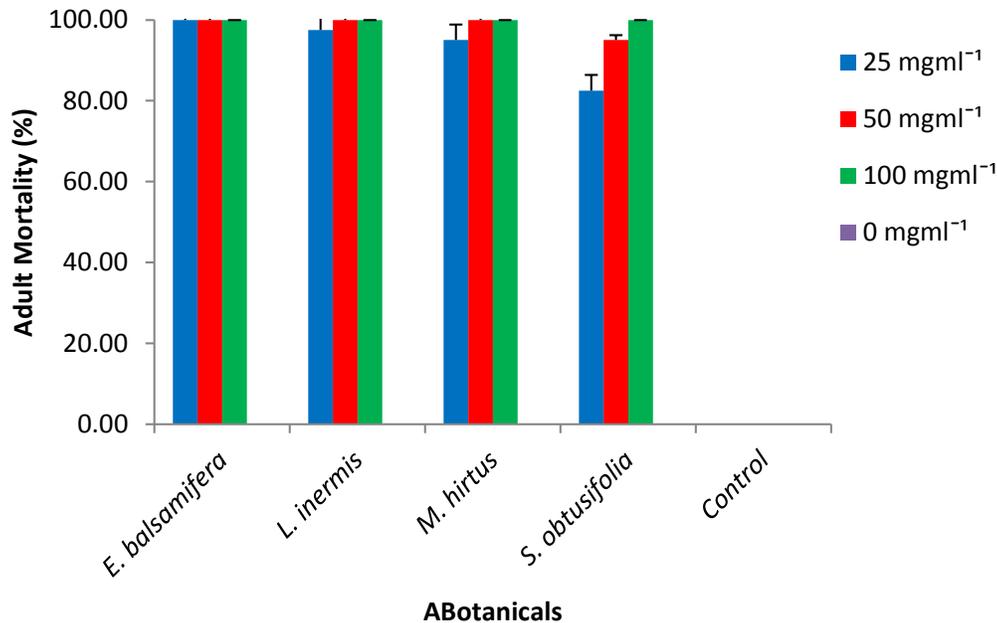


Figure 2. Mean percentage adult mortality of *S. zeamais* in sorghum grains treated with ethanolic extracts of different botanicals applied at different concentrations within 3 DAT.

of the botanical extracts, the decreasing order of their repellent action was *E. balsamifera* > *L. inermis* > *S. obtusifolia* > *M. hirtus*. At 100 mgml⁻¹, the percentage repellency was in the order *E. balsamifera* > *S. obtusifolia* > *L. inermis* > *M. hirtus*. There was no significant interaction between the ethanolic extracts and concentrations on the repellency levels ($F_{(6,36)} = 0.6139$; $p = 0.7176$). It however, showed that the repellency was significantly different among both the botanical types ($F_{(3,36)} = 8.513$; $p = 0.0002$) and the varying concentrations ($F_{(2,36)} = 7.553$; $p = 0.0018$) within 24 h.

Adult mortality of *S. zeamais* in sorghum grains treated with ethanolic leaf extracts

Ethanolic extracts of the selected botanicals applied at different concentrations of 25, 50 and 100 mgml⁻¹ exhibited varying percentage mortalities of *S. zeamais* within 3 DAT. Treating the grains with *E. balsamifera* resulted in total (100.00 ± 0.00%) adult mortality of the weevils at all the three concentrations within 3 DAT (Figure 2). This was followed by treatments with *L. inermis* and *M. hirtus* where 97.50 ± 2.50 and 95.00 ± 2.89% were respectively recorded at 25 mgml⁻¹, while 100.00 ± 0.00% of the weevils died at both 50 and 100 mgml⁻¹. The adult mortalities recorded in grains treated with *S. obtusifolia* ranged from 82.50 ± 2.50 to 100.00 ± 0.00%. None of the weevils died in untreated grains at the end of 3 DAT (Figure 2). There was a significant difference in adult mortality of *S. zeamais* among the botanicals ($F_{(4,45)} = 2947$; $p = 0.0001$).

Emergence of adult *S. zeamais* in sorghum grains treated with ethanolic leaf extracts

There was no emergence of adult *S. zeamais* in sorghum grains treated with ethanolic extracts of the selected botanicals. However, the mean number of emerged weevils in untreated grains was 156.80 ± 5.41. Therefore, ethanolic extracts of all the botanicals at the three concentrations of 25, 50 and 100 mgml⁻¹ resulted in total (100%) inhibition rate in adult emergence of *S. zeamais* in sorghum grains.

Grain perforations in sorghum treated with ethanolic leaf extracts

Highest grain perforations (7.50 ± 1.44 to 10.00 ± 2.04%) in sorghum grains were obtained in *S. obtusifolia* treatments, while the least (2.50 ± 1.44 to 5.20 ± 0.00%) were in sorghum treated with *E. balsamifera*. Grain perforations in sorghum treated with *L. inermis* and *M. hirtus* ranged from 3.75 ± 1.25 to 6.25 ± 1.25% and 5.00 ± 2.04 to 8.75 ± 1.25%, respectively, but 66.25 ± 3.15% of the grains were perforated in untreated samples. The grain perforations in the different treatments appeared to be in the following order: Control > *S. obtusifolia* > *M. hirtus* > *L. inermis* > *E. balsamifera* and 25 > 50 > 100 mgml⁻¹ in terms of concentration (Figure 3). There was a highly significant difference ($F(4, 45) = 588.30$, $p < 0.0001$) in grain perforations among the treatments. Further, there was no significant difference in grain

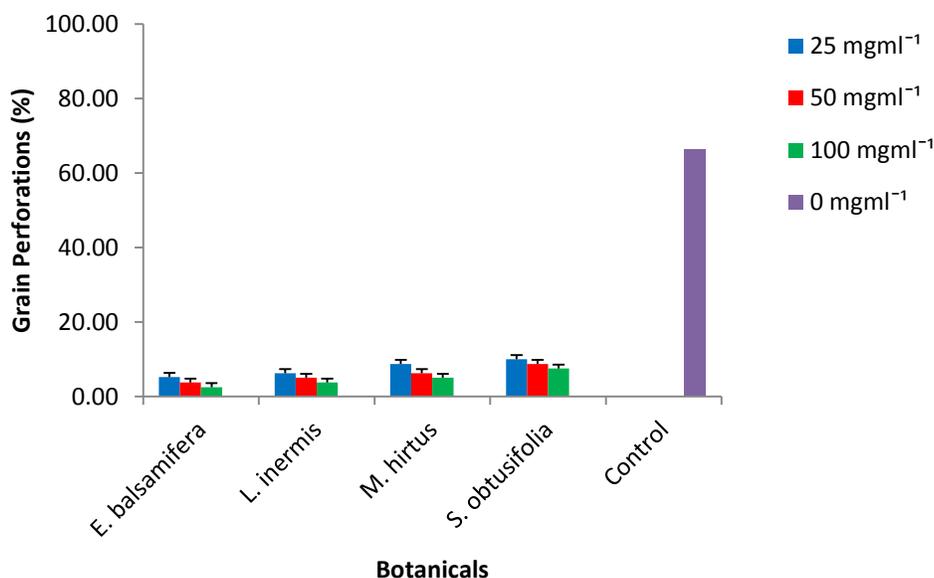


Figure 3. Mean grain perforations in sorghum grains treated with ethanolic extracts of different botanicals applied at different concentrations infested by *S. zeamais*.

perforations among the varying concentrations ($F(2, 45) = 1.71, p = 0.1924$).

DISCUSSION

Repellency of the botanicals against *S. zeamais*

All the botanicals, *E. balsamifera*, *L. inermis*, *M. hirtus* and *S. obtusifolia*, tested in this study have repellent potentiality against adult *S. zeamais* infesting sorghum. The present investigation has shown that the repellency of ethanolic extracts of the test botanicals against *S. zeamais* within 24 h was encouraging. The high repellent activity of ethanolic extract of *E. balsamifera* concurs with Idris et al. (2014) and Suleiman et al. (2018a) who recorded > 90.90% repellency of ethanolic and methanolic extracts of *E. balsamifera* against *Anopheles gambiae* and *S. zeamais*, respectively.

Similarly, the repellency potential of ethanolic extracts of *L. inermis* is in line with Datti and Idris (2013) that the extracts were highly repellent against mosquitoes. Application of the extract at 12.50% concentration on a marked area of forearm repelled 83.90% of *Anopheles gambiae*.

Findings of Akinbuluma et al. (2015) revealed the repellent effects of ethanolic extracts of *Capsicum frutescens* and *Dennettia tripetala* on *S. zeamais* where the botanicals repelled 91.68 and 89.58% of the weevils, respectively. These outcomes concur with Vigilanco et al. (2008) who earlier reported that ethanolic extracts of *Solanum argentinum* Bitter and Lillo repelled 55.0% of *S. oryzae*.

It is interesting to note that all the botanicals were found to demonstrate repellent activities against adult *S. zeamais* in sorghum grains. This shows that the plant materials could probably reduce *S. zeamais* infestations to sorghum by repelling the insect from the grains. The study revealed that *E. balsamifera* and *L. inermis* exhibited greater repellency against the weevils than the rest. The repellent activities of the selected botanicals could be attributed to the presence of non-host volatile odour components of plants with insects' repellent activities (Kuhns et al., 2016). Active ingredients such as alkaloids, flavonoids, saponins, phenolics and tannins, have been identified in some plant species and suggested to confuse the olfactory receptors so that the insects could not smell the host (Effiom et al., 2012). Further, Adesina et al. (2016) reported that secondary metabolites present in *Bridelia micrantha*, *Chasmanthera dependens* and *Vernonia cinerea* were responsible for repelling *Dysdercus superstitionis* (Herrich Schaffer).

Effect of the botanicals on adult mortality of *S. zeamais*

This study has revealed that leaves of *E. balsamifera*, *L. inermis*, *M. hirtus* and *S. obtusifolia* caused significant mortalities of adult *S. zeamais*. Ethanolic extracts of the selected botanicals were found to be highly effective by resulting in total mortality of adults of *S. zeamais* in sorghum grains at all concentrations of *E. balsamifera* and higher rates of *L. inermis*, *M. hirtus* and *S. obtusifolia*. The efficacy of the test botanicals concurs with earlier findings that ethanolic extracts of some plant

species caused significant adult mortality of maize weevils (Ajayi, 2013; Ibrahim et al., 2016). Also, findings of Ibrahim et al. (2016) showed that application of ethanolic leaf extract of *C. odorata* at 10 ml per 50 g maize caused 14.00% adult mortality of *S. zeamais* at 7 DAT. Findings of this study showed that the mortality of adult *S. zeamais* was directly proportional to the post treatment period agreeing with Ajayi (2013) and Ibrahim et al. (2016).

All the selected botanicals were effective against the weevil even at lower concentrations. This was possible because plant species contain secondary metabolites which are huge store of compounds such as the steroids, phenolic compounds, tannins, terpenoids, flavonoids, alkaloids, saponins and glycosides with wide range of biological activity reported to have great impact on insecticidal activities (Biswas et al., 2016; Hikal et al., 2017). The characteristic smell of the study botanicals might have also contributed to their insecticidal activity by repelling the insects away from the grains (Suleiman et al., 2018a). Ethanolic leaf extracts of *E. balsamifera*, *L. inermis*, *M. hirtus* and *S. obtusifolia* were generally found toxic to adult *S. zeamais* and suppressed their survival in sorghum grains by causing considerably high mortality.

Effect of the botanicals on adult emergence of *S. zeamais*

Ethanolic extracts of the test botanicals exhibited total inhibition in adult emergence of *S. zeamais* in treated sorghum grains as there was no adult emergence recorded. Application of plant powders to reduce adult emergence of *S. zeamais* was previously reported (Rivera et al., 2014; Oni and Ogunbite, 2015).

The total inhibition rate in adult emergence of the weevils in grains treated with ethanolic leaf extracts of the test botanicals at varying concentrations recorded in this study agrees with Ileke (2014) who reported a complete inhibition of adult emergence of *S. zeamais* when exposed to aqueous extracts of *A. boonei*. The suppression activity in adult emergence of *L. inermis* was also recorded against *C. maculatus* (Suleiman and Suleiman, 2014; Chudasama et al., 2015). It is found out that the botanicals might be lethal to the eggs deposited and as such led to inhibition in adult emergence of the weevils in the treated grains, concurring with Chudasama et al. (2015) that poisonous substances present in the extracts may enter into the egg through chorion and suppressed further embryonic development.

Influence of ethanolic extracts in reducing grain perforations in of stored sorghum infested by *S. zeamais*

The effectiveness of *E. balsamifera* and *L. inermis* in reducing grain perforations of sorghum by *S. zeamais*

corroborates with Suleiman and Suleiman (2014) who reported that application of leaf powders of the botanicals at 1.0 g / 20 g cowpea reduced perforations caused by *C. maculatus* from 18.33% in the control to 1.67%. Other botanicals were tested for their effectiveness in reducing grain perforations caused by *S. zeamais*. The weevil did not cause any grain perforation in stored wheat treated with powders of *A. indica* and *A. boonei* at 2.5, 5.0, 12.5 and 25.0% (Ileke and Oni, 2011). Similarly, no grains were perforated by *S. zeamais* 45 days after infestation when treated with 2.0 g *Corymbia citrodora* per 30 g of maize (Longe, 2016).

The significant reduction in grain perforations by the selected botanicals in this study could be due to total inhibition in emergence of the adult weevils in ethanolic extracts, since most of the perforations occurred as a result of emergence holes made by the insects. In addition to this, the secondary metabolites present in the botanicals might serve as antifeedants, and therefore the weevils found the treated grains unpalatable, hence reduction in perforations. This is supported by Dales (1996) and Rajashekar et al. (2012) that secondary compounds can affect insects by causing rapid death and acting as antifeedants, among others.

Conclusion

Findings have revealed that ethanolic extracts of *E. balsamifera*, *L. inermis*, *M. hirtus* and *S. obtusifolia* are repellents against *S. zeamais* in stored sorghum. The botanicals were found to be toxic to adult *S. zeamais*. The extracts were highly effective as adult emergence inhibition agents against *S. zeamais*. All the botanicals reduced grain perforations in sorghum at all the concentrations. The repellent activity, adult mortality effects, inhibition in adult emergence and reduction in grain perforations of the extracts indicated that the botanicals could be used as protectants of stored sorghum against *S. zeamais*, thereby contributing to IPM strategies. However, further investigations on their toxicity on mammals and other insect pests are hereby recommended.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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