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INSECTICIDAL ACTIVITY OF SESAME LEAF AND STEM EXTRACTS ON *Clavigralla tomentosicollis* Stal (Hemiptera: Coreidae)

***Negbenebor, H.E.,[1] Abdullahi, R.I.,[2] Nura, S.,[3] and Sharif, U.[4]**
1. Department of Biological Science, Baze University, Abuja, Nigeria.
2. Department of Botany, Ahmadu Bello University, Zaria, Nigeria.
3. Department of Biology, Ahmadu Bello University, Zaria, Nigeria.
4. Department of Biological Science, Yusuf Maitama Sule University, Kano, Nigeria.
*Corresponding Author: Helen.ehimemen@bazeuniversity.edu.ng

ABSTRACT
A study was conducted to investigate the insecticidal potency of leaf and stem ethanolic extracts of white sesame (Sesamum indicum L.) on the pod-sucking bug (Clavigralla tomentosicollis Stal.) under laboratory conditions. Four different concentrations of the extracts (5.00, 10.00, 15.00 and 20.00 mg/l) of ethanolic extracts were prepared from the leaf and stem parts of the Sesame plant. Water and Cypermethrin were used as negative and positive controls. Fresh cowpea pods were treated with the various concentrations of the stem and leaf ethanolic extracts and the pod-sucking bugs were introduced into the jars containing the pods. The result obtained revealed significant differences (P≤0.05) in the effect of various concentrations in inducing mortality of the insects at different instars. The extracts were more effective in the first (1^{st}) instars against the pod-sucking bugs. The activity is concentration dependent as it increases with increase in concentration of the extract. However, leaf ethanolic extract proved to be more effective. More so, the percentages of pods infested by the bugs were found to be lower among the 20.00 mg/l treated pods in both stem and leaf ethanolic extracts. The phytochemical result indicated the presence of certain phytochemicals such as alkaloids, carbohydrates, cardiac glycosides, diterpenoids, flavonoids, proteins, saponins, steroids, tannins and triterpenoids that were proved to be vital in the insecticidal activity of the extracts. Thus, the stem and leaf ethanolic extracts of sesame are effective botanical insecticides against C. tomentosicollis especially at 20.00 mg/l of the leaf extract.
Keywords: Clavigralla tomentosicollis, Concentrations, Ethanolic Extract, Sesame

INTRODUCTION

Cowpea (*Vigna unguiculata* L.) is a pulse crop produced and consumed largely by subsistence farmers in the semi-arid and sub-humid regions of Africa (DeBoer, 2003). It is an important cash and food crop for many poor farmers and also noted for its high nutritional value. The seeds and foliage are rich in protein, carbohydrate, minerals (iron and calcium), vitamins and carotene (Adedire *et al*., 2011), and are used in preparing several dishes for man and livestock (Bressani, 1985). It is an important food to millions of people in Africa in general (Ndiaye, 2007; Adedire *et al*., 2011) and Nigeria in particular. Nigeria is the largest producer and consumer of cowpea in the world (Prereia *et al*., 2001; Lowenberg-Deboer and Ibro, 2008). But, despite the significance of cowpea to food security and that animal protein tends to be far from the reach of common man in Nigeria, the crop is still facing the problem of serious damage by insects' pests in the field, thereby reducing yield of the plant. One such important field insect pest of cowpea in Nigeria is the pod-sucking bug (*Clavigralla tomentosicollis*) that attacks the pods and seeds of cowpea (The Mcknight Foundation, 2006). *C. tomentosicollis* sucks the sap from cowpea pods, causing them to shrivel and dry prematurely, with resultant losses in yield estimated between 20 -100% in various parts of Africa (Singh and Allen, 1980).

The use of chemical insecticides, in practice appears to be the major control measure to pod-sucking bugs (Jackai and Adalla, 1997). Chemical insecticides however, have detrimental effects to humans and the environment, and also are not affordable to a majority of peasant farmers (Alebeck, 1996). Thus, botanical insecticides remain an option in insect pest management of this pod-sucking bug because they are believed to provide the most effective control against insect pests that have become resistant to other insecticides (Weinzierl, 2000).

They provide sustainable, safe, available and cheap alternative to synthetic insecticides in the control of storage insect pests threatening stored food and these have led to the belief that plant-derived insecticides are safer and more eco-friendly than synthetic products. This study therefore aimed at investigating the insecticidal efficacy of sesame leaf and stem ethanolic extracts against *Clavigralla tomentosicollis*.

MATERIALS AND METHODS

Collection and Authentication of Plant Materials

Fresh leaves and stem of Sesame (*Sesamum indicum* L.) were obtained from the Botanical Garden of the Department of Botany, Ahmadu Bello University Zaria. The samples were authenticated at the herbarium of the Department of Botany, Ahmadu Bello University, Zaria. A voucher number 02859 was assigned to the plant material.

Extracts Preparation

The fresh leaf and stem of sesame were air dried at room temperature, pounded to fine powder by pestle and mortar according to the protocols described by Dabire *et al*. (2008). The ethanolic extracts were prepared by soaking 100 g of each powder in 150 ml of 95% ethanol and shaken in orbital shaker at 120 rpm. The preparations were left to stand for another 24 hours and then filtered through Whatman No 1 filter paper. The filtrates were concentrated to dryness at 40°C under reduced pressure on a rotary evaporator and were stored in a refrigerator at −4°C until needed.

Preparation of Ethanolic Concentrations

Four different concentrations of the extracts (5.00, 10.00, 15.00 and 20.00 mg/l) of ethanolic extracts were prepared from the leaf and stem parts of the Sesame plant. Water and Cypermethrin were used as negative and positive controls.

Phytochemical Analysis

The phytochemical screening of the ethanolic leaf and stem extracts of *Sesamum indicum* was carried out inorder to determine the presence of Bioactive components according to the method described by Sofowara (1993) and Adegoke *et al*. (2010). The ethanolic extracts of *S. indicum* were subjected to qualitative test for the presence of bioactive components that include Molisch's test for detection of Carbohydrates, Lead acetate test for detection of Tannins, Keller-Killiani's test for detection of Cardiac Glycosides, Frothing/Foaming test for detection of Saponins, Libermann-Burchard's test for detection of Steroids and Triterpenoids, Copper Acetate's test for detection of Diterpenoids, Alkaline test for the detection of Flavonoids, Xanthoprotein test for the detection of Protein, Borntrager's test for the detection of Anthraquinones and Meyer's and Wagner's test for detection of Alkaloids,.

Preparation of Test Animals

The larvae and adults of *C. tomentosicollis* were derived from a laboratory mass rearing facility. Insects were supplied with fresh cowpea pods and reared in wood cages according to the technique described by Dabire *et al*. (2005). Toxicity and direct contact tests were carried out on 1st, 3rd and 5th instars larvae and adults of *C. tomentosicollis*. A chemical insecticide cypermethrin (25g active ingredient/litre) procured in Sabon Gari market, Kano, Nigeria was used as positive control.

Determination of the Effects of the Ethanolic Extracts on *C. tomentosicollis*

The tests were carried out in 374 cm^3 capacity plastic flasks. For each insect stage (1st, 3rd and 5th instars larvae and adults) five treatments equivalent to the six different ethanolic concentrations (including controls) were setup. For each insect stage, 120 individuals were split in 20 flasks (6/flask) and were replicated three times. Four flasks correspond to the four ethanolic concentrations while the fifth and sixth ones represented the controls (cypermethrin and untreated control). Cowpea pods (5 pods/flask) were treated with the ethanolic extracts for 5 seconds then allowed to dry for 5 minutes and put in the flask containing the insects. The untreated control pods were treated by dipping in tap water as negative control and some in the insecticide as positive control. Insects were kept in flasks for 24 hours and the number of dead insects and damaged pods was recorded for each treatment.

Data Analysis

The data obtained was subjected to statistical analysis using Analysis of variance (ANOVA) via SPSS (11.0 versions) with Duncan's Multiple Range Test used in separating the means.

RESULTS

The result for the phytochemical screening of the ethanolic leaf and stem extracts of *S. indicum* are presented in Table 1. The results revealed the presence of active constituents in the form of alkaloids, carbohydrates, cardiac glycosides, diterpenoids, flavonoids, proteins, saponins, steroids, tannins, Triterpenoids and anthraquinoones.

Table 1: Phytochemical composition of the leaf and stem ethanolic extracts of Sesame

Phytochemical Compound	Test	Leaf	Stem
Alkaloids	Wagner's test	+	+
Anthraquinones	Bornfrager's	+	-
Carbohydrate	Molisch	+	+
Cardiac glycosides	Keller-Killani	+	+
Diterpenoids	Copper acetate	+	+
Flavonoids	Alkaline reagent test	+	+
Protein	Xanthoproteic	+	+
Saponins	Frothing test	+	+
Steroids	LibermannBuchard's test	+	+
Tannins	Lead subacetate test	+	+
Triterpenoids	LibermannBuchard's tes	+	+

Key: (+) = Present (-) = Absent

The result for the effect of leaf ethanolic extracts of sesame on *C. tomentosicollis* is presented in Table 2. The result showed significant difference (P≤0.05) in the effect of the various concentrations of the extracts in inducing mortality of the insects' pest. It also revealed that, the extracts caused the mortality of 96.8-100% of the pests at first instar. However, the mortality rate decreased by 12.6-29.6% of third instar with increase in concentration of the extracts. Furthermore, the effects of the extracts reduced significantly on adults via the 5th instar to between 5.1-11.1%.

Table 2: Effects of various Sesame leaf ethanolic extracts on *C. tomentosicollis* mortality

Treatments (mg/l)	1st Instar (%)	3rd Instar (%)	5th Instar (%)	Adult (%)
5.00	96.8^{a*1}	12.6^{e}	4.8^{d}	5.1^{c}
10.00	99.7^{a}	17.1^{d}	7.3^{c}	6.8^{c}
15.00	99.7^{a}	23.4^{c}	8.6^{c}	6.6^{c}
20.00	100^{a}	29.6^{b}	13.7^{b}	11.1^{b}
Cypermethrin	100 ^{a}	87.3^{a}	92.4^{a}	96.5^{a}
Water	0.0^{b}	0.0^{f}	0.0^{e}	0.0^{d}

N.B: *^{1} Means within the columns with the same superscript letter(s) are not significantly different (P≤0.05)

Similarly, the result for the effect of stem ethanolic extracts of sesame on *C. tomentosicollis* (Table 3) revealed significant difference (P≤0.05). The result showed that 65.2% of the first instars were killed by 5.00 mg/l of the sesame stem ethanolic extract. The percentage increased to 87.4% at 20.00 mg/l concentration. But, the mortality rate decreased with progression in the instars to adult stage. At adult stage, 3.4-8.7% of the insects were killed by the extracts. The insecticidal effect of the extract is also concentration dependent, increases with increase in concentration at all the insects stages of development.

Table 3: Effect of various Sesame stem ethanolic extracts on *C. tomentosicollis* mortality

Treatments (mg/l)	1st Instar	3rd Instar	5th Instar	Adult
5.00	65.2^{c*1}	5.3^{e}	2.2^{e}	3.4^{c}
10.00	67.6^{c}	9.7^{d}	3.9^{d}	4.2^{c}
15.00	82.1^{b}	14.8^{c}	5.1^{c}	5.0^{c}
20.00	87.4^{b}	27.2^{b}	9.8^{b}	8.7^{b}
Cypermethrin	100^{a}	85.7^{a}	96.1^{a}	91.9^{a}
Water	0.0^{d}	0.0^{f}	0.0^{f}	0.0^{d}

N.B: *^{1} Means within the columns with the same superscript letter(s) are not significantly different (P≤0.05).

However, the result for the number of cowpea pods infested under the protection of the sesame leaf ethanolic extracts is presented in Table 4. The result indicated no significant difference in the number of infested pods at first instar. But significant difference (P≤0.05) exists in the number of infested pods treated with the leaf ethanolic extracts from 3rd instar to adult stage. The result revealed significant decreased in the number of damaged pods with increase in the concentrations of the extracts.

The number of infested pods decreased from 1-4 infested pods at the third instar with increase in the concentrations of the leaf ethanolic extracts of sesame. The number of infested pods decreased from 29 to 10 pods at adult stage with increase in concentrations.

Table 4: No. of sesame leaf ethanolic extracts treated cowpea pods infested by *C. tomentosicollis*

Treatments (mg/l)	1st Instar	3rd Instar	5th Instar	Adult
5.00	$2.8^{b*!}$	3.9^{b}	19.9^{b}	28.7^{b}
10.00	2.0^{b}	3.4^{b}	16.2^{c}	23.5^{c}
15.00	2.1^{b}	2.6^{c}	9.3^{d}	14.2^{d}
20.00	2.0^{b}	1.8^{d}	4.5^{e}	9.7^{e}
Cypermethrin	0.0^{b}	0.0^{e}	1.38^{f}	4.59^{f}
Water	9.8^{a}	16.5^{a}	29.2^{a}	52.7^{a}

N.B: *[1] Means within the columns with the same superscript letter(s) are not significantly different (P≤0.05)

More so, the result for the number of treated pods infested by *C. tomentosicollis* is shown in Table 5. The result indicated significant differences (P≤0.05) in the degree of infestation of the treated pods at different developmental stages of the test insect except at 1 st instar where no significant difference is found. The number of infested pods at first instar was between 0-2 pods. But the number decreased from 3 to 1 pod at 3rd instar and from 14 to 8 pods at the 5th instar with increase in concentration of the extract. However, the number of infested pods decreased from 30 to 18 pods at adult stage. The effect is also concentration dependent, decreased with increase in concentrations of the extract.

Table 5: No. of sesame stem ethanolic extract treated cowpea pods infested by *C. tomentosicollis*

Treatments (mg/l)	1st Instar	3rd Instar	5th Instar	Adult
5.00	2.1^{b}	3.5^{b}	14.2^{b}	30.1^{b}
10.00	1.8^{b}	3.1^{b}	11.0^{c}	28.0^{c}
15.00	1.7^{b}	2.8^{b}	11.2^{c}	26.3^{d}
20.00	0.0^{b}	1.3^{c}	7.9^{d}	18.2^{e}
Cypermethrin	0.0^{b}	0.0^{d}	5.02^{e}	9.69^{f}
Water	11.3^{a}	14.7^{a}	21.6^{a}	49.3^{a}

N.B: *[1] Means within the columns with the same superscript letter(s) are not significantly different (P≤0.05)

DISCUSSION

The insecticidal activities of botanical extracts have proved vital in the control of insect pests. The organic solvent extractions of active ingredient might enhance the contact insecticidal activity of Sesame leaf and stem ethanolic extracts on all the development stages of *C. tomentosicollis* as reported by Dabire *et al.* (2005). This is also in line with the previous finding by Oparaeke *et al.* (2005) who maintained that, this kind of formulation was successful in Nigeria with plant extracts for controlling cowpea insect pest. The differences in toxicity may be associated to the proportion of the active chemicals in the extracts due to differential solubility in the ethanol solvent between leaf and stem. This probably was the reason for the high mortality of *C. tomentosicollis* in leaf extracts than in the stem extract.

The present study revealed that all the tested plants extracts were toxic and could be used as protectant against *C. tomentosicollis*. However, the high toxicity of leaf ethanolic extracts of sesame signifies the relative importance of the extracts in controlling the pod-sucking bug infestation in cowpea field. The high dose (20 mg/l) protects high percentage of the pods and induces 100% mortality of *C. tomentosicollis* at the first instar. Therefore the application of the extracts at the early fruiting stage of cowpea can control the devastating effect of the pod-sucking bug. This finding agrees with the work of Opareke and Dike (2005), Ba *et al.* (2009), Mukanga *et al.* (2010), Adedire *et al.* (2011), Ileke and Oni (2011) and Mwine *et al.* (2013) who observed that certain botanicals are effectively toxic against insect pests including *C. tomentosicollis*. This result indicates that all the extracts displayed considerable effectiveness to reduce *C. tomentosicollis* infestation on cowpea.

The efficacy of the leaf and stem extracts of Sesame presented in this study is in line with the previous findings of Mundi *et al*. (2012), Asawalam and Anaeto (2014) Suleiman and Suleiman (2014) and Danga *et al*. (2015) who individually reported high efficacy of botanical extracts from members of Lamiaceae and Myrtaceae leaf extracts as important pesticides against insects pests. The ability of the evaluated plant extracts to significantly induce adult mortality indicated that the plant might possess ovicidal and larvicidal properties as reported by Jose and Adesina (2014). This finding agrees with the work of Adeniyi *et al*. (2010) who reported that plant extracts from *Vernonia amygdalina* at 4.0% concentration resulted in higher toxicity (measured as percentage mortality) to *Acanthoscelides obtectus*. This finding therefore adds to the existing data on the efficacy of plant extracts as biopesticides of cowpea as highlighted by Adeniyi *et al*. (2010) that extracts from leaves of *Oscimum gratissimum*, *Sida acuta*, *Telfaria occidentalis* and *V. amygdalina* possess good insecticidal potential because of their phytochemical constituents and the order of toxicity at different concentrations was 4.00 > 3.00 > 2.00 > 1.00%. This is similar to the finding of the present study that insecticidal effects of botanicals increase with increase in concentration. The toxicity of the ethanolic extracts of the test plants used in this study is in agreement with that reported by Alabi *et al*. (2005), Ijeh and Ejike (2011) who individually reported that, aqueous extract of the leaves of *Vernonia sp*. has high phytotoxic properties.

Phytochemical analysis is very useful in the evaluation of some active biological components of some plants as reported by Anwar *et al*. (2007) which could be used to predict the activity of such plant parts on some test organisms. Phytochemical screening helps to reveal the chemical nature of the constituents of the plant extract which may also be used to search for bioactive agents that could be used in the synthesis of very useful drugs as stressed by Sibanda and Okoh (2008). The profile of the bioactive compounds of a plant therefore indicates its insecticidal value as such is of great importance as stressed by Alphonso and Saraf (2012). The presence of active constituents in the leaves and stem of *S. indicum* revealed in this study is in conformity with the findings of Fasola and Ogunsola (2014) who reported similar compounds in the leaves and stem of *S. indicum* and *Ceratotheca sesamoides*. Similar finding was also found in the work of Neeta *et al*. (2015).

CONCLUSION

It was concluded that, ethanolic extracts obtained from stem and leaf of *Sesamum indicum* are very effective botanical insecticides that are toxic to *C. tomentosicollis* and can therefore be used as protectants against the bug's infestation in cowpea fields. From the study, it is generally understood that, the efficacy increased with increase in concentrations of the extract. It was therefore recommended that, 20.00 mg/l of the leaf extract of sesame be used at early stage of the podding.

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