Full Length Research Paper

Evaluation of ethanol plant extract for protection of *Cola nitida* against kola weevils (*Balanogastris kolae* and *Sophrorhinus* spp) (Coleoptera: Curculionidae) in storage

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The kola weevils Balanogastris kolae and Sophrorhinus spp (Coleoptera: Curculionidae) are the most destructive of all kola pests. This study was designed to evaluate the protective properties of ethanol extracts of 5 plant materials each at 1x10³, 2.5x10³, 5x10³ and 1x10⁴ ppm, against the kola weevils on stored kolanuts. The development and emergence of adult weevils were assessed by counting newly emerged adult weevils at fortnightly intervals for 112 Days. The number of weevil exit holes on the kolanuts and the number of kolanuts with colour change in each treatment were also determined. The mean number of adult B. kolae emergence from the various extract treatments did not differ significantly from each other. However, none of the extract treatments compared effectively with the standard treatment (1.38 ± 0.25). A similar trend was observed for Sophorhinus spp, but emergence of adult weevils was extremely low $(0.03 \pm 0.13$ to $0.34 \pm 0.10)$, even for the control treatment (0.78 ± 0.14) . There was no significance difference in the mean number of weevil exit holes recorded for all the extracts at 2.5x10³, 5x10³ and 1x10⁴ ppm treatment levels. However, they all differed significantly (P < 0.05) from their control treatment (101.16 ± 11.26), but did not compare effectively with the standard treatment (8.16 ± 0.75). The mean number of kolanuts with colour changes recorded for the standard treatment (2.28 ± 0.18) differed only completely from the various treatment means of Cederela odorata (6.16 ± 0.39; 5.81 ± 0.38; 5.28 ± 0.30; 4.97 ± 0.16). Generally, there was no significant difference amongst the various extract treatments means, so none could be claimed to be superior to the other. The various extracts therefore could be proffered as alternatives to kola farmers, so as to reduce their total dependence on synthetic insecticides for kolanut storage. Storage of kolanuts at 2.5x10³ ppm was found adequate and recommended taking into consideration their general protective effectiveness of the kolanuts and for economic reasons.

Key words: Kolanuts, weevils, plant extracts, treatments, emergence, exit holes, colour change.

INTRODUCTION

Cola acuminata and *Cola nitida* (Schott and Endl) are the only edible species of kolanuts grown in commercial scale in Nigeria (Jacob, 1973). The cotyledons of the kolanuts are red, pink or white and nuts extracted from the same pod often have a particular colour. The colour, size, flavour and storage quality of the kolanuts often determine the price of the kolanuts in the market. The kola weevils *Balanogastris kolae* and *Sophrorhinus* spp are the most destructive of all kola pests (Ivbijaro, 1976a; Daramola, 1981, 1983). The weevils feed, oviposits and completes their life cycles entirely within the kolanuts thereby exposing the kolanuts to secondary invasion by micro organisms, especially fungi (Ivbijaro, 1976a; Daramola, 1981, 1983; Odebode, 1990). The female weevils lay eggs about 1 cm deep into the kolanuts or accesses other parts of the fruit through wounds and holes made by other insects such as *Ceratitis colae*. It

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may also get into the kolanuts through cracks on the husk created when the follicles dehisce before harvest. Incubation lasts for about 4-6 days. The larval stage takes 17-20 days within which time the larva feeds extensively on the kolanuts and reduces them to brown powdery mass. Pupation lasts for about 5-6 days. The average period from oviposition to the emergence of the adults of *B. kolae* is 29 days while it is 31 days for *Sophrorhinus* spp. There was no specific season for the reproduction of the weevils, which were noted to continue throughout the year on left-over kolanuts and kolanuts produced between the main harvest seasons (Alibert and Mallamaire, 1955; Daramola, 1974; NRI, 1996).

The use of physical and synthetic chemical means was suggested in the past as reliable method of controlling the kola weevils (Ivbijaro, 1976b). However, the non availability, high cost of synthetic pesticides and irradiation equipment, emergence of pesticide-resistant weevils, and the potential hazards posed by pesticides to the environment, have all necessitated the search for alternative control measures against the kola weevils (Anikwe and Ojelade, 2005). Moreover, kolanuts do not require further processing after skinning and curing before consumption and for that reason the use of synthetic insecticides no matter how minimal should be discouraged.

In Nigeria, many local plant products such as peppers, ashes, vegetable oils, citrus peals, pawpaw leaves, cedar, neem tree products etc have been used successfully for the control of insect pests of stored products. These alternative bio-pesticides have been adjudged safe, biodegradable and environmental/user friendly (Ivbijaro, 1983; Sowunmi and Akinnusi, 1983; Ogunwolu and Idowu, 1994; Okorie et al., 1990; Don-Pedro, 1985; Pereira, 1982; Su, 1977; Asogwa and Osisanya, 2000; 2003; Ofuya, 1986; Jackai, 1993).

The present study was designed to evaluate the protective properties of ethanol extracts of five plant materials against the kola weevils *B. kolae* (Desbr) and *Sophrorhinus* spp (Coleoptera: Curculionidae) on stored kolanuts.

MATERIALS AND METHODS

This research work was carried out at the Entomology Laboratory of the Cocoa Research Institute of Nigeria (CRIN), Ibadan, at a temperature of 28 \pm 3°C and 75 \pm 5% relative humidity.

Collection of the plant materials for extraction

The following plant materials: *Azadirachta indica (stem bark); Cedrela odorata (stem bark); Chrysophyllum albidum (stem bark), Khaya spp (stem bark),* and *Chromolaena odorata (leaf)* were collected from the Forestry Research Institute of Nigeria, Jericho-Ibadan.

The samples were chopped into bits and air- dried on side benches in the laboratory for one week and thereafter, oven-dried at 60°C for 6 h. The oven-dried materials were pulverized with a highspeed mill for subsequent use for the extraction.

Crude plant extraction procedures

The pulverized plant materials were weighed out and Soxhletextracted using absolute ethanol for minimum of 8 hours according to methods by Ofuya et al., (1992).

Collection of kola samples

Two baskets of fresh unskinned kolanuts were bought from Ogumakin market in Ogun State, Nigeria. The kolanuts were skinned, washed and cured for 72 hours in the Entomology laboratory before use.

Application of treatments

Twenty-five cured kolanuts were randomly sorted out into transparent plastic 1 litre bowls each containing four concentrations (1 x 10^3 , 2.5 x 10^3 , 5 x 10^3 and, 1 x 10^4 ppm) of each plant extract and left to soak for 12 hours. The control kolanuts were soaked in distilled water (0 ppm) for the same period of time. The standard treatment kolanuts for comparism were soaked in Cypermethrin 10 EC for 1 hour. Each of the treatments was replicated 4 times in a completely randomized design. The kolanuts were removed after the soaking period and placed in small flat baskets for excess water to drain off. The nuts were thereafter cured for 72 hours in order to reduce the moisture content to a minimal level.

Storage of the nuts

The 25 cured kolanuts were each placed in black light gauge polythene bag of dimension 42.5 x 21.0 cm and tied up and placed on the laboratory bench. All kolanuts were stored at the Entomology laboratory for 112 days.

Post storage assessment

The various treatments in separate polythene bags were sieved every 2 weeks in order to monitor the development and emergence of adult *Balanogastris kolae* and *Sophrorhinus* spp by direct counting of newly emerged adult weevils at fortnightly intervals for 112 Days Post Experimental Period (DPEP). The efficacy and suitability of the various treatments were also considered by determining the number of weevil exit holes on the kolanuts and the number of kolanuts with colour change in each treatment.

Statistics

The total number of newly emerged weevil, their exit holes and kolanuts with colour change in each of the 4 replicates were recorded at fortnightly intervals at 14, 28, 42, 56, 70, 84, 98, 112 DPEP. The total mean \pm S.E. (standard error of the mean) of all the 32 observations were computed out and data obtained were subjected to the analysis of variance (ANOVA) at P < 0.05.

RESULTS

The rate of development and emergence of the adult weevils (*B. kolae* and *Sophrorhinus* spp) generally decreased with increased concentrations of the various extracts (Tables 1 and 2). The development and emergence of *B. kolae* from the stored kolanuts treated with the various extracts were significantly lower (P < 0.05) than that re-

Conc. of plant extract (ppm).	Exposure periods (Days Post Experimental Period (DPEP)								
	14	28	42	56	70	84	98	112	Mean ± SE*
	Total number of adult <i>B. kolae</i> emergence**								
Cederela odorata									
1x10 ³	8	104	117	232	269	143	9	7	27.78 ^b ± 4.31
2.5x10 ³	7	89	106	198	218	134	11	2	23.91 ^{bc} ± 3.60
5x10 ³	0	58	101	164	189	120	7	0	19.94 ^{bc} ± 3.19
1x10 ⁴	0	46	67	157	176	112	5	0	17.5 ^{bc} ± 3.01
Khaya spp.									
1x10 ³	0	118	125	231	217	130	13	7	26.28 ^b ± 3.88
2.5x10 ³	0	97	98	224	162	119	11	5	22.38 ^{bc} ±3.47
5x10 ³	0	71	89	215	143	109	3	1	19.72 ^{bc} ±3.29
1x10 ⁴	0	48	64	139	94	85	2	0	13.5 ^{bc} ± 2.19
Azadirachta indica									
1x10 ³	5	69	117	266	134	79	13	4	21.47 ^{bc} ± 3.73
2.5x10 ³	2	56	94	167	119	70	1	3	16.0 ^{bc} ± 2.61
5x10 ³	0	46	81	147	109	53	1	2	13.72 ^{bc} ± 2.34
1x10 ⁴	0	29	74	109	102	47	1	1	11.34 ^{bc} ± 1.95
Chromolena odorata									
1x10 ³	3	93	105	240	250	58	11	4	23.88 ^{bc} ± 4.23
2.5x10 ³	1	64	88	229	185	53	7	3	19.69 ^{bc} ± 3.64
5x10 ³	0	51	77	214	151	47	2	1	16.97 ^{bc} ± 3.32
1x10 ⁴	0	37	58	201	130	40	2	0	14.63 ^{bc} ± 3.08
Chrysophyllum albidum									
1x10 ³	4	87	113	286	229	86	4	9	25.56 ^b ± 4.48
2.5x10 ³	1	66	103	279	166	65	3	9	21.63 ^{bc} ± 4.11
5x10 ³	0	53	85	249	150	54	1	3	18.59 ^{bc} ± 3.70
1x10 ⁴	0	27	74	213	105	41	1	1	14.44 ^{bc} ± 3.11
Control (0)	9	140	295	545	628	385	185	94	71.28 ^a ± 9.29
Standard (1x10 ²)	0	4	8	10	14	8	0	0	1.38 [°] ± 0.25

 Table 1. Effect of various ethanol plant extract treatments on the progress of Balanogastris kolae development and emergence from stored kolanuts

corded in the control treatment with a mean adult emergence of 71.28 \pm 9.29. The mean number of adult *B. kolae* emergence from the various extract treatments (1x10³, 2.5x10³, 5x10³ and 1x10⁴ ppm) did not differ significantly from each other (Table 1). However, none of the various extract treatments compared effectively with the standard treatment with a mean record of 1.38 \pm 0.25. A similar trend was observed for *Sophrorhinus* spp but emergence of adult weevils was extremely low (ranging from 0.03 \pm 0.13 to 0.34 \pm 0.10), even for the control treatment with a mean of 0.78 \pm 0.14 (Table 2).

The plant extract effectively suppressed the development and emergence of *B. kolae* within the first 42 days, which coincided with the second generation emergence of the weevils. This was followed by an unprecedented high population and increased number of exit holes created by the emergence of the subsequent generation of weevils from the 56th day (Tables 1 and 3). There was no significance difference in the mean number of weevil exit holes recorded for all the extracts at 2.5x10³ ppm, 5x10³ ppm, 1×10^4 ppm treatment levels. However, they all differed significantly (P < 0.05) from their control with a mean weevil exit holes of 101.16 ± 11.26, but did not compare effectively with the standard treatment (8.16 ± 0.75) (Table 3). At 1×10^3 ppm treatment level, only *C. odorata* with mean exit hole of 59.03 ± 7.60 differed significantly (P < 0.05) from their control, but values for all the extracts at that level did not differ from each other (Table 3).

The few colour changes noted on the stored kolanuts progressed steadily to the 42^{nd} day after which there were no further changes (Table 4). The colour changes on the nuts did not increase progressively with the increased concentration of the extracts applied. There was no significant difference between the various extract treatments and the control treatments with a mean colour change of 4.97 ± 0.41 . The mean number of colour changes recorded for the standard treatment (2.28 \pm 0.18) differed only completely from the various treatment means of *Cederela odorata* (6.16 \pm 0.39; 5.81 \pm 0.38;

Exposure periods (Days Post Experimental Period (DPEP)									
Conc. of plant extract	14	28	42	56	70	84	98	112	Mean± SE
(ppm)	То	tal nu	umbe	r of a	adult	Soph	orhi	<i>nus</i> sp	p. emergence**
Cederela odorata									
1x10 ³	2	3	2	4	0	0	0	0	0.34 ^b ± 0.10
2.5x10 ³	2	2	2	3	0	0	0	0	0.28 ^b ± 0.19
5x10 ³	0	2	2	3	0	0	0	0	0.22 ^b ± 0.19
1x10 ⁴	0	0	0	1	0	0	0	0	0.03 ^b ± 0.13
<i>Khaya</i> spp.									
1x10 ³	0	0	3	2	0	0	0	0	0.16 ^b ± 0.07
2.5x10 ³	0	1	2	1	0	0	0	0	0.13 ^b ± 0.06
5x10 ³	0	1	1	2	0	0	0	0	0.13 ^b ± 0.06
1x10 ⁴	0	0	1	3	0	0	0	0	0.13 ^b ± 0.06
Azadirachta indica									
1x10 ³	0	2	2	3	0	0	0	0	0.22 ^b ± 0.11
2.5x10 ³	0	3	2	1	0	0	0	0	0.19 ^b ± 0.08
5x10 ³	2	0	1	1	0	0	0	0	0.13 ^b ± 0.06
1x10 ⁴	0	0	1	1	0	0	0	0	0.06 ^b ± 0.04
Chromolena odorata									
1x10 ³	2	3	2	1	0	0	0	0	0.25 ^b ± 0.10
2.5x10 ³	1	1	2	3	0	0	0	0	0.22 ^b ± 0.87
5x10 ³	0	3	1	1	0	0	0	0	0.16 ^b ± 0.07
1x10 ⁴	0	0	2	1	0	0	0	0	0.09 ^b ± 0.05
Chrysophyllum albidum									
1x10 ³	2	1	2	2	0	0	0	0	0.22 ^b ± 0.09
2.5x10 ³	1	1	2	1	0	0	0	0	0.16 ^b ± 0.07
5x10 ³	0	0	1	1	0	0	0	0	0.06 ^b ± 0.04
1x10 ⁴	0	0	1	1	0	0	0	0	0.06 ^b ± 0.04
Control (0)	2	3	7	6	4	3	0	0	0.78 ^ª ± 0.14
Standard (1x10 ²)	0	0	0	0	0	0	0	0	$0.00^{b} \pm 0$

Table 2. Effect of various ethanol plant extract treatments on the progress of Sophorhinus spp.development and emergence from stored kolanuts.

*Means with the same superscript are not significantly different (P > 0.05) by Tukey's test **Each value represents a total of four replicates

5.28 ± 0.30; 4.97 ± 0.16) (Table 4).

The plant extracts at 2.5×10^3 ppm treatment level generally conferred good protective ability on the kolanuts against the weevil effects. It reduced tremendously the progress of weevil development and emergence, which were not significantly different from the effects of the extracts at higher treatment levels (5×10^3 and 1×10^4 ppm) (Tables 1, 2 and 3).

DISCUSSION

The kola weevils are said to be "field to store pests" as their infestation is initiated in the field and persists in storage (Daramola and Ivbijaro, 1975). All the kola trees in Africa are believed to be infested with a significant infestation of 30-70% which can be as high as 100% in some cases of late harvest in Cote D'Ivoire, Guinea and Nigeria (Alibert and Mallamaire, 1955; Goormans and Pujol, 1955; Daramola, 1973; Daramola and Ivbijaro, 1975). According to Daramola and Taylor (1975), the havoc caused by these insect pests approximately claims 60% of the total kolanut production in Nigeria.

The plant extracts generally suppressed the weevil oviposition and progeny emergence from the stored kolanuts in the first 42 DPEP. However, there was an increased number of exit holes and high population of the weevil emergence recorded from 56 DPEP due to the breakdown of the protective properties of the extracts.

The few colour changes observed on the kolanuts could have been as a result of physiological factors associated with storing fresh kolanuts with polythene materials and not from the plant extracts or standard insecticide used.

Plant extracts are slow acting and degrade easily in the environment. Earlier research findings therefore recommended their application at high rates and at an increased frequency to achieve effective pest control (Golob et al., 1982; Sharaby, 1988; Ofuya et al., 1992; Ewete et al.,

Conc. of plant extract (ppm).	Exposure periods (days post experimental period (DPEP)								
	14	28	42	56	70	84	98	112	Mean ± SE*
	Total number of weevil exit holes on the kolanut**								
Cederela odorata									
1x10 ³	8	77	108	230	512	512	512	512	77.22 ^{ab} ± 9.75
2.5x10 ³	7	65	82	190	387	387	387	387	59.13 ^{bc} ± 7.29
5x10 ³	0	49	78	183	364	364	364	364	55.19 ^{bc} ± 6.89
1x10 ⁴	0	39	73	173	283	283	283	283	44.28 ^{bcd} ± 5.33
<i>Khaya</i> spp									
1x10 ³	0	101	121	242	469	469	469	469	73.13 ^{abc} ± 8.51
2.5x10 ³	0	80	113	220	349	349	349	349	56.53 ^{bc} ± 6.13
5x10 ³	0	57	82	208	316	316	316	316	50.34 ^{bc} ±5.74
1×10 ⁴	0	40	63	158	265	265	265	265	41.28 ^{bc} ±4.90
Azadirachta indica									
1x10 ³	4	72	97	285	460	460	460	460	71.81 ^{abc} ± 8.54
2.5x10 ³	2	58	86	173	320	320	320	320	49.34 ^{bc} ±5.84
5x10 ³	0	30	46	165	280	280	280	280	42.53 ^{bcd} ± 5.43
1x10 ⁴	0	20	39	130	239	239	239	239	35.78 ^{cd} ± 4.69
Chromolena odorata									
1x10 ³	2	35	70	226	389	389	389	389	59.03 ^{bc} ±7.60
2.5x10 ³	1	29	69	201	294	294	294	294	46.13 ^{bc} ± 5.55
5x10 ³	0	21	65	180	270	270	270	270	42.06 ^{bcd} ±5.16
1x10 ⁴	0	18	61	175	260	260	260	260	40.44 ^{bcd} ± 5.04
Chrysophyllum albidum									
1x10 ³	5	29	86	304	428	428	428	428	66.75 ^{abc} ± 8.23
2.5x10 ³	2	28	54	294	425	425	425	425	64.94 ^{abc} ± 8.42
5x10 ³	0	18	33	267	375	375	375	375	56.81 ^{bc} ± 7.56
1×10 ⁴	0	21	31	226	309	309	309	309	47.31 ^{bc} ±6.20
Control (0)	10	72	254	385	629	629	629	629	101.16 ^a ±11.26
Standard (1x10 ²)	2	23	23	38	45	45	45	45	8.16 ^d ± 0.75

Table 3. Effect of various ethanol plant extracts on the number of weevil exit holes on stored kolanuts

*Means with the same superscript are not significantly different (P > 0.05) by Tukey's test $\frac{1}{2}$

**Each value represents a total of four replicates

1996).

There are vast types of secondary metabolites in higher plants: acids, alcohols, aldehydes, alkaloids, esters, fatty acids, flavones, glycosides, hydrocarbons, lactones, nitrogen-containing compounds, sterols, phenols, and terpenoids, which confer pesticidal activity on them (Dales, 1996). Although this aspect was not investigated in the present study, it is logical to suggest that the reduction in population of kola weevils with these plant extracts may have been imparted by their bioactive chemical components. For example, Neem products contain, aza-dirachtin, salanin and nimbin, all known to have insecticidal properties, which could have been responsible for the reduced weevil progeny emergence observed and better kolanuts protection as compared to the no treatment control. Research should therefore be intensified on the extraction, isolation and identification of the various active ingredients in the various plant extracts with the aim of formulating them into insecticides. This

will provide a sustainable alternative control measure in low-input agriculture.

Conclusion

The application trials on stored kolanuts with the various plant extracts showed significant effects (P < 0.05) on the weevils as against their control. Generally, there was no significant difference amongst the various extract treatments means, so none could be claimed to be superior to the other. It is therefore recommended that the various extracts could be used by kola farmers for routine storage of kolanuts.

Storage of kolanuts at 2.5×10^3 ppm was found adequate and recommended taking into consideration their general protective effectiveness of the kolanuts against the weevils in storage and for economic reasons. The extracts were not too effective compared to the standard insecticide. However, due to their significant effects (P <

Conc. of plant extract (ppm)	Exp	Exposure periods (Days Post Experimental Period (DPEP)							
	14	28	42	56	70	84	98	112	Mean± SE*
		Тс	otal n	umbe	er of k	olanı	ut wit	h colo	ur change*
Cederela odorata									
1x10 ³	10	25	27	27	27	27	27	27	6.16 ^a ± 0.39
2.5x10 ³	10	20	26	26	26	26	26	26	5.81 ^{ab} ± 0.38
5x10 ³	11	21	22	23	23	23	23	23	5.28 ^{abcd} ± 0.30
1x10 ⁴	12	21	21	21	21	21	21	21	4.97 ^{abcde} ± 0.16
<i>Khaya</i> spp									
1x10 ³	2	8	18	18	18	18	18	18	3.69 ^{def} ± 0.42
2.5x10 ³	0	17	17	17	17	17	17	17	3.72 ^{def} ± 0.37
5x10 ³	0	17	17	17	17	17	17	17	3.72 ^{def} ± 0.29
1x10 ⁴	0	13	15	15	15	15	15	15	3.22 ^{ef} ± 0.29
Azadirachta indica									
1x10 ³	2	15	23	23	23	23	23	23	4.84 ^{abcde} ± 0.48
2.5x10 ³	0	17	17	17	17	17	17	17	3.72 ^{def} ± 0.26
5x10 ³	2	8	17	17	17	17	17	17	3.50 ^{de} ^f ± 0.37
1x10 ⁴	3	14	14	14	14	14	14	14	3.16 ^{ef} ± 0.22
Chromolena odorata									
1x10 ³	1	22	26	26	26	26	26	26	5.59 ^{abc} ± 0.44
2.5x10 ³	1	21	23	23	23	23	23	23	5.0 ^{abcde} ± 0.41
5x10 ³	1	18	18	18	18	18	18	18	3.97 ^{bcdef} ± 0.27
1x10 ⁴	0	11	20	20	20	20	20	20	4.09 ^{bcdef} ± 0.49
Chrysophyllum albidum									
1x10 ³	7	11	25	25	25	25	25	25	5.25 ^{abcd} ± 0.44
2.5x10 ³	4	11	16	22	22	22	22	22	4.38 ^{abcde} ± 0.46
5x10 ³	2	17	18	19	19	19	19	19	4.13 ^{bcdef} ± 0.28
1x10 ⁴	2	17	17	17	17	17	17	17	3.78 ^{cdef} ± 0.36
Control (0)	5	11	23	24	24	24	24	24	4.97 ^{abcde} ± 0.41
Standard (1x10 ²)	2	7	9	11	11	11	11	11	2.28 ^f ± 0.18

Table 4. Effect of various ethanol plant extracts on the number of stored kolanuts with colour change

*Means with the same superscript are not significantly different (P > 0.05) by Tukey's test.

**Each value represents a total of four replicates.

0.05) over their controls, they can be proffered as a temporary alternative to kola farmers, so as to reduce their total dependence on synthetic insecticides for kolanut storage.

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