

## Suppression of Seed Beetle (*Callosobruchus maculatus*) Population with Root Bark Powder of *Zanthoxylum Zanthoxyloides* (Lam.) Waterm. (Rutaceae) on Cowpea (*Vigna unguiculata* (L.) Walp

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### ABSTRACT

*In view of the recently increased interest in developing botanical pesticides as an alternative to synthetic pesticides, this investigation was carried out to examine the insecticidal potentials of the Zanthoxylum Zanthoxyloides (Lam.) Waterm. (Rutaceae) Root Bark Powder against Seed Beetle, Callosobruchus maculatus (F.). The experiment was carried out at ambient temperature of  $28 \pm 3^{\circ}\text{C}$  and relative humidity of  $68 \pm 3\%$  in the laboratory. Population suppression activities of Z. zanthoxyloides Root Bark Powder against Seed Beetle was recorded at 5, 7 and 10 g/150 g cowpea grains laid out in completely randomized design replicated four times including the untreated control. Plant products applied at rates greater than 20% of the grain weight would not be economically viable in suppressing the population of insect pests. The root bark powder was applied to assess contact adult mortality, adult emergence, percentage weight loss and percentage grains damaged. Application rates of Z. zanthoxyloides were found to possess insecticidal activity against adult seed beetle. Topical application of the root bark powder caused significant ( $P < 0.05$ ) increase in adult mortality, reduced adult emergence, reduced weight loss of the grains and offered protection of cowpea grains against damage in small scale storage. Highest rate of treatment significantly increased adult mortality of C. maculatus compared to lower rates at 2 DAT. From the results, it could be concluded that the adults of C. maculatus were susceptible to Z. zanthoxyloides root bark powder. On the whole application of the plant material was better than no treatment at all. Such findings would be helpful in promoting research aimed at the development of new agents for C. maculatus control. The plant powder could also, be incorporated in Integrated Pest Management approach against C. maculatus in storage.*

Keywords: Beetle, Coleoptera, Emergence, Plant Material, Powder, Survival

### INTRODUCTION

The food security of any nation depends, largely on adequate storage of farm produce (e.g., seeds) against insect infestation. Coulibaly *et al.*, (2010) opines that cowpea seeds have the potential to contribute to food security and to poverty reduction in West Africa. Insect pests constitute the most visible and important constraint to cowpea production in most parts of Nigeria, infesting mature pods and accounting for post-harvest reduction of seeds. Thus, it is essential to protect the seeds from storage

insect pests because of their benefits in consumption, planting and sale. Cowpea, *Vigna unguiculata* (L.) Walpers, is a common food crop throughout Nigeria but particularly in the middle belt and drier northern regions (Ojuederie *et al.*, 2009). Sub-Saharan Africa accounts for over 70% of cowpea produced and consumed worldwide (Coulibaly *et al.*, 2010). Most often the seeds and pods of cowpea are boiled and eaten as vegetable or are consumed after cooking to provide protein and several vitamins and minerals while the leaves or stems serve as fodder for livestock feeding. In Nigeria, cowpea is consumed in the form of bean pudding, bean cake, baked beans, fried beans, bean soup among others (Mbah and Silas, 2007). High protein contents and lysine contents make cowpea a natural supplement to staple diets of cereals, roots and tubers commonly grown in many poor countries (Adekola and Oluleye, 2007). The research on cowpea is rapidly increasing due to population explosion, food shortage and demand for plant protein. However, insects are by far the largest group of crop pests that cause damage to the crop both in the field and in the store (Stansly, 2011).

Seed beetle, *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae), attacks cowpea pods in the field and continue in stored seeds reducing their nutritional value. The female deposits eggs singly on the surface of cowpea seeds. On hatching, the larvae penetrates the testa and remain in the seeds until fully developed. Adults emerge out of the pupae in about a month (Ofuya and Lale, 2001). The use of conventional synthetic insecticides has played a major role in the control of the pest at all stages of development. However, there are problems associated with the use of toxic synthetic pesticides which include human and eco-toxicity (Ukeh, 2008), while their increased use has been linked with the problems of bio-magnification, resurgence and the development of insecticide tolerant strains of pest species (Singh, 2011). There is therefore, a pressing need for the development of safer, alternative crop protectants such as botanical insecticides, which undergo biodegradation rapidly and do not contaminate the environment. The search for alternative sources for the containment of storage insect pests has continued in order to discourage the use of poisonous insecticides. Generally, botanical insecticides cause less damage to human and environmental health than conventional insecticides (Ukeh, 2009).

In Nigeria, candlewood tree, *Zanthoxylum* species are common components of the rainforest vegetation. The tree is a morphologically variable plant specie occurring throughout southern Nigeria. It is traditionally used for the treatment of stomach-ache, urinary and venereal diseases and rheumatism (Ogunwolu 1998). *Zanthoxylum zanthoxyloides* (Lam.) Waterm. also known as *Fagara zanthoxyloides* is an indigenous plant used widely as chewing stick for tooth cleaning in West Africa (Adebiji *et al.*, 2009; Adegbolagun and Olukemi, 2010). This present study was undertaken to investigate the insecticidal effects of *Z. zanthoxyloides* root bark powder in the suppression of seed beetle, *C. maculatus*, population and damages caused in stored cowpea.

## MATERIALS AND METHODS

### Insect Culturing

Adults of *C. maculatus* were collected from infested stock of cowpea seeds in the insectary of Nigerian Stored Products Research Institute (NSPRI), Asa Dam Road, Ilorin, Nigeria. About 50 unsexed bruchids were used to infest about 300 g cowpea seeds in a 500 ml Kilner jar covered with white muslin cloth. The jar had previously been sterilized in an oven at 50° C for 2 h. After being allowed 2 weeks of oviposition, the parents adults were sieved and killed by freezing. New generation of beetles that emerged after 32 days were used in this experiment according to the procedure of Savon *et al.*, 2010.

### Collection and Preparation of Plant Material

*Zanthoxylum zanthoxyloides* root was bought from a local market, Oja-Oba, Ilorin, Kwara State. The root bark was removed with the aid of razor blade, cut into smaller pieces and air-dried for 7 days on the verandah of the laboratory but covered at night to prevent dew. The pieces of *Z. zanthoxyloides* root bark were ground in a mortar and pestle into powder and then passed through 0.1 mm mesh sieve to standardize particles. The fine powder was stored in a black polythene bag and kept in the dark until required for use about 24 hours later.

### Source and Preparation of Cowpea Grains

Cowpea variety (IAR 48) obtained from the National Seed Council, Ilorin, Nigeria, has brown, smooth and irregular grains. The grains were disinfested, after sorting and winnowing, in the freezer compartment of a refrigerator at -4°C. After 10 days of freezing, the black polythene bag containing the grains was emptied on a laboratory desk in the open air to allow the seeds to thaw for 72 hours before use.

### Experimental Protocol

Cowpea grains were weighed into separate clean cylindrical plastic containers of 9 cm in diameter and 15 cm in height. *Zanthoxylum zanthoxyloides* root bark powder was applied at the rates of 5, 7 and 10 g/150 g cowpea grains and the mixture were thoroughly agitated to ensure even spread of the powder. Ten pairs of adults of *C. maculatus* were introduced into each container; the open end of the containers was covered with white muslin cloth fastened in place with a rubber band to prevent escape and suffocation of the bruchids. All treatments were arranged in completely randomized design (CRD) replicated four times including the untreated control. The experimental set-up took place at prevailing temperature (28±3° C) and relative humidity (68±3%).

### Data Collection

Data were collected on various parameters including adult mortality, emergence, percentage grain weight loss and grain damage. Adult mortality was scored after counts of dead beetles at 1, 3 and 5 days after treatment (DAT). A beetle was considered dead, if it remained still after being probed with an insect pin on the abdomen. Surviving and dead beetles were removed and Discarded at 7 days After Treatment (DAT)

when they would have laid eggs and to prevent overlap with subsequent first filial generation ( $F_1$ ).

A keen observation showed early adult emergence in the untreated control. Cowpea grains treated with *Z. zanthoxyloides* root bark powder were assessed for the emergence of the first filial generation ( $F_1$ ) at 30 DAT. Adults that emerged subsequently were removed and discarded to prevent overlap with second filial generation ( $F_2$ ). Counts of the number of emerged adults for the second filial generation ( $F_2$ ) were recorded at 60 DAT.

In scoring for the percentage grain damaged, samples of 100 g grains were taken from all the treatments and the number of damaged grains (grains with characteristic holes) and undamaged grains were counted and weighed. Percentage damage was calculated using the method of Food and Agriculture Organisation (1985).

The Percentage Grain Weight Loss (PGWL) was determined by the difference between the initial weight and final weight divided by the initial weight multiplied by 100.

#### Statistical Analysis

Data collected were subjected to analysis of variance (ANOVA) using Statistical Package of Social Science (SPSS) version 11.0. Significantly different means were separated by Least Significant Difference (LSD) test at  $P=0.05$

## RESULTS

#### Effects of *Z. zanthoxyloides* on Survival of *C. maculatus*

Table 1 shows that *Z. zanthoxyloides* root bark powder applied at the highest rate of 10 g/150 g grains was most effective in causing adult mortality of *C. maculatus* with 9.00 and 12.00 mean dead adults representing 45% and 60% mortality at 3 and 5 DAT respectively. During the same period, 7 g/150 g grains gave 7.25 and 10.25 mean dead adults representing 36.25% and 51.25% respectively. The rates of mortality in the treatments were significantly different ( $P<0.05$ ) compared to the untreated control which recorded no mortality. The least effective treatment was applied at the rate of 5 g/150 g grains which showed no significant difference ( $P>0.05$ ) compared to other rates of treatment except the control at 5 DAT. However, the various rates of treatment were significantly different from one another and the control at 3 DAT. Adult beetle mortality varied with the rates of treatment and exposure time.

#### Effect of *Z. zanthoxyloides* root bark powder on emergence

Treatments applied at the rates of 5, 7 and 10 g/150 g grains gave 1.00, 1.00 and 0.25 mean emerged adults at 30 DAT respectively. Significantly ( $P<0.05$ ) lower numbers of emerged adults were recorded at different rates at first filial generation ( $F_1$ ) compared to the untreated control (7.25) (Table 1). Treatments applied at the rates of 5, 7 and 10 g/150 g grains gave 1.75, 1.00 and 1.00 mean emerged adults at 60 DAT respectively. Similarly, significantly lower numbers of emerged adults were recorded at different rates at

second filial generation (F<sub>2</sub>) compared to the untreated control (8.50) (Table 1). The different treatments significantly ( $P < 0.05$ ) reduced adult emergence of *C. maculatus*. Each rate of the *Z. zanthoxyloides* plant powder reduced emergence significantly when compared to the control (Table 1).

Effects of *Z. zanthoxyloides* root bark powder on seed damage/seed weight loss  
Treatments applied at the rates of 5, 7 and 10 g/150 g gave grain weight loss of 3.72%, 3.51% and 2.89% respectively which were significantly different compared to the grain weight loss of 14.88% recorded in the control. There was appreciable reduction in cowpea grain weight loss in those grains treated with *Z. zanthoxyloides* root bark powder when compared to the control. Treatments applied at the rates of 5, 7 and 10 g/150 g gave grain damage of 11.69%, 3.34% and 1.00% respectively which were significantly different compared to the grain damage of 20.67% recorded in the control. Even the grains treated with *Z. zanthoxyloides* root bark powder were riddled with holes showing some degree of damage (Table 1).

#### DISCUSSION

Findings in this investigation were determined by rates of treatment and exposure time. Increasing the rate of *Z. zanthoxyloides* root bark powder caused increase in adult mortality of the beetle, suggesting that the plant material will be required in large quantities to provide protection for bulk storage. *Z. zanthoxyloides* root bark powder is however, weakly toxic to the *C. maculatus* adults because its contact action could not induce 100% adult mortality within 24 h even at the highest rate of treatment. Its effectiveness is based on the fact that using the plant material is better than no control at all. The differences in number of dead adults recorded in the various treatments may be attributed to differences in the amount of insecticidal components. Among the components of *Z. zanthoxyloides* earlier reported is the presence of highly pungent secondary metabolites, Zanthoxylol, as a phenolic compound reputed for insecticidal activity (Wongo, 1998). Botanicals have shown potential for stored cowpea against bruchid attack (Dabire *et al.*, 2008).

Grains in storage are damaged by several species of insect pests, the commonest being beetles and moths, leading to loss in weight and seed quality (Udo, 2011). In this study, percentage grain weight loss of 14.9% was caused to cowpea as a result of *C. maculatus* infestation within three months storage period. This report contradicts higher grain loss reported by various researchers. IITA (1995) reported Africa estimate losses of stored grain at an average of 30% per annum. Musa (2007) reported that 42.2% of cowpea grains were lost to bean beetle within three months storage period. The difference might be ascribed to period/length of exposure, environmental factors and nutritional value of the cowpea variety. Various rates of *Z. zanthoxyloides* root bark powder reduced the number of holes riddled by *C. maculatus* in the cowpea grains. Severity of damage in stored untreated grains was significantly higher compared to

treated grains. This type of damage led to loss of aesthetic quality and economic value of the grains. Damage by insect pests on cowpea can be as high as 80–100% if not effectively controlled. Denloye *et al.* (2007) reported that once infestation is established, insect pests cause gradual and progressive damage leading to losses in weight, nutritional, organoleptic and aesthetic quality of stored grains. It had been demonstrated that *Z. zanthoxyloides* root bark powder was toxic to adults of *C. maculatus* and therefore protected cowpea from depredation by the pest (Ogunwolu and Odunlami, 1996). Several studies on the various effects of *Z. zanthoxyloides* extracts have been reported. For example, Patel *et al.* (2010) named the compound nitidine as the active agent in *Z. zanthoxyloides*. More recently, the potential of *Z. zanthoxyloides* leaf, bark and root extracts as a biopesticide for stored food protection has been reported (Udo, 2011).

The different rates of treatment could not afford 100% inhibition of adult emergence. However, each rate of the root bark powder appreciably reduced development to adult stage suggesting that it could afford protection of the seeds against infestation. The reduction in number of emerged adults may be linked to reduction in viability of eggs. Ogunwolu *et al.*, (1998) reported that short duration exposure of adult bruchids to *Z. zanthoxyloides* powder reduces their reproductive fitness. The effectiveness of various plant parts against stored product pests had earlier been reported by several researchers (Gendo *et al.*, 2008; Boussaada *et al.*, 2008; Manzoomi *et al.*, 2010). The present study shows that *Z. zanthoxyloides* root bark powder could be added to the list of botanical insecticides against seed beetle, *C. maculatus*. Thus, the plant powder could be incorporated in the integrated pest management approach designed against the insect pest.

#### CONCLUSION

The findings from this study indicated that *Z. zanthoxyloides* root bark powder afforded some degree of protection against *C. maculatus* population in stored cowpea grains and has also appreciably reduced cowpea grains damage. It could be concluded that *Z. zanthoxyloides* root bark powder has insecticidal potential against adults of *C. maculatus* and then could be used by resource-poor farmers who store small amounts of cowpea grains meant for consumption and next season planting. The plant material is biodegradable and available in the country compared to imported synthetic insecticides and its current use as tooth chewing stick depicts its non-toxicity to man.

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Table 1: Insecticidal Activity of *Zanthoxylum zanthoxyloides* Root Bark Powder on *Callosobruchus maculatus* Population and Cowpea Grain Damage

| Treatment (g)/150g grains | Mean              | Adult C. <i>maculatus</i> | Mortality          | Mean F <sub>1</sub> | Mean F <sub>2</sub> | % Weight loss      | %Grains damaged    |
|---------------------------|-------------------|---------------------------|--------------------|---------------------|---------------------|--------------------|--------------------|
|                           | 1 DAT             | 2 DAT                     | 3 DAT              | 30 DAT              | 60 DAT              |                    |                    |
| 10                        | 2.50 <sup>a</sup> | 9.00 <sup>a</sup>         | 12.00 <sup>a</sup> | 0.25 <sup>b</sup>   | 1.00 <sup>b</sup>   | 2.89 <sup>b</sup>  | 1.00 <sup>b</sup>  |
| 7                         | 2.50 <sup>a</sup> | 7.25 <sup>b</sup>         | 10.25 <sup>a</sup> | 1.00 <sup>b</sup>   | 1.00 <sup>b</sup>   | 3.51 <sup>b</sup>  | 3.34 <sup>b</sup>  |
| 5                         | 2.25 <sup>a</sup> | 5.50 <sup>c</sup>         | 8.75 <sup>a</sup>  | 1.00 <sup>b</sup>   | 1.75 <sup>b</sup>   | 3.72 <sup>b</sup>  | 11.76 <sup>b</sup> |
| 0                         | 0.00 <sup>b</sup> | 0.00 <sup>d</sup>         | 0.00 <sup>b</sup>  | 7.25 <sup>a</sup>   | 8.50 <sup>a</sup>   | 14.88 <sup>a</sup> | 20.67 <sup>a</sup> |
| .                         | 0.45              | 0.76                      | 0.81               | 0.69                | 0.49                | 1.59               | 0.85               |

Values with the same superscript in the same column are not significantly different at P=0.05 using Least Significant Difference (LSD) test

DAT Days After Treatment

F<sub>1</sub> First Filial Generation

F<sub>2</sub> Second Filial Generation

SE Standard Error

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