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Efficacy of Balsam Spurge [*Euphorbia balsamifera* (Ait.)] as Storage Protectant Against Cowpea Bruchid (*Callosobruchus maculatus)*

Oladejo A.O.[1,*], Sikiru G. K.[1], Adedire, O.[2], Olorundare O. O.[1]; Ayorinde J.O. [1] and Ishaya M.[1]

[1]Department of Pest Management Technology, Federal College of Forestry, Jos Nigeria.
[2]Department of Statistics, Federal College of Forestry, Jos. Nigeria
*Corresponding Author: Email: folabiola60s@gmail.com

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ABSTRACT

The present study investigated the effects of Euphorbia balsamifera Leaf and stem powders and the combination on the adult's bruchids establishment, mortality, oviposition, and subsequent emergence. Varying quantities of the powders at 0.0g (control), 10.0g, 15.0g and 20.0g respectively were added to the 100g cowpea in each jar. Thereafter, five pairs (male and female) of newly emerged adult bruchids were introduced into each of the treated cowpea in the bottle jar. Each treatment was replicated three times and was arranged in Completely Randomized Design (CRD). The results revealed that the bruchids were able to establish but after 24hours, adult mortality commenced and increased significantly as the dosage and duration of the experiment increases, except in the control (0.0g) treatment. These had direct effects on the oviposition and subsequent progeny emergence. The mortality effects of these powders on insects may depend on chemical composition of the treated powders which may suggest a role in its pesticidal, anti-feedant and repellent potencies against C. maculatus. The highest mean mortality (88.33%) was recorded on the combination (20.0g). It can be concluded from this study that combinations of both powders at 20.0g could be used as alternative pesticides against bruchids infestations in stored cowpea grains.

Keywords: Cowpea grains, *Euphorbia balsamifera*, *Callasobrochus maculatus*, Balsam-spurge, Phytochemical.

1.0. Introduction

African cowpea farmers suffer heavy yield loss often over 90% due to pests and diseases most specifically by bruchids (*Callosobruchus maculatus*) (Caswell, 2001). Despite its short life cycle, *C. maculatus* is a very destructive insect pest that causes perforations and weight losses, leading to losses in nutritional as well as commercial values of cowpea seeds (Suleiman, 2016; Ojebode *et al*., 2016). This pest is reported to be the most damaging pest of legume seed and its larva infest grains such as cowpea, chickpea and Bambara groundnut, which are the most common and important legume crop in the world (Mahdi and Rahman, 2008). Pests and diseases constitute the most limiting factor affecting intensive cowpea production and may cause total loss of the grain (Radha, 2014). A major limitation of the production of cowpea in Nigeria is the emergence and menace of storage pests which do not only spoil the product but the development of larvae inside the grain when feeding also make it unfit for human consumption. *C. maculatus* is a worldwide pest and its larvae develop within various cultivated legumes, such as black-eyed beans (*Vigna unguiculata)* (Nabael *et al*., 2012). Considerable physical and nutritional losses sustained in Nigeria due to infestation of stores food products by *C. maculatus* is very high both qualitatively and quantitatively (Atwal, 2011).

Currently, insect control in stored products relies primarily upon the use of gaseous synthetic fumigants and residual insecticides, both of which may pose serious hazards to humans and environment (Ogunsin, 2011; Bourguet and Guillemaud, 2016). Residue of methyl bromide, one of the two synthetic fumigants still used in the disinfestations of storage foods have been found to exhibit carcinogenic effects in human beings (Dansi and Chou, 2005).

The persistent de-emphasizing of the use of modern or orthodox chemical pesticides in the developed world for the prevention and control of various field and storage pests is gradually gaining grounds due to their considerable side effects coupled with the cost ineffectiveness or even the inability of the farmer to have access to them (Popp *et al*., 2013; Bhandari, 2014; Mabe, *et al*., 2017). Therefore, to achieve a sustainable production of marketable cowpea for consumption, for domestic and for export trades, it is very important to search for alternative insect pest control methods that are safe for the environment and efficient in the management of *C. maculatus* for sustainable agriculture. The use of traditional medicinal plants has been identified as one of the most economical and environmentally safe method of preventing, control and preservation during the course of production and storage of cowpea in the field or store as the case may be (Oluwasma, 2011). *E. balsamifera* is one of such plants, it was reported to have some repellent activities against mosquito which are still extensively used in the traditional communities in the tropics (Adedapo *et al.*, 2004; Moore *et al*., 2006). The sap (latex) of *E. balsamifera*, which is rather poisonous if ingested, but widely used in odontology as traditionally antalgic treatment of acute dental pulpitis (Yam *et al*., 1997). Berhanu and Emana (2018) have included *E. balsamifera* in the Integrated Pest Management for the control of cowpea bruchids and was found to be effective. The plant has been found effective in the control of termite when combined with solignum and gamalin on Triplochiton scleroxylon (Nasiru and Zayyanu, 2021). It is on this note that this work centered on investigating the effects of *E.balsamifera* as an alternative pesticide for sustainable management of cowpea bruchids.

2.0. Methodology

2.1 Description of the Study Area

This research work was carried out in the Federal College of Forestry, Jos experimental site in the Jos North Local Government Area of Plateau state. The College falls between latitude 9° 51' N and longitude 8° 53' E with an altitude of 1200 m above sea level, and it is located within the northern guinea savanna agro-ecological zone of Nigeria (Figure1). Mean annual rainfall is about 1413 mm characterized by an estimation of 200 to 300 mm mean monthly rainfall between May and September, while the pick period (July) is characterized by mean monthly of above 321mm and outside of these months, rainfall decline rapidly (Kowal and Knabe, 2019). Daily temperature ranges between 10°C to 32°C, minimum and maximum respectively (University of Jos Meteorological section, 2013).

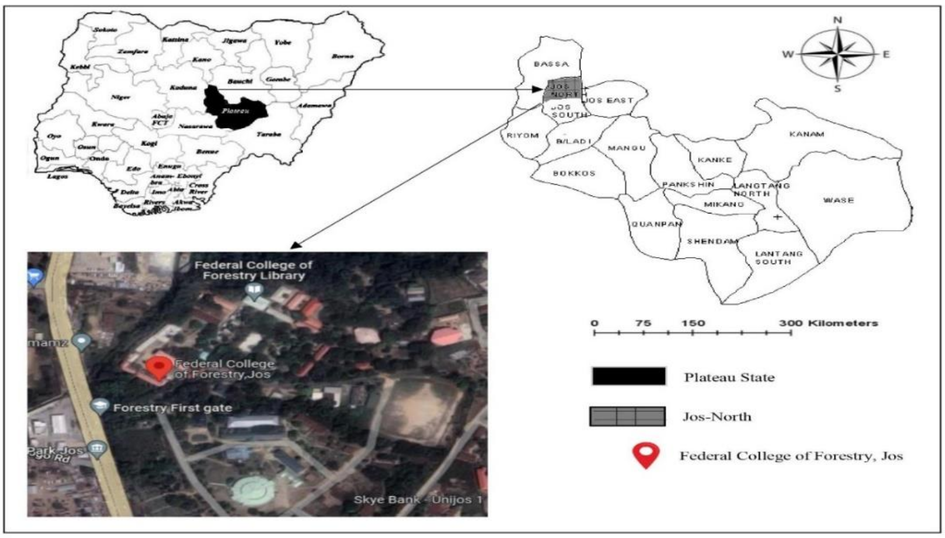

Figure 1: Location Map of the study Area

2.2. Methods of data collection

2.2.1 Preparation of plant materials

Three kilograms of dried cowpea grains were purchased from Terminus market. The grains were sieved to remove non-variable seed, dirt and broken particles. Then, 2.5 kg grains were randomly sampled and stored in a refrigerator for two weeks; to kill any prior sources of cowpea bruchids inoculums and eggs which might be already preexisting in the grains (Parugrug and Roxas, 2008).

2.2.2 Preparation of powder

Euphorbia balsamifera leaves and stem commonly known as Balsam spurge were collected from the bushes around British America Junction, Jos in Jos North LGA of Plateau State. The plant was identified at herbarium of Federal College of Forestry, Jos. The leaves and stem were dried under shade, for 14 days, in a well-ventilated area in the Laboratory of the Department of Entomology to avoid loss of active compounds through photo degradation of active ingredient by ultra-violet ray. The dried leaves and stem were milled and sieved (with a 10 mm siever) into fine powders. Electric-weighing scale was used to weigh the different doses of the powders and was separately kept in air-tight containers until required.

2.2.3 Determination of Phytochemical constituents

The leaf and stem powders of Balsam spurge (*E. balsamifera*) were subjected to standard phytochemical analyses for different constituents such as alkaloids, anthraquinones, tannins, glycosides, steroids, terpenoids, saponins and oil as described by Jigna and Sumitta (2006); Thomas and Krishnakumari (2015).

2.2.4 Culturing of Calasobruchus maculatus

Some adult *C. maculatus* were obtained from infested cowpea in Katako market, Jos. The insects were identified using morphometric method and were paired sexually then introduced into some fresh cowpea in plastic jar in order to mate. This was carried out under laboratory conditions inside a growth chamber at 30 ± 2^0C and 70±5% RH. The jars tops were sealed with 1mm muslin cloth to prevent escape. Copulation and oviposition commenced after 24 – 48 hours and this was left for 7 days. Then parent stocks were removed and cowpea seeds containing eggs were picked and kept in another jar under temperature 30 ± 2^0C and 70±5% for F1 generation to emerge for the experiment (Akinwumi *et. al*., 2007).

2.2.5 Application of Treatments and Experimental Design

One hundred grammes (100g) of clean disinfested cowpea were weighed into10 sterilized 1litre bottle jars each. In the treatments, *E. balsamifera* leaf powder, stem powder and their combination were introduced at varying quantities [0.0g (control), 10.0g, 15.0g, 20.0g] respectively to the 100g cowpea in each jar. The powders were thoroughly mixed with the disinfested cowpea using glass rod to ensure thorough admixture. The treated cowpea was left undisturbed for an hour. Thereafter, five pairs (male and female) of newly emerged adult bruchids were introduced into each of the treated cowpea in the bottle jar and were covered with 2 mm muslin net and tied with rubber ring to prevent the escape of the bruchids. Each treatment was replicated three times. The bottle jar was arranged in Completely Randomized Design (CRD).

2.2.6 Data Collection

Activity such as bruchids establishment was checked through their movement after one hour of introducing the plants powders into the cowpea grains. Adult bruchids mortality was observed by physically counting and recorded at regular intervals of 24, 48, 72, 96 and 120hours exposure to the treatments. Any bruchids that cannot move up to half of its body length is assumed to be dead after probing them with forceps in accordance with Ileke and Oni, (2011); Oladejo *et al.,* (2020).

After 120 hours, oviposition was checked by removing the entire dead bruchids and each grain were carefully observed for a transparent dot or black dot inside the grain using hand lens and Light microscope (sign of laid egg on cowpea grain), the egg numbers were counted and recorded for each treatment. The treatments were kept back in the jar for 35days, to check and record the new emergence of cowpea bruchids. Observation commenced on the 30 days after oviposition and lasted for 5 days in accordance with Oladejo *et al.,* (2022).

2.3. Method of data analyses

2.3.1 Data Analysis
Data was collected and analyzed using analysis of variance to determine if there are any differences among the treatments and Fisher's Lest Significant Difference (FLSD) test was used to determine the factor that was responsible for the difference, using SAS software.

3.0. Results and Discussion

3.1 PhytoChemical Constituents of E. balsamifera Leaf and Stem Powders
Table 1 showed the phytochemical screening of Balsam spurge (*Euphorbia balsamifera L.)* stem and leaf powders**.** It revealed the presence of a wide range of phytochemical constituents including steroids, flavonoids, alkaloids, saponins, anthraquinone, volatile oil and tannin (only in stem powder). This could be the reason for their wide range of biological activities.

Table 1: PhytoChemical Constituents of *E. balsamifera* Leaf and Stem Powders

Phytochemical composition	Leaf	Stem
Anthraquinone	+	+
Alkaloids	+	+
Balsams	+	+
Cardiac glycosides	-	-
Flavonoids	+	+
Saponins	+	+
Steroids	+	+
Tannin	-	+
Terpenoids	-	-
Volatile oil	_	+

Key: *+ Present; - Absent*

3.2 *The effects of leaf and stem powder of E. balsamifera on the adult establishment, mortality and oviposition*

The introduced bruchids were seen moving towards the surface of the cowpea under the muslin net after an hours of introduction in all the treatments showing the evidence of establishment. This is in line with Suleiman *et al.,* (2012) who used *Lawsonia inermis* L for the control of *Sitophilus zeamais* and were seeing moving near the surface of the treatment after the application of the botanical.

It was observed after 24 up to 120hours of bruchids exposure to the powders that the adult mortality increases as the dosage and duration of the experiment increases, except for control (0.0g) treatment that had the least mortality. At 96 hours and dosage 15.0g, bruchids mortality in stem powder and combined powders had 50.00% and increases until we had 63.33% and 66.67% at 120 hours in stem powder and combined powders respectively. At 20.0g dosage, only combined powder had 50.00% at 72 hours while at leaf and stem powders had 50.00% and 56.67% at 96 hours respectively. They all increased to 60.00%, 66.67% and 83.33% respectively at 120 hours. These differences were significantly different statistically (Table 2). This is in conformity with Edeldouk *et al.,* (2015) who reported that application of cotyledon powder of melon (*Cirullus vulgaris*) on *C. maculatus.* The mortality effects of different plant materials on insects may depend on several factors such as chemical composition and species susceptibility (Aktar *et al.,* 2004). In the present study, mortality of *C. maculatus* varied with dosage of the powders which means that grounding the plants parts allows them to release their insecticidal effect on bruchids, while untreated grain offers free environment where bruchids suffer no developmental limitations hence, the highest

feeding rates (Mbaiuinam *et al*., 2006). Also, the preponderance of tannins, steroids, anthraquinones, and cardiac glycoside in stem powder may suggest a role in its pesticidal, anti-feedant and repellent potencies against some insect and pathogens (*Idris et al*., 2014; Thomas and Krishnakumari, 2015). Karamanoli *et al*., (2011) complemented this report by explaining that tannins exert its action by a combination of mechanisms that include iron chelation and enzyme inhibition which suggest the higher efficacy of stem powder over leaf that does not have tannin (Table 1). It was also observed that Saponins and Cardiac-glycosides are present in both leaf and stem powders; this could be linked to the insecticidal interaction with cholesterol which results in impairing ecdysteroid synthesis (Chaieb, 2010; Lengai *et al*., 2020). Terpenoids are also known to have a pungent odour and act as a deterrent to the insect (Martini *et al*., 2003).
Oviposition of bruchids decreases as the dosage increases in all the powders applied, with the control recording the highest oviposition values (47.17%), while other values ranges from 30.50% down to 21.17% at 10.0g to 20.0g with combined powders having the least. All these treatments were significant different from the control (Table 3). The findings of this study are in accordance with Edeldouk *et al.,* (2012); Khaliq *et al.,* (2014) and Kosar *et al.,* (2016) who concluded that plant powders reduce oviposition of bruchids.

Cowpea bruchids emergence decreases as the dosages increases in all the plant powder, with the control recording the highest values (52.17%), while the least values (7.34%) was observed in the combined treatment (Table 4). This is line with Ojo and Ogunleye (2013); Rivera *et al.,* (2014) and Oni and Ogungbite (2015) who previously reported the use of plant powders in suppressing adult *C. maculatus* emergence.

Table 5 shows the values obtained from bruchids weight loss in the experimental setup. Bruchids weight loss decreases as the dosages increases in all the plant powders, with the control recording the significantly highest values of 17.22, 17.89 and 18.33grams in leaf, stem powders and combination respectively. While the least values of 5.48 grams was observed in the combination treatment.

Table 2: Percentage Means adult mortality after exposure to different plant powders

Powders	Dosage	24 hours	48 hours	72 hours	96 hours	120 hours
Leaf	Control	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a
	10 g	3.33±5.77a	10.00±0.00b	23.33±5.77b	33.33±5.77b	46.67±5.77b
	15 g	6.67±5.77a	20.00±0.00c	33.33±5.77c	43.33±5.77bc	56.67±5.77bc
	20 g	10.00±10.00a	23.33±5.77c	36.67±5.77c	50.00±10.00c	60.00±10.00c
Stem	Control	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a
	10 g	0.00±0.00a	10.00±0.00b	23.33±5.77b	36.67±5.77b	50.00±0.00b
	15 g	6.67±5.77b	23.33±5.77c	36.67±11.55c	50.00±10.00c	63.33±5.77c
	20 g	6.67±5.77b	23.33±5.77c	40.00±0.00c	56.67±5.77c	66.67±5.77c
Leaf and Stem	Control	0.00±0.00a	0.00±0.00a	0.00±0.00a	0.00±0.00a	13.33±11.55a
	10 g	10.00±10.00ab	16.67±5.77b	33.33±5.77b	46.67±11.55b	63.33±15.28b
	15 g	6.67±5.77ab	23.33±11.55bc	36.67±15.28bc	53.33±11.55bc	66.67±15.28b
	20 g	16.67±5.77b	30.00±0.00c	50.00±0.00c	66.67±5.77c	83.33±5.77b

Values with the same alphabet are not significantly different at P≤ 0.05

Table 3: Percentage means Oviposition of Bruchids after Exposure to Different Dosages of Plant Powders

Concentrations	Leaf Powder	Stem Powder	Combination of Leaf and Stem
Control	47.17±1.15a	47.17±1.15a	47.17±1.15a
10g	34.40±3.85b	30.13±6.15b	27.17±2.36b
15g	28.33±5.58bc	30.50±2.00b	24.83±2.48b
20g	25.17±1.26c	22.83±0.76c	21.17±0.76c

Values with the same alphabet are not significantly different at P≤ 0.05

Table 4: Percentage means Bruchids Emergence after Exposure to Different Dosages of Plant Powders

Concentrations	Leaf Powder	Stem Powder	Combination of Leaf and Stem
Control	51.07±1.68a	51.24±0.75a	52.17±2.25a
10g	11.66±1.48b	11.97±1.55b	9.54±1.64b
15g	10.75±1.12bc	9.62±0.80c	9.16±1.89b
20g	8.48±1.29c	9.62±0.73c	7.34±0.57b

Values with the same alphabet are not significantly different at P≤ 0.05

Table 5: Percentage means Weight loss after Exposure to Different Dosages of Plant Powders

Concentrations	Leaf Powder	Stem Powder	Combination of Leaf and Stem
Control	17.22±1.39a	17.89±1.07a	18.33±0.57a
10g	7.55±0.40b	6.98±0.13b	6.46±0.98b
15g	7.30±0.21b	6.77±0.29b	5.98±0.54b
20g	7.13±0.12b	6.34±0.90b	5.48±0.99b

Values with the same alphabet are not significantly different at P≤ 0.05

4.0. Conclusions

Based on the results of this study, both stem and leaf powders of Euphorbia balsamifera can be use as storage protectant against *C. maculatus*, but its combination was more effective (83.33% mortality) just as conventional chemicals. However, efficacy depends on the dosage and the exposure duration of the powders. The pesticidal effects of these powders were capable of anti-feedant and repellent potency. The powders also have a tendency of blocking the spiracles of the insect thus impairing their respiration leading to their death. Therefore, it can be concluded that small holder farmers can use of *E. balsamifera* leaf and stem powders and the combination of both powders at 20.0g as an alternative control option in integrated storage pest management strategies of *C. maculatus*. However, a standardized method of quantity formulation for this botanical should be established.

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