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Effects of various doses of Neem-Afri biopesticide on the mortality and emergence of adult *Callosobruchus Maculatus* (Fabricius) [Coleoptera: Chrysomelidae] reared on cowpea seeds

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ABSTRACT

Studies were conducted at the Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto, Nigeria, to determine the effects of various doses of commercial biopesticide, Neem-Afri, on the mortality and emergence of adult Callosobrucus maculatus (Fabricius). Thirty air-dried Cowpea, Vigna unguiculata (L.) Walp. Grains, treated with four (10.0, 5.0, 2.0 and 1.0 ml: 100 ml) doses of emulsify able biopesticide, were infested with five pairs of freshly emerged adult C. maculatus and were observed under ambient laboratory conditions. Results showed over 70% adult mortality in all the seeds treated with Neem-Afri biopesticide, and 100% inhibition in seeds treated with 10 ml dose of the commercial pesticide at 4 days and 30 days post treatment respectively.

Keywords: Biopesticides, Cowpea, *Callosobruchus maculatus*, Neem-Afri, Mortality, Emergence.

INTRODUCTION

The practice of using plant materials as biopesticides or medicines is well known. These products are generally referred to as Botanicals. As many as 2000 plant species are use globally in the control of insect pests, most of which are used as food or traditionally as medicines [1]. Local farmers in Nigeria use leaves, roots and stems of dozens plant species in the control of stored produce pests. The plant materials provide small-scale farmers with biodegradable, risk-free and inexpensive substitute for the control of pests [1,2,3]. However, the increased interest in the use of plant-derived pesticides for the control of both field and storage pests have led to the standardization in formulation of the plant based products, including Neem to produce more effective and risk free doses [4,5,6].

Different formulations of Neem, *Azadirachta indica* A. Juss (Fam.: Meliaceae) leaf powder and its extracts have been tested for their efficacy on stored produce pests either alone or in comparison with other botanicals both in the fields and under laboratory settings [7]. Laboratory studies have shown that Neem has both medicinal and pesticidal importance [3]. Neem provides extensive agro forestry products [8]. Neem had also proved antiseptic against fungi, protozoans, bacteria, virus and helminthes [8, 9]. Neem powder and extracts are effective to over 400 insect pests. Neem seed and leaf extracts contained an array of structurally related tetranortriterpenoids isomers collectively referred to as the azadirachtins that play a role as insect antifeedants and growth regulators [10]. Despite its potentials, reports showed general unavailability of Neem- or plant derived pesticides in local markets in Northern Nigeria, nether is the extent of this pesticide effect on stored insects is known to farmers [11].

Being it a major cowpea pest in Nigeria, the cowpea weevil, *C. maculatus*, is known to course as much as 80% damage depending on the storage period and method of preservation employed [12]. Considering the importance of cowpea and the loss it suffered due to *C. maculatus* infestation both before and during storage, this research is design to ascertain the rates of mortality and emergence of *C. maculatus* reared on cowpea treated with varying doses of Neem-Afri Biopesticide.

MATERIALS AND METHODS

This study was conducted at Zoology Laboratory, Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto under ambient temperature and relative humidity.

Insect Culture

Adults *C. maculatus* collected from Sokoto granary were introduced into a 500 ml jar and allowed to oviposit for four days before it was discarded. Egg bearing seeds were kept in the jar undisturbed until the emergence of fully developed adults. Culture was maintained until the emergence of F_5 adults, each time by reintroduction of the newly emerged adults into a new jar containing fresh cowpea seeds.

Test Material

Neem-Afri biopesticide was purchased at Abubakar Rimi Market Sabon Gari, Kano, Nigeria. It is a product of Adebar Trade and Industries Ltd, Maiduguri, Borno State. It contained 3000 ppm Azadirachtin, 0.4% Nimbin and 0.56% Salanin. 100 ml of distilled water was added to 1 ml of emulsify able pesticide to make up the recommended dose of the pesticide to distilled water in field spray. Treatment doses of 2.5, 5.0 and 10.0 ml each were also added to 100 ml of water in separate jars. Forty (40 g) of healthy untreated cowpea seeds were placed in glass jar containing 1:100 pesticide to distilled water formulation and allowed to soak for three minutes. Treated grain was removed and spread on a laboratory table and allowed to dry for 24 hours. Thirty (30) air dried seeds were randomly selected, weighed and placed in a Petri dish. Similar procedure was repeated for each of the treatment doses following Jayakumar [13].

Mortality Assessment

To study the effect of Neem-Afri biopesticide on adult *C. maculatus*, the modified procedure of Silassie [14] was adopted. Five pairs of day-old adult cowpea weevil were introduced into each of the Petri dishes containing treated and control cowpea seeds, and mortality was recorded at 12 hours intervals each time by picking out the dead insect and continued for 4 days (96 hours) post infestation. The weevils were considered dead when there was no response after proving the abdomen with a pin. The percentage insect control or corrected mortality was determined using Abbott [15] formula as follows:

$$\text{Corrected Mortality} = \left(\frac{\text{Pc - Pt}}{\text{Pc}} \right) \times 100$$

Where: Pt and Pc = Percent mortality in the treated and control samples respectively.

Adult Emergence/Inhibition Rate

Adult emergence was assessed by recording the number of emergent adults from both treated (At) and untreated or control (Ac), the counting was stopped after 30 days post infestation to avoid overlap of generation following Agona and Muyinza [16], Silassie [14] and Jayakumar [13]. The Percentage Reduction (PR) or Inhibition Rate in F_1 adults was determined from the recorded data using the formula provided in Jayakumar [13] as follows:

$$\text{Percentage Inhibition (PI)} = \left(\frac{\text{Ac - At}}{\text{Ac}} \right) \times 100$$

Data Analysis

Data were analyzed using SPSS Inc. version 17, 2006 respectively. To normalize the variances, data were Square Root Transformed before being subjected to analysis following Bland and Altman [17], and Agona and Muyinza

[16]. One way ANOVA was used to compare the difference in insect's mortality and number of emerged adult. Means separations were conducted using Duncan New Multiple Range Test at 5% significant level.

RESULTS

The result in Table 1 showed a progressive rise in mortality of adult *C. maculatus* from 24 to 94 hours in both treated and untreated cowpeas. Never the less, significantly highest mortality was recorded in seeds treated with higher doses of Neem-Afri biopesticide.

Table 1 Effects of various doses of Neem-Afri biopesticide on the mortality of bean weevil, *Callosobruchus maculatus* reared on treated cowpea seeds.

		Cumulative mean no. of dead adults±SE/post-exposure duration (Corrected Mortality %)			
Treatment	**Dose (ml)**	**24 hours**	**48 hours**	**72 hours**	**96 hours**
Neem-Afri	10.00	2.00^{cd}±0.58 (17.24)	3.33^{bc}±0.33 (20.00)	7.67^{cd}±0.67 (70.83)	9.67^{d}±0.58 (95.83)
	5.00	1.00^{abc}±0.58 (6.90)	2.67^{bcd}±1.45 (12.00)	6.67^{c}±0.67 (58.33)	9.00^{bcd}±0.58 (87.50)
	2.50	0.67^{abc}±0.33 (3.45)	2.00^{a}±0.58 (4.00)	3.33^{b}±0.33 (16.67)	8.00^{bc}±0.58 (75.00)
	1.00	0.67^{abc}±0.33 (3.45)	1.33^{a}±0.33 (0.00)	2.67^{b}±0.67 (8.33)	7.67^{b}±0.33 (70.83)
Control	0.00	0.33^{a}±0.33 (0.00)	1.67^{a}±0.33 (0.00)	2.00^{a}±0.58 (0.00)	2.00^{a}±0.58 (0.00)

****Means in the same column followed by similar alphabets are significantly the same (P≥0.05), DMRT (1951).*

Table 2 Effects of various doses of Neem-Afri Biopesticide on the emergence of bean weevil, *Callosobruchus maculatus* reared on treated cowpea seeds

		Cumulative mean no. of emerged adults±SE/post exposure duration (Inhibition (%)				
Treatment	Dose (ml)	**23 days**	**25 days**	**27 days**	**29 days**	**30 days**
Neem-Afri	10.0	0.00^{a}±0.00 (100.00)	0.00^{a}±0.00 (100.00)	0.00^{a}±0.00 (100.00)	0.00^{a}±0.00 (100.00)	0.00^{a}±0.00 (100.00)
	5.0	0.00^{a}±0.00 (100.00)	0.00^{a}±0.00 (100.00)	0.33^{a}±0.33 (98.88)	0.33^{a}±0.33 (99.22)	0.33^{a}±0.33 (99.45)
	2.5	0.33^{a}±0.33 (97.62)	0.33^{a}±0.33 (98.57)	1.00^{ab}±0.58 (96.63)	1.33^{ab}±0.33 (97.03)	1.33^{ab}±0.87 (97.79)
	1.0	2.00^{ab}±1.16 (85.71)	7.67^{b}±0.88 (67.14)	12.00^{c}±2.52 (59.55)	15.33^{c}±3.18 (64.34)	19.33^{b}±2.91 (67.96)
Control	0.00	14.00^{c}±6.08 (0.00)	23.33^{c}±4.86 (0.00)	29.67^{d}±5.81 (0.00)	43.00^{d}±7.77 (0.00)	60.33^{c}±11.32 (0.00)

****Means in the same column followed by similar alphabets are significantly the same (P≥0.05), DMRT (1951).*

From Table 2 above, 100% inhibition (0.00 emerged adults) was recorded in seeds treated with higher doses of Neem-Afri Biopesticide. Highest adult emergence was recorded in untreated cowpea seeds followed by those treated with lower doses (1.0 ml and 2.5 ml) of the biopesticide.

DISCUSSION

The results from the study clearly showed that the commercial pesticide, Neem-Afri, is effective in the control of cowpea weevil. The effects tend to increase with increase in pesticide concentration. The effects may not be unconnected with the composition of Neem as is reported by Gunasekaran and Anita [18] and Eleazu *et al*. [19].

Phytochemical analysis of Neem leaf revealed the presence of saponin, tannin, flavonoid, alkaloid and cyanogenic glucosides [19]. Biswas *et al*. [20] also reviewed the effect of the various compounds that were isolated from the seed, leaf and root powder and extracts of Neem and studied for their biological activities against insects and other animals. Azadirachtins when consumed will suppress the function of ecdysone, thus preventing moulting which fatally restricted larval growth. The presence of azadirachtins also trigger the development of an anti-peristaltic sensation in the insect's alimentary canal that ultimately prevents feeding. Sani *et al*. [21] studied the effects of Neem-Afri on the organoleptic qualities of treated cowpea, the result showed that it is acceptable by the general public.

CONCLUSION

From the above study, it is evident that the commercial Neem-Afri biopesticide is capable of reducing cowpea weevil, *C. maculatus* infestation in storage condition.

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