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COMPARATIVE EFFICACY OF *Jatropha curcas* L. LEAF POWDER FOR THE CONTROL OF *Callosobruchus subinnotatus* (Pic) on STORED BAMBARA NUT, *Vigna subterranea* (L.) Verdcourt

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ABSTRACT
The research was conducted in the year 2016 at the Entomology laboratory of the Department of Crop Protection, Bayero University Kano to assess the biocidal and damage reduction effect of leaf powder from physic nut, Jatropha curcas L. caused by Callosobruchus subinnotatus (Pic) on stored bambara nut, Vigna subterranea (L.) Verdcourt. Also, the study aimed at comparing the effectiveness of the plant product with conventional chemical insecticide (pirimiphos-methyl). Factorial experiment of 4×2 levels (leaf powder at the rates of 0, 0.5. 1.0 and 1.5 g, with and without addition of the synthetic chemical at 0.01 g/20 g bambara nut seed) were laid out in a Completely Randomized Design and repeated three times. Results showed that, admixture of 0.5g leaf powder proved effective in causing high biocidal effect and reduced seed damage. However, this was statistically same with seeds treated with 1.0g leaf powder. Comparatively, all treatments were statistically different from the untreated control seeds but similar with the check treatment. Highly significant (p<0.001) persistent effect was observed in seeds treated with 1.5g leaf powder mixed with 0.5g seed powder at 60 days after treatment (DAT) without any negative effect on seed viability. This was also statistically similar in all other treatments apart from the untreated control. In conclusion, appreciable level of protection on bambara nut seeds was achieved using leaf and seed powder of J. curcas applied at different mixture rates and singly. Therefore, it is recommended that for effective management of C. subinnotatus infesting bambara nut, farmers could use 1.5 g leaf powder or 1.0 g seed powder per 20 g bambara nut seeds singly each. Alternatively, 0.5:1.0, 1.0:1.0 or 1.5:0.5 leaf/seed combinations could be used for safe bambara nut storage.
Keywords: Biocidal, Callosobruchus subinnotatus, bambara nut, Jatropha, damage, insecticide

INTRODUCTION

An underutilized legume, bambara nut, *Vigna subterrabea* L. (Verdcourt) was once the third most important grain legume after peanut and cowpea. Mainly grown by women for the sustenance of their families, bambara nut was cultivated in extreme tropical environments by peasant farmers who lacked access to irrigation and/or fertilizers and with little guidance on improved practices (Mukurumbira, 1985; Mwale *et al.*, 2007). In addition to human consumption and fodder for livestocks, bambara nut is used in traditional medicines as a remedy for many ailments in humans and livestock, which included *helminthosis*, *schistosomiasis*, leprosy, diarrhoea and *psoriasis* (Burkill, 1995). However, in spite of its usefulness and nutritional values, bambara nut, is reported to be threatened by the devastating activities of stored products pests, notably *Callosobruchus subinnotatus* Pic. (Lale and Vidal, 2003a).

Although, successes have been recorded with the use of synthetic insecticides, abuse and inappropriate use of the synthetic insecticides caused a lot of problems ranging from high costs to mammalian toxicity (Dike and Mshelia, 1997). In addition, the use of synthetic chemical insecticides reduced viability of seeds (Bamaiyi *et al.,* 2007). These serious limitations posed by the use of synthetic insecticides as preservatives during storage on one hand and losses caused by *C. subinnotatus* during storage on the other called for search of a new alternative method of controlling the pest. Recently, particular interest has been focused on the use of natural plant products because they are available locally, cheap, less hazardous and environmentally friendly as well as safe and easy to handle. Moreover, botanical pesticides are biodegradable thereby leaving no residual toxicity to man. However, little information exists on the insecticidal properties of the physic nut, *Jatropha curcas* L. (Euphorbiaceae) for the control of *C. subinnotatus* in stored bambara nut.

MATERIALS AND METHODS

Preparation of Bambara Nut Seeds

Fifty Kilogrammes of unshelled bambara nut, cream/brown eye variety seeds were purchased at a local market in Dambatta Local Government Area, Kano State. The seeds were decorticated manually. Shrivelled (shrunken) and damaged seeds as well as all other debris were removed. To disinfest the cleaned whole seeds, they were put in a polythene bag together with two phostoxin tablets (in an envelope) for 24 hours. The mouth of the bag was tied securely to ensure that any insect pest present within the seeds was killed according to the method of Ogunwolu *et al.,* (2002). Thereafter, the seeds were opened and spread in a shaded well ventilated place for 48 hours to ensure that the seeds were free from the phostoxin residue. To avoid subsequent re-infestation and to ensure that any insect pest that might still remain within the seeds was killed the previously fumigated seeds were transferred into a fresh and different polythene bag and kept at -4˚C inside a fridge for four days (Ahmed, 2007).

Collection of The *Jatropha curcas* Leaves

The leaves of the physic nut *J. curcas* were collected at the orchards of Audu Bako College of Agriculture, Dambatta, Kano State and identified as *Jatropha curcas* L. in the Department of Biological Sciences, Ahmadu Bello University, Zaria. The Actellic dust was purchased from a pesticide store at Abubakar Rimi market, Sabon-gari, Kano, Kano State, Nigeria.

Preparation of The *Jatropha curcas* Leaves

The leaves were dried in shade to crispy condition. Thereafter, it was pounded in a mortar with pestle and then passed through a sieve 40 µmm to give a very fine powder as described by Youdeowei (2004) and Yusuf and Ahmed (2005). The fine powdered plant materials were kept in plastic bags until needed.

Source and Rearing of Insect Culture

The initial culture of the bambara nut bruchids was obtained from naturally infested bambara nut seeds at Kurmi market, Kano city, Kano State, Nigeria. A sample of the insects on infested seeds was taken to the insectary of the Department of Crop Protection, Ahmadu Bello University, Zaria, Kaduna State, Nigeria for proper taxonomic identification as *Callosobruchus subinnotatus* (Pic.). These were then massively reared in transparent plastic buckets measuring 30 cm in height and 15 cm top diameter. The top end of the plastic buckets was covered with white muslin cloth, secured firmly into place with rubber bands, hence allowing for ventilation. These were incubated in Kliner jar at an ambient temperature and relative humidity (32±3^{o}C and 57±3% respectively) with alternating light and dark cycle for 12 hours as previously described by Abduljalal *et al.* (2011).

Experiment and Experimental Design

The experiment was conducted in the Crop Protection Laboratory I, Faculty of Agriculture, Bayero University Kano State (11°59'47"N, 8°31'0"E) (Kowal and Knabe, 1972) to assess the effect of the *J. curcas* leaf powder on biocide (adult mortality) of, and damage (number of eggs, adult emergence and holes as well as percentage seed damage, seed weight lost and germination) caused by *C. subinnotatus* on bambara nut. In a 4×2 factorial experiment, the leaf powder and pirimipos-methyl were assessed for the management of *C. subinnotatus* infesting stored bambara nut. The First factor (leaves of *J. curcas*) had four (4) levels (0, 0.5, 1.0 and 1.5 g/20 g seed) while the second factor (pirimiphos-methyl) had two levels (with and without) applied at the reduced standard rate of 0.01 g/20 g (Gwinner, *et al.,* 1996). There were eight (8) treatments, which were replicated three (3) times in a Completely Randomized Design. The treatments were admixed with the bambara nut and shaken vigorously after which, five pairs of freshly emerged adult *C. subinnotatus* were introduced into each treatment in plastic cups. A total of 24 transparent plastic cups measuring 10 cm in depth and 9 cm top diameter were kept in the laboratory at ambient temperature and relative humidity of 32±3^{o}C and 57±3%, respectively. The top of the plastic cups were covered with white muslin cloth held in place with rubber bands to secure it firmly.

Assessment of Potentials of the Plant Products

Number of Eggs Laid

The number of eggs laid was counted with the aid of hand lens at 14 days after treatment (DAT) (Aliyu and Ahmed, 2006) when all the introduced adult insects were dead and those that were still living removed (Appleby and Credland, 2001).

Number of Adult Emergence

When all eggs laid were expected to have hatched, the number of adult emergence was taken as the total number of adults that emerged considering the period of emergence from egg to adult to be from 34 – 42 days after oviposition as described by Mbata (1992). Adult *C. subinnotatus* that emerged were removed and recorded daily in all the treatments and replicates and their cumulative numbers were considered as F_I generation emergence.

Number of Emergence Holes

Number of emergence (exit) holes was assessed by counting the number of holes that appeared on each seed. This was conducted with the aid of a needle, which was used to standardize the holes in such a way that no hole was counted more than once. The seeds were turned upside down and from side to side to ensure that no holes were left uncounted as described by Aliyu and Ahmed (2006) and Abduljalal *et al.* (2011).

Adult Mortality

Assessment of adult mortality was carried out from the first day after treatment (DAT) and continued subsequently until all insects were dead by counting the number of insects that died daily as a result of the treatment applied as described by Lale and Yusuf (2001); and Yusuf and Ahmed (2007). Per cent adult mortality was determined as the number of dead insects divided by the total number of insects introduced, multiplied by 100.

$$\%\ Adult\ mortality = \frac{Number\ of\ dead\ insects}{Total\ number\ of\ insects\ introduced} \times 100$$

Data on percentage adult mortality was corrected using Abbot's (1925) formula.

$$Pr = \frac{Po - Pc}{100 - Pc}$$

Where,
Pr = Corrected mortality (%)
Po = Observed mortality (%)
Pc = Control mortality (%)

Percentage Seed Damage

This parameter was measured as the percentage of the difference between the number of seeds without holes (in each experimental unit) and the number of seeds with holes (in the same experimental unit) divided by the total number of seeds multiplied by 100 (Ogunwolu *et al.,*2002).

$$PSD = \frac{NGWH}{TLNG} \times 100$$

Where: PSD = Percentage seed damage;
NGWH = Number of grains with holes;
TLNG = Total number of grains.

Percentage Seed Weight Lost

This parameter was also measured as the percentage of the difference between the initial and final weight of the seeds divided by the initial weight as described by Golob *et al.* (1982)

$$PSWL = \frac{[UaN - (U + D)]}{UaN} \times \frac{100}{1}$$

Where: PSWL = Percentage seed weight loss
U = Weight of unaffected seeds in the sample
N = Total number of seeds in the sample
Ua = Average weight of unaffected seeds
D = Weight of affected seeds in the sample

Potency

Data on residual toxicity (potency of the leaf powder) were collected by counting the number of dead insects out of the first generation (F_1) progeny reproduced by the parents which were introduced into the treated seeds 60 days after treatment when all the introduced parents were dead and the off springs had emerged (Abduljalal *et al.,* 2011).

Data Analysis

Data in percentages were transformed into arc sine percentages, while numerical data with low or zero count were transformed using √n+1 before analysis, as described by Little (1978). The transformed data was subjected to One-way Analysis of Variance using computer software (SAS for windows) statistical package. Treatments with significant differences were compared and separated at 0.05% level of probability using Duncan's multiple range test (DMRT).

RESULTS

In Table 1, the effects of *J. curcas* leaf powder, with and without addition of pirimiphos-methyl was assessed on the oviposition, F_1 emergence of, and damage caused by *C. subinnotatus* on bambara nuts. Number of eggs laid by the bruchids on bambara nut admixed with different rates of the leaf powder, with and without the synthetic chemical, was significantly ($p<0.001$) lower than the number of eggs (37.42) observed on the untreated bambara seeds. Although statistically similar, the number of eggs (6.17 and 7.17) laid on bambara seeds treated with 1.0 and 1.5 g leaf powder without the pirimiphos-methyl, respectively, was lower than eggs (13.75 and 7.42) laid on bambara seeds admixed with the same treatments (1.0 and 1.5 g leaf powder) with addition of the pirimiphos-methyl each, respectively. In addition, rate of F_1 progeny emergence from seeds treated with the different concentrations of the leaf powder, with and without the pirimipos-methyl was significantly ($p<0.001$) less than the rate of emerged adults (30.20%) from the control treatment. Although insignificantly different, the rate of emerged adults (7.92%) in 1.0 g leaf powder treated seeds was lower than emerged adults (13.69%) in the same treatment (1.0 g leaf powder) with the addition of the synthetic chemical. Moreover, compared to the number of exit holes (24.42) found on the untreated bambara seeds, significantly ($p<0.001$) lower exit holes were found on seeds treated with the leaf powder at all rates, with and without the addition of the Actellic dust.

Table 2 shows that except at one DAT where the bambara seeds were admixed with 1.5 g leaf powder only, all concentrations of the leaf powder, with and without addition the synthetic chemical, expressed significantly ($p<0.001$) higher mortality percentages than the control. At this

Table 1: Effect f *Jatropha curcas* Leaf Extract With and Without Synthetic Chemical Application on Oviposition, Emergence of, and Damage Caused By *Callosobruchus subinnotatus*

Treatment		Oviposition	F_1 Emergence$^{\triangle}$	Damage
Leaf powder(g)	Pirimiphos-methyl(g)			
0.0	0.00	$37.42^{a\Omega}$	$25.33^{a\Omega}$ (30.20)	$24.42^{a\Omega}$
0.0	0.01	9.17^{b}	1.92^{b} (7.92)	1.92^{b}
0.5	0.00	8.67^{b}	3.00^{b} (9.98)	2.17^{b}
0.5	0.01	6.92^{b}	2.17^{b} (8.33)	1.17^{b}
1.0	0.00	6.17^{b}	1.92^{b} (7.92)	1.83^{b}
1.0	0.01	13.75^{b}	5.67^{b} (13.69)	4.58^{b}
1.5	0.00	7.17^{b}	3.33^{b} (10.47)	2.08^{b}
1.5	0.01	7.42^{b}	3.08^{b} (9.98)	2.75^{b}
	L.S.*	***	***	***
	SE±	3.241	1.628	2.397

$^{\Omega}$Means within a column followed by different letters are statistically significantly different at *** = P ≤ 0.00, Duncan's multiple range test.
$^{\triangle}$Figures in parentheses are Arcsine √percentage transformations.
*L.S = level of significance.

Table 2: Effect of *Jatropha curcas* Leaf Extract With and Without Synthetic Chemical Application on Per Cent Adult Mortality of *Callosobruchus subinnotatus* Infesting Stored Bambara Nut

Treatment		Days after treatment (DAT)					
Leaf powder(g)	**Pirimiphos-methyl(g)**	**1**	**2**	**3**	**4**	**5**	**6**
0.0	0.00	$10.83^{d\Omega}$	$27.50^{f\Omega}$	$42.50^{e\Omega}$	$65.83^{e\Omega}$	$73.33^{c\Omega}$	$78.33^{d\Omega}$
0.0	0.01	48.33^{a}	65.83^{ab}	81.67^{a}	89.17^{b}	98.33^{a}	100.00^{a}
0.5	0.00	37.50^{bc}	55.00^{d}	56.66^{d}	86.67^{bc}	90.83^{b}	93.33^{bc}
0.5	0.01	48.33^{a}	63.33^{bc}	67.50^{c}	78.33^{cd}	90.83^{b}	95.83^{a-c}
1.0	0.00	43.33^{ab}	52.50^{d}	64.17^{c}	75.83^{d}	89.17^{b}	91.67^{c}
1.0	0.01	46.67^{a}	60.00^{c}	69.17^{bc}	82.50^{b-d}	90.00^{b}	97.50^{ab}
1.5	0.00	12.50^{d}	45.00^{e}	48.33^{e}	83.33^{b-d}	93.33^{ab}	99.17^{a}
1.5	0.01	35.00^{c}	68.33^{a}	75.00^{ab}	97.50^{a}	100.00^{a}	100.00^{a}
	L.S.*	***	***	***	***	***	***
	SE±	2.480	1.415	2.406	2.810	2.287	1.500

$^{\Omega}$Means within a column followed by different letters are statistically significantly different at *** = P ≤ 0.001, Duncan's multiple range test.
*L.S = level of significance.

time (one DAT), mortality figure (12.50%) using 1.5 g leaf powder was similar to that observed in the control(10.83%). In addition, substantial mortality trend (43.33%), similar to that observed (48.33%) in the check treatment (0.01 g synthetic chemical) was observed in 1.0 g leaf powder treatment. At two and three DAT, increase in per cent mortality was observed with increase in the leaf powder concentration when pirimiphos-methyl was added. The higher the concentration of leaf powder, with actellic dust, the more efficacious the treatment. This was especially more apparent when compared to the same concentrations of leaf powder but without the pirimiphos-methyl. Moreover, at four, five and six DAT, even though appreciable insect kill was observed in all concentrations of the treatments, with and without the actellic dust, better than that observed in the control, however, the trend in insect kill (83.33, 93.33 and 99.17%, respectively) provided by 1.5 g leaf powder without the synthetic chemical was statistically similar to that obtained in the check treatment (89.17, 98.33 and 100.00%, respectively). Insect mortality (86.67%) observed using 0.5 g leafpowder without actellic dust was also similar to that observed in the check treatment (89.17%) at four (4) DAT.

In terms of seed weight loss, Table 3 shows that there was neither significant difference among treatments, with and without synthetic chemical application nor between the check (pirimiphos-methyl) and the control. However, significant ($p<0.001$) reduction in the proportion of damaged seeds was observed between all treatments with the control. Although all treatments, including the check, were statistically similar, the least damaged seed (6.70%) portion was observed in seeds treated with 1.0 g leaf powder without the synthetic chemical. This was even lower than that observed in the check treatment (8.96%).

Table 3: Effect of *Jatropha curcas* Leaf Extract With and Without Synthetic Chemical Application on Per Cent Seed Damage and Weight Loss Caused By *Callosobruchus subinnotatus*

Treatment		Per cent (%)	
Leaf powder(g)	**Pirimiphos-methyl(g)**	**Seed damage**	**Seed weight loss**
0.0	0.00	$32.37^{a\Omega}$	7.83
0.0	0.01	8.96^{b}	4.47
0.5	0.00	12.43^{b}	4.21
0.5	0.01	9.02^{b}	4.21
1.0	0.00	6.70^{b}	3.88
1.0	0.01	17.12^{b}	4.08
1.5	0.00	10.27^{b}	4.69
1.5	0.01	10.13^{b}	3.98
	L.S.*	***	N.S.
	SE±	3.730	1.324

$^{\Omega}$Means within a column followed by different letters are statistically significantly different at *** = P ≤ 0.001 and N.S. = not significant, Duncan's multiple range test.
*L.S = level of significance.

Similar potency (residual toxicity) effect was observed in all concentrations of the treatment, with and without the addition of the synthetic chemical, as well as the check. In most of the treatments, few or no bruchids were found. However, even the highest number of live bruchids (3.00) observed in 1.0 g leaf powder with synthetic chemical was significantly ($p<0.001$) lower than that observed in the control (21.50).

Table 4: Potency Effect of *Jatropha curcas* Leaf Extract With and Without Synthetic Chemical Application on *Callosobruchus subinnotatus* At 60 DAT

Treatment		Potency[△]
Leaf powder(g)	Pirimiphos-methyl(g)	
0.0	0.00	21.50[aΩ] (27.63)
0.0	0.01	0.08[b] (0.00)
0.5	0.00	1.33[b] (6.55)
0.5	0.01	1.17[b] (6.02)
1.0	0.00	0.17[b] (0.57)
1.0	0.01	3.00[b] (9.98)
1.5	0.00	1.25[b] (6.29)
1.5	0.01	0.33[b] (0.99)
L.S.[*]		***
SE±		2.024

[Ω]Means within a column followed by different letters are statistically significantly different at *** = P ≤ 0.001, Duncan's multiple range test.

[△]Figures in parentheses are Arcsine √percentage transformations.

[*]L.S = level of significance.

Discussion

Brattesten (1983) stated that research into natural plant products (botanicals) could have an advantage over synthetics as cost-effective and environmentally sustainable alternative for protecting stored food against insect attack. However, studies suggested that information and scientific support on botanicals is generally inadequate and it is often difficult to recommend particular plant materials as a replacement for chemical insecticides, because efficacy levels of botanicals could vary among storage pests, application methods and stored products. Many species and herbs and their extracts were known to possess insecticidal activities, which may be frequently present in their extracts (Schmidt *et al.,* 1991).

The use of plant extracts as insecticides to protect grains, especially legumes, against storage insects is traditional practice in many countries in Asia and Africa. The method was reported as convenient and inexpensive for the protection of stored seeds in households and on small farms and many different edible plant products have been studied as stored grain protectants (Ahmed *et al*., 1988; Don Pedro, 1989; Pacheco *et al.,* 1995).

Kumar and Sharma (2008) stated that oil and other extracts from *J. curcas* can be used as bio-pesticides, due to their insecticidal, molluscicidal, fungicidal, and nematicidal properties. Similarly, Gübitz *et al*. (1999) stated that extracts from the plant when used as natural crop pesticides in controlling insect pests, could be a promising alternative to hazardous chemicals. In addition, Heller (1996) reported that *J. curcas* extracts had the potential of controlling several insect pests and unlike spraying with synthetic chemicals, treatments with *J. curcas* extracts seemed not to affect populations of beneficial arthropods.

Conclusion

The plant extract used in this study proved effective and provided substantial reduction of ovipositon, progeny emergence and consequently lower number of exit holes (seed damage). Substantial protection was achieved by using by the plant extract singly, which was similar to that provided by using the residual insecticide powder (Actellic dust, 2%). This agreed with findings of Saxena *et al.* (1988); Schmidt *et al*. (1991) and Asawalam and Adesiyan (2001) which stated that plant parts; oil, extract, and powder mixed with grains reduced insect oviposition, egg hatchability, postembryonic development. Also, Obeng-Ofori and Reichmuth, (1997) reported that there is scientific evidence, which proved that plant derivatives inhibit progeny production by causing insect egg mortality.

Recommendations

From the findings of this study, it could be recommended that:

i. the leaf and seed extracts of *J. curcas,* singly and combined, could be admixed with bambara nut seeds during storage;
ii. 1.5/20 g leaf powder and 1.0/20 g seed powder, singly each provided the best result for the control of *C. subinnotatus* on bambara nut during storage;
iii. alternatively, 1.0:1.0 leaf/seed combinations could also be applied to protect stored bambara nut seeds against the invasion by *C. subinnotatus*.

Contribution of Authors

Dattijo, S. A. conceived the concept of this research, designed and acquired data, as well as analysis of the data. Ahmed, B. I., Adebitan, S. A. and Gurama, U. A. encouraged the investigation, supervised the findings of this work, verified the analytical methods and assist in the interpretation of the data, while Yusuf, S. R. reviewed, critically, the intellectual content of the final manuscript of this article and give final approval for the version submitted for publication.

Conflict of Interest

In accordance with Taylor and Francis policy and my ethical obligation as researcher, I am reporting that, in the conduct of this research, I enjoyed the use of facilities, equipment and personnel resources of the Department of Crop Protection, Faculty of Agriculture, Bayero University Kano, Kano State, Nigeria, a University that may be affected by the research report in this paper. I have in place an approved document for managing any potential conflict that may arise from this research.

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