Asian Journal of Agricultural Science 6(2): 62-68, 2014
DOI:10.19026/ajas.6.5304
ISSN: 2041-3882; e-ISSN: 2041-3890

Submitted: October 09, 2013 Accepted: October 22, 2013 Published: March 25, 2014

Research Article

Preservation of Cowpea (*Vigna unguiculata*) Seeds: Incidence of Ethanolic Extract from *Balanites aegyptiaca*, *Melia azedarach* and *Ocimum gratissimum* Leaves on *Callosobruchus maculatus* (Coleptera: Bruchidae)

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Abstract: This study aims to evaluate the effect of ethanolic extracts of *Balanites aegyptiaca*, *Melia azedarach* and *Ocimum gratissimum* leaves as bioinsecticide in the preservation of cowpea seeds (*Vigna unguiculata*) against the pest *Callosobruchus maculatus*. The extracts were obtained by maceration of leaves powdersin ethanol (95%). These extracts were then used to achieve anti-insecticides tests in jars at doses of 10, 15, 25 and 50% compared to a reference compound (Stargrain). The weevils rearing and some biological tests were conducted in laboratory conditions at a temperature of 29.1°C and a relative humidity of 74%. The results showed that these extracts have anti-oviposition activity and ovicidal dose-dependent. Doses 25 and 50% of *Melia azedarach* and *Ocimum gratissimum* have completely inhibited the spawning activity of *C. maculatus* 24 h after treatment. There was significant difference ($p<0.05$) between oviposition due to the positive control (Stargrain) and that due to dose 50% of the three extracts. Compared to the negative control (ethanol 95%), all these plant extracts have significantly reduced ($p<0.05$) oviposition of *C. maculates* female (38 eggs per 100 seeds after 4 days). The dose 10% of *B. aegyptiaca* and 50% of *O. gratissimum* showed no weevil emergency. It have been also noticed a reduction in seeds depreciation and rate attack in all the treated settings compared to the control. So the rate attack and the mass loss were proportional to the doses of B. *aegyptiaca* leaves extract but conversely proportional to doses of *Melia azedarach* and *O. gratissimum* leaves extracts. No depreciation of seeds was recorded at doses 10 and 15% of *B. aegyptiaca* and 50% of *O. gratissimum*. The different treatments did not affect the germination of seeds; the highest germination rate (93.67%) was recorded with seeds treated with the dose 50% of *O. gratissimum* against only 65% with those treated with the positive control. These ethanolic extracts have shown insecticidal effect against *C. maculatus* and can alternatively use for the cowpea seeds preservation.

Keywords: *Balanites aegyptiaca*, bio-insecticide, *Callosobruchus maculates*, *Ocimum gratissimum*, Oviposition, *Melia azedarach*, *Vigna unguiculata*

INTRODUCTION

Given the problems link to the use of synthetic pesticides such as the emergence of resistant insects, food poisoning, pollution of the environment (Elhag, 2000; Isman, 2000; Regnault-Roger, 2002) and the inadequacy of other farmers in preservation technics, the use of plants and their derivatives as alternative insecticides in protecting food grainers, dating very long, remains current (Bell *et al*., 1988; Stoll, 2000). Several studies have contributed to extract from certain plants, compounds whose insecticide properties side effects have not been reported in humans (Isman, 2000). Those plant extracts are biodegradable. Another advantage of these products is their low cost of production and their availability to farmer or small scale storage. Plant derivatives such as powders or extracts from leaves, seeds and bark influence pests activities through several mechanisms including toxicity on larvae and adult insects, egg-laying reduction, adult emergence inhibition and reduction of seeds impairment (Kiendrebeogo *et al*., 2006; Ngamo *et al*., 2007; Rajapakse and Ratnasekera, 2008; Epidi and Udo, 2009; Denloye *et al*., 2010).

This study aims to evaluate the effect of ethanolic extracts of some plants leaves (*Balanites aegyptiaca*, *Melia azedarach* and *Ocimum gratissimum*) as botanical insecticide for the preservation of cowpea (*Vigna unguiculata*) against *Callosobruchus maculatus*. The tests were carried out with the aim to estimating the

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oviposition, seed viability and damage caused by weevils on cowpea seeds treated with leaves extracts.

MATERIAL AND METHODS

Biological material and pretreatment:

Seeds sampling: The plant material used in this study consisted of dry seeds of *Vya* cultivar cowpea. They were bought from farmers in the Mayo-Kani division (Far North, Cameroon).Cowpea seeds were pretreated as performed by Koubala *et al*. (2013). After sorted and cleaned, the grains were stored in the freezer (-20°C) for 48 h before drying (sun) for 2 h. About 100 g of seeds were introduced into glass jars initially labelled. This procedure was done in triplicate.

Leaves sampling and extracts preparation: Leaves of *Balanite saegyptiaca*, *Melia azedarach* and *Ocimum gratissimum* were collected from the locality of Maroua (Far North, Cameroon) in the month of July. Ethanolic leaves extracts were obtained according to the procedure proposed by Koubala *et al*. (2013). The different fresh leaves were dried at the room temperature (26.8°C and humidity of 74%) for 7 days. The dried leaves were blended and sieved (0.25 mm) to obtain a powder.

Within 48 h, 50 g of the each powder was macerated under stirring in 100 mL of ethanol (95%). The mixture was filtered with What man filter paper (No. 4) and the filtrate was concentrated (60°C) in a rotary evaporator (Heidolph type) to obtain a dried extract which was stored (4°C).

Animal sampling: The animal parasites used were none sexed beans weevil (*Callosobruchus maculatus*) of 2 to 3 days old. A mass rearing was carried out with pairs of beans weevil from previously infested bunkers of cowpea section (Agriculture Research for Development Institute, Maroua). This breeding was done on cowpea grain at a temperature of 29.1°C and a relative humidity of 74% (Tapondjou *et al*., 2002). The obtained young animals (2 days old) were thus used for the study.

Bioassays:

Evaluation of weevil oviposition on treated grain: Four different doses of ethanolic extracts were previously prepared. About 100 g of seeds were uniformly coated with 5 mL of different doses (10, 15, 25, 50% w/v) of ethanolic extract by homogenization in 500 mL glass jars. As previously described by Koubala *et al*. (2013) seed jars negative and positive controls were, respectively treated with ethanol (95%) and with Deltamethrin (2 g per kg of seed). After evaporation of the solvent, an aspirator was used to infest each jar with 10 pairs of weevils (two days old). The jars were then covered with canvas gauze. The number of eggs laid on 100 grains was obtained by counting under a binocular microscope. This was done in each jar from different batches (42 jars each) every 24 h within a period of 4 days. This was used to assess the effect of the extracts on the weevil egg-laying.

Emergence of weevils and ovicidal activity of ethanolic extracts: The test media were left in laboratory conditions at an average temperature of 29.1°C and a relative humidity of 74% until emergence of weevil. The contents of the jars were regularly observed in order to determine the day of weevils emergence and deduce their development cycle according to the dose and treatment used (Delobel and Tran, 1993). In order to kill all adults and hidden juvenile forms, the experiment was stopped in an oven at 100°C for 1 h. After haven determined the number of emerging adults per jar, the emergence reduction ratio or emergence Inhibition Ratio (IR) was calculated according to the following equation:

$$IR\ (\%) = \frac{Cn - Tn}{Cn} \times 100$$

where, *Cn* and *Tn* are the number of weevils emerging from negative control and treated jars, respectively.

Estimation of the treated seeds protection: The attack and mass loss percentages are two most commonly used criteria for assessing seed damages. Given that N is a set of seeds (100 seeds) which is composed of healthy and attacked seeds, Nh the number of healthy seeds and Na the number of attacked seeds. The attack ratio (A %) was given by the following equation:

$$A\ (\%) = \frac{Na}{Nh + Na} \times 100$$

And the mass loss (M %) was also be determined by:

$$M\ (\%) = \frac{Mi - Mf}{Mi} \times 100$$

where, *Mi* and *Mf* are, respectively the initial (at the beginning of the text) and final (at the end of the text) mass of cowpea seed.

Estimation of the viability of the treated seeds: The viability of cowpea seeds processed and stored for 3 months was performed according to the method described by Rahman and Talukder (2006). Thus 25 cowpea seeds pretreated with leaves extracts or controls (negative and positive) were stored in plastic box for three months. They were then sown (Ns) and the number of germinated seed (Ng) was noted. Germination rate or viability percentage (V) was given by the following equation:

$$V\ (\%) = \frac{Ng}{Ns} \times 100$$

Data processing and statistical analysis: The data obtained from three replicates were subjected to Analysis of Variance (ANOVA) to compare the means for factor treatment. The Waller-Duncan test at 5% significance level was performed to determine differences between means. The software used was SPSS (Statistical Package for Social Sciences) version 10.0 for Windows.

RESULTS AND DISCUSSION

Effect of leaves extracts on the bruchidsoviposition: The number of eggs laid in jars treated with leave extract was used to assess the effect of these extracts on weevil eggs. On day 1, Fig. 1 shows that in areas treated with leave extracts from *B. aegyptiaca* spawning activity was 10.33% for the smallest dose against 4.00% for the high dose. We noted that the spawning activity of *C. maculatus* is conversely proportional to the dose of *B. aegyptiaca*, but this activity increases with time. This could be due to the increment of active substances present the extracts. It was also observed a highly significant difference ($p<0.05$) of rearing between treated and control media (18.67% of eggs laying on day 1 against 38.00% at day 4). The spawning activity in media treated with *M. azedarach* leave extract at doses 10 and 15%, is conversely proportional to the dose (Fig. 2). But at highest doses (25 and 50%), the number of laid eggs remains constant over the time ($p>0.05$). In jars treated with *O. gratissimum* leaves extract, there is no significant difference in spawning activity between media treated with lowest doses (10 and 15%) (Fig. 3). However, highest doses (25 and 50%) completely inhibit spawning activity of *C. maculates* for 24 h after that oviposition remains constant throughout the treatment period.

Generally, 96 hours after treatment, we observed that the number of laid eggs on the seeds is conversely proportional to the dose per hundred of seeds taken randomly. Compared to negative control (38.00%), all these ethanolic extracts reduce significantly ($p<0.05$) oviposition of *C. maculates* female. Dose 50% of these extracts is the most effective and it exhibit similar effect compared to the positive control (4.67%). This reduction of spawning activity may be due in part to the early death of weevils right after their introduction into the jars related to the toxicity of the extract. Indeed previous works (Koubala *et al*., 2013) showed that the ethanolic extracts from *Balanites aegyptiaca*, *Melia azedarach* and *Ocimum gratissimum* leaves had insecticidal effects on *C. maculatus*. Dose 50% of *O. gratissimum* caused 83.33% of mortality 24 h after treatment. On the other hand, the constituents of the extracts inhibit bruchids oviposition (Kiendrebeogo *et al*., 2006) by inducing ovarian changes similar to those caused by chemo-sterilizers which block spawning of bruchids (Schmidt *et al*., 1991). Similar results were obtained (Rajapakse and Ratnasekera, 2008) on the inhibition of oviposition of *C. maculatus* by the essential oils of *Ocimum sanctum* and

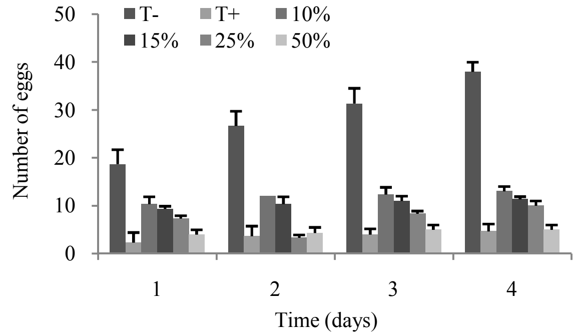

Fig. 1: Number of produced eggs on cowpea seed by *C. maculatus* adult in function of time at various doses (10-15-25-50%) of *B. aegyptiaca* leaves extract. A reference compound (Stargrain; T+) and ethanol (90%) (T-) were respectively used as positive and negative control

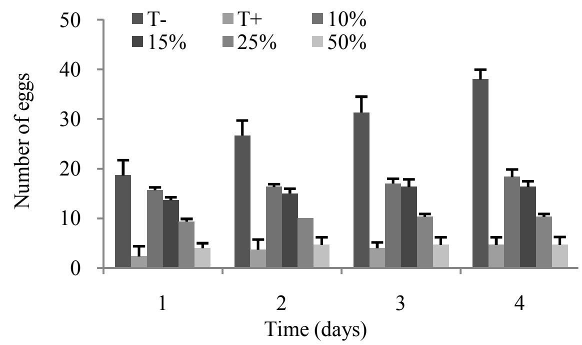

Fig. 2: Number of produced eggs on cowpea seed by *C. maculatus* adult in function of time at various doses (10-15-25-50%) of *M. azedarach* leaves extract. A reference compound (Stargrain; T+) and ethanol (90%) (T-) were respectively used as positive and negative control

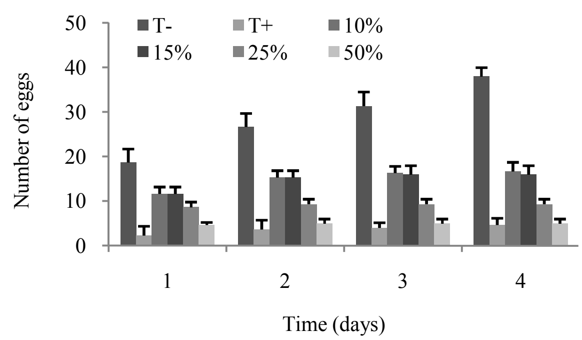

Fig. 3: Number of produced eggs on cowpea seed by *C. maculatus* adult in function of time at various doses (10-15-25-50%) of *O. gratissimum* leaves extract. A reference compound (Stargrain; T+) and ethanol (90%) (T-) were respectively used as positive and negative control

Table 1: Number of weeds emerged from cowpea seeds and seed onset ratio after treatment with ethanolic extract of *B. aegyptiaca*, *M. azedarach* and *O. gratissimum* leaves. Leave extracts were used at various doses (10-15-25-50%) compare to a reference compound (Stargrain; T+) and to ethanol (90%) (T-)

Extracts	Dose (%)	Number of weeds emerged	Percentage inhibition of weeds emergence (%)	Seed onset ratio after treatment (%)
Ethanol (90%)	10 to 50	179.33 ± 10.03^{a}	0	26.33 ± 3.06^{a}
Stargrain	10 to 50	3.00 ± 0.20^{c}	98.33	13.33 ± 3.06^{b}
B. aegyptiaca	10	0.00 ± 0.00^{d}	100	0.00 ± 0.00^{c}
	15	0.33 ± 0.08^{c}	99.82	0.00 ± 0.00^{d}
	25	10.67 ± 1.15^{bc}	94.05	2.67 ± 0.38^{c}
	50	28.00 ± 5.89^{b}	84.39	4.00 ± 1.00^{c}
M. azedarach	10	35.00 ± 5.57^{b}	80.48	13.33 ± 1.53^{b}
	15	31.67 ± 6.67^{b}	82.34	6.67 ± 0.58^{c}
	25	21.33 ± 5.86^{b}	88.11	3.00 ± 0.20^{c}
	50	7.00 ± 1.00^{c}	96.10	1.00 ± 0.23^{d}
O. gratissimum	10	17.67 ± 3.06^{c}	90.15	12.33 ± 2.52^{b}
	15	9.33 ± 1.53^{c}	94.80	9.00 ± 1.00^{c}
	25	1.67 ± 0.28^{c}	99.07	1.00 ± 0.20^{d}
	50	0.00 ± 0.00^{d}	100	0.00 ± 0.00^{e}

Mean values from triplicate measurements±standard deviation. Values in the same column followed by different superscripts are significantly different ($p<0.05$)

Anona reticulate. Larvicidal effects and anti-oviposition of fruits and leaves extracts from *M. azedarach* on *Aedesaegypti* have also been testified (Coria *et al*., 2008). In addition, ethanolic extracts from *O. sanctum* and *O. basilicum* have significantly reduced spawning activity of *C. chinensis* (Kiradoo and Srivastava, 2011).

Emergence in treated seeds: The number of individuals emerging from jars containing seeds (about 570 seeds/jar) treated with plant extracts is between 0 and 35 bruchids (Table 1). With extracts from *B. aegyptiaca* leaves, the number of weevils that comes from the seeds is proportional to the dose. With the dose of 10% no emergence was recorded. This gives a 100% reduction in adults emerging against 84.39% reduction with the dose 50%. Since it takes to much time to concentrated extracts at 50%, we can explain this by the denaturation of secondary active metabolites during the preparation of this extract (Schumutterer, 1990). However, leaves extracts from *M. azedarach* and *O. gratissimum* we note that the number of individual emerging is conversely proportional to the dose due to the concentration effect. No weevil emergence was recorded with the dose 50% of *O. gratissimum*. There was a significant difference ($p<0.05$) between data obtained from the negative control (179.33 weevils) and those from all other treated jars. We also notify that no significant difference was observed between treatment with synthetic insecticide (Stargrain) and those treated with extract at doses of 10 and 15% of *B. aegyptiaca*, dose 50% of *M. azedarach* and dose 15, 25 and 50% from *O. gratissimum* (Table 1). Because of their ability to reduce the emergence of weevils, these extracts have an ovicidal effect similar to that of the synthetic insecticides which can be replaced by the formers.

The way to coat the seeds with ethanolic extracts has as overall effect on the reduction of number of emerged individuals. This is a consequence of the adult weevils' mortality or the destruction of larvae and eggs initially produced after extracts contact. This ovicidal and/or larvicidal activity could be caused by volatile compounds coming from the extracts. Credland (1990) showed that these compounds could enter the egg through the breathing pores and cause death of the embryo. In the same way, non-volatile compounds could block these pores causing asphyxiation of the embryo. The insecticidal activity could also be due to the presence of secondary compounds (Kabaru and Gichia, 2001). Triterpenes are known for their diverse insecticidal properties, including ovicidal effect. This is the case of azadarachtine isolated from *Azadirachta indica* (Su and Mulla, 1998) or meliacarpine from *Melia azedarach* (Al-Rubae, 2009). Kiendrebeogo *et al*. (2006) showed that, an isolated fraction of *Striga hermontica* extract, mainly constituted of triterpenes, had an ovicidal effect (73%) on *C. maculatus*. In addition, the bruchids emergence time depends on the type of treatment applied to the seeds. Compare to the negative control (28 days), treatment with ethanolic extracts (24-27 days) exhibit an early onset of weevils. This is in contradiction with the works done by Kiendrebeogo *et al*. (2006) who found that the emergence in cowpea seeds treated with crude extracts of *Striga hermonthica* started with a delay of three days compare to the negative control. This could be related to the difference in chemical composition of the extracts used. Mbata and Ekpendu (1992) showed that alcoholic extract of *Piper guineense* preserved cowpea with 100% of ovicidal activity against *C. maculatus*. In the present study the same percentage of activity is obtained with the dose 10% of B. *aegyptiaca* and dose 50% of *O. gratissimum*. Because of their ability to reduce or prevent the emergence of bruchids, the evaluated ethanolic extracts can be used for the conservation of cowpea seeds.

Appearance of the treated seeds: Table 1 shows that seeds depreciation rate depend on the treatment used. At dose 10% of *B. aegyptiaca*, no seeds depreciation was recorded. However at dose 50%, 4% of seeds were

Table 2: Weight loss ratio and viability percentage ofcowpea seeds treated with ethanolic extract of *B. aegyptiaca*, *M. azedarach* and *O. gratissimum* leaves. Leave extracts were used at various doses (10-15-25-50%) compare to a reference compound (Stargrain; T+) and to ethanol (90%) (T-)

Extracts	Dose (%)	Seed weight loss ratio (%)	Seed viability percentage (%)
Ethanol (90%)	10 to 50	4.17 ± 0.65^{a}	83.50 ± 0.50^{a}
Stargrain	10 to 50	0.43 ± 0.06^{d}	65.00 ± 1.00^{d}
B. aegyptiaca	10	0.00 ± 0.00^{e}	71.00 ± 0.36^{c}
	15	0.00 ± 0.00^{d}	76.00 ± 0.00^{d}
	25	0.25 ± 0.05^{c}	79.33 ± 0.06^{c}
	50	1.07 ± 0.12^{b}	91.00 ± 0.33^{b}
M. azedarach	10	1.70 ± 0.12^{b}	71.67 ± 0.29^{c}
	15	1.21 ± 0.28^{b}	82.00 ± 1.00^{b}
	25	0.50 ± 0.10^{b}	89.67 ± 0.29^{a}
	50	0.07 ± 0.01^{d}	92.00 ± 0.50^{b}
O. gratissimum	10	0.90 ± 0.03^{c}	73.00 ± 0.00^{b}
	15	0.54 ± 0.14^{c}	80.50 ± 0.17^{c}
	25	0.14 ± 0.08^{c}	90.00 ± 1.32^{a}
	50	0.00 ± 0.00^{e}	93.67 ± 1.52^{a}

Mean values from triplicate measurements ± standard deviation. values in the same column followed by different superscripts are significantly different ($p<0.05$)

attacked. This suggests that the attack rate increases with the dose of the extract. On the other hand, the rate of attack is proportional to the doses of *M. azedarach* and *O. gratissimum* leaves extract (Table 1). We also noticed that seeds treated at dose 50% of *O. gratissimum* are not attacked while those treated with smallest dose (10%) are considerably attacked (12.33%). Treated seed exhibits significantly different ($p<0.05$) attack rates from one extract to another compared to the negative control (26.33%). It is also noted that the ethanolic extracts significantly reduce ($p<0.05$) seed loss mass more than the negative control which causes over 4.17% of mass loss (Table 2). Seeds treated at doses 10, 15 and 25% of *B. aegyptiaca*, doses 25% and 50% of *M. azedarach* and doses 15, 25 and 50% from *O. gratissimum* show similar mass losses compared to the positive control (0.43%). These doses can be used as an alternative in the fight against *C. maculatus*.

Results also show that the seed attack by weevils is closely related to the type of treatment applied to the dose. In agreement with Ngamo and Hance (2007), the attack rate of seeds is directly related to the number of eggs laid and the number of emerged weevils from seeds. Rajapakse and Ratnasekera (2008) have shown that oils from *Ocimum sanctum* and *Anona reticulata* prevent any cowpea seeds impairment after a development cycle of weevils. In addition, Epidi and Udo (2009) showed that ethanolic extracts from *Dracaenena arborea* prevent significant mass losses caused by *Sitophilus zeamais* and *Callosobruchus maculates* on maize and cowpea, respectively. These relatively small seed damages may be due to an important reduction of the number of individual emerging from treated seeds (Vijaya and Khader, 1990). Leaves extracts used in the present study have less bitter taste than leaves extract and seed oil from neem (*Azadirachta indica*) which were used in seed preservation because of their bitter taste (Rajapakse and Ratnasekera, 2008). This encourages the use of these extracts in the storage of cowpea seeds. Another advantage is the fact that these products are available, clean, biodegradable and non-polluant to the environment compared to chemical insecticides. It should be added that the extracts used in this study are from edible plant leaves commonly used in sauce.

Viability of the treated seeds: The test of viability carry on treated cowpea seeds showed that five days after seedling germination rates ranges from 71 to 93.67% in jars treated with leave extracts against 83.5% in those treated with the negative control (Table 2). Jars containing seeds treated with Stargrain (positive control) showed the lowest germination rate (65%). There is no significant difference ($p>0.05$) between seed viability from the negative control jars (83.50%) and from jars treated with doses 25 and 50% from *O. gratissimum*. It also appears that the viability increases with the doses extract. For example, with *O. gratissimum* the highest germination rate (93.67%) was recorded at the highest doses.

All these treatments do not affect the cowpea seeds germination (Table 2). In agreement with Adeniyi *et al*. (2010) the viability of seeds treated with ethanolic extracts from *O. gratissimum*, *S. acuta*, *T. occidentalis* and *V. amygdalina*is dose-dependent and the treatment used does not affect the quality of seeds. Keita *et al*. (2001) mentioned that the powders of *O. gratissimum* and *O. basilicum* allow full seeds protection against *C. maculatus* without significantly affecting their ability to germinate. Nevertheless, it would be convenient to test these plant extracts in combination with other means against pest in order to generate integrated methods.

CONCLUSION

It follows from this study that due to their ability to reduce or prevent the emergence of weevils (*Callosobruchus maculatus*) and to significantly reduce damages caused on treated cowpea seeds, the ethanolic extracts of *Balanites aegyptiaca*, *Melia azedarach* and *Ocimum gratissimum* leaves can be used in the preservation of cowpea seeds. Doses of 25 and 50% of *Melia azedarach* and *Ocimum gratissimum* completely inhibit egg-laying of *C. maculatus* 24 h after treatment. No weevil emergence is recorded at doses 10% of *B. aegyptiaca* and 50% of *O. gratissimum*. The attack in all treated areas are of lower rates compared to that of negative control ($p<0.05$). No seeds impairments are recorded at doses 10 and 15% of *B. aegyptiaca* and dose 50% of *O. gratissimum*. These treatments do not affect the viability of the seeds. With thedose of 50% from *O. gratissimum* we have the highest germination rate (93.67%) while positive control (Stargrain) causes only 65% of germination rate. These ethanolic extracts also have insecticidal effect against weevils and can therefore be used as an alternative for the preservation of seeds.

ACKNOWLEDGMENT

Our thanks go towards IRAD (Agriculture Research for Development Institute) of Maroua (Cameroon) for giving us weevil strains (*Callosobruchus maculatus*) and hot house for the viability tests of cowpea seeds.

REFERENCES

Adeniyi, S.A., C.L. Orjiekwe, J.E. Ehiagbonare and B.D. Arimah, 2010. Preliminary phytochemical analysis and insecticidal activity of ethanolic extracts of four tropical plant (*Vernonia amygdalina*, *Sidaacuta*, *Ocimum gratissimum* and *Telfaria occidentalis)* against beans weevil (*Acanthscelides obtectus*). Int. J. Phys. Sci., 5(6): 753-762.

Al-Rubae, A.Y., 2009. The potential uses of *Melia Azedarach* L. as pesticidal and medicinal plant. Am-Eurasian J. Sust. Agric., 3(2): 185-194.

Bell, A., O. Muck, P. Mutlu and H. Scheider, 1988. Integrated Protection Denrées stockéesestune Profitable Business! GTZ. Eschborn, Germany, pp: 37.

Coria, C., W. Almiron, G. Valladers, C. Carpinella, F. Luduena, M. Defago and S. Palacios, 2008. Larvicidal and oviposition deterrent effects of fruit and leaf extracts from *Melia azedarach* L. on *Aedes aegypti* (Dipetra: Culicidae). Bioresource Technol., 99(80): 3066-3070.

Credland, P.F., 1990. Biotype Variation En Host Plant Change in Bruchids: Cause and Effect in the Evolution of Bruchids Pests. In: Fujii, *et al*. (Ed.), Bruchids and Legumes: Economics Ecology and Coevolution. Kluwer Academic Publishers, The Netherlands, pp: 271-287.

Delobel, A. and M. Tran, 1993. Beetles food stored in hot areas. Tropical wildlife. 32 Edn., ORSTOM/CTA, Paris, pp: 24.

Denloye, A.A., W.A. Makanjuola, O.K. Teslim, O.A. Alafia, A.A. Kasali and A.O. Eshilokun, 2010. Toxicity *of Chenopodium ambrosioides* L. (Chenopodiaceae) products from Nigeria against three storage insects. J. Plant Protect. Res., 50(3): 379-384.

Elhag, E.A., 2000. Deterrent effects of some botanical products on oviposition of the cowpea bruchid *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae). Int. J. Pest Manage., 46(2): 109-113.

Epidi, T.T. and I.O. Udo, 2009. Biological activity of ethanolic extract fraction of *Dracaena arborea* against infestation of stored grains by two storage insect pest. Pak. J. Biol. Sci., 12(13): 976-980.

Isman, M.B., 2000. Plant essential oils for pest and disease management. Crop Prot., 19: 603-608.

Kabaru, J.M. and L. Gichia, 2001. Insecticidal activity of extracts derived from different parts of the mangrove tree *Rhizophoram ucronata* (rhizophoraceae) Lam. against three anthropoids. Afr. J. Sci. Tech., 2(2): 44-49.

Keita, S.M., C. Vincent, J.P. Schmit, J.T. Arnason and A. Belanger, 2001. Efficacy of essential oil of *Ocimum basilicum* L. and *O. gratissimum* L. applied as an insecticidal fumigant and powder to control *Callosobruchus maculatus* (Fab.) (Coleoptera: Bruchidae). J. Stored Prod. Res., 37(4): 339-349.

Kiendrebeogo, M., A.P. Ouedraogo and O.G. Nacoulma, 2006. Insecticidal activity of *Striga hermonthica* (Del.) Benth (Scrophulaceae) sur *Callosobruchus maculatus* (Fab.) (Coleoptera: Bruchidae). Biotechnol. Agron. Soc., 10(1): 17-23.

Kiradoo, M.M. and M. Srivastava, 2011. Insecticidal efficacy of some Lamiaceae plant extracts against *Callosobruchus chinensis* Linn. (Coleoptera: Bruchidae). Asian J. Agr. Sci., 3(2): 100-103.

Koubala, B.B., T.A.P. Miafo, D. Bouba, A.G.S. Kamda and G. Kansci, 2013. Evaluation of insecticide properties of ethanolic extract from *Balanites aegyptiaca*, *Melia azedarach* and *Ocimum gratissimum* leaves on *Callosobruchus maculatus* (Coleptera: Bruchidae). Asian J. Agr. Sci., 5(5): 93-101.

Mbata, G.N. and O.T. Ekpendu, 1992. The insecticidal action of four botanicals against three storage beetles. Med. Fac. Landbouww. Univ. Gent., 57(3A): 723-733.

Ngamo, L.S.T. and T. Hance, 2007. Variety of pests and commodities and alternative control methods in tropical. Tropicultura, 25(4): 215-220.

Ngamo, L.T.S., M.B. Ngassoum, P.M. Mapongmestsem, W.F. Noudjou, F. Malaisse, E. Haubruge, G. Lognay, H. Kouninki and T. Hance, 2007. Use of essential oils of aromatic plants as protectant of grains during storage. Agr. J., 2(2): 204-209.

Rahman, A. and F.A. Talukder, 2006. Bioefficacy of some plant derivatives that protect grain against the pulse beetle, *Callosobruchus maculates*. J. Insect Sci., 6(3): 1-10.

Rajapakse, R.H.S. and D. Ratnasekera, 2008. Pesticidal potential of some selected tropical plant against *Callosobruchus maculatus* (F) and *Callosobruchus chinensis* (L) (Coleoptera: Bruchidae). Trop. Agr. Res. Extension, 11: 69-71.

Regnault-Roger, C., 2002. New Phyto-insecticides for the Third Millennium. In: Philogène, B.J.R., C. Regnault-Roger and C. Coord Vincent (Eds.), Biopesticidesd'origine Plant. Éditions Lavoisier Tec and Doc, Paris, pp: 19-39.

Schmidt, G.H., E.M. Risha and A.K.W. El Nahal, 1991. Reduction of progeny of some stored product Coleoptera by vapours of *Acorus calamus*. J. Stored Prod. Res., 27(2): 121-127.

Schumutterer, H., 1990. Properties and potential of natural pesticides from the neem tree, *Azadirachta indica*. Ann. Rev. Entomol., 35: 271-297.

Stoll, G., 2000. Natural Crop Protection in the Tropics. CTA/AGRECOL, Margraf Verlag, Weikersheim, pp: 376.

Su, T. and M.S. Mulla, 1998. Ovicidal activity of neem product (azadirachtin) against *Culex tarsalis* and *Culex quinquefasciatus* (Diptera: Culicidae). J. Am. Mosquito. Contr., 14(2): 204-209.

Tapondjou, L.A., C. Adler, H. Bouda and D.A. Fontem, 2002. Efficacy of powder and essential oil from *Chenopodium ambrosioides* leaves as post-harvest grain protectants against six-stored product beetles. J. Stored Prod. Res., 38: 395-402.

Vijaya and Khader, 1990. *Pachymerus chinensis*, its biology and extent of damage to grains plant. Prot. Bull., 25: 23-28.