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Insecticidal Activity of *Cyperus rotundus* L. and *Datura stramonium* L. Co-Administered with Sesame Oil Against African Bollworm *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae)

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Abstract

African bollworm is a worldwide-spread species that shows a high degree of polyphagia and it is considered as one of the main agricultural pests in the world. Laboratory experiments were conducted in the Research Laboratory, College of Agricultural Studies, Sudan University of Science and Technology to evaluate the lethal effect of *C. rotundus* and *D. stramonium* on the *H. armigera* and to investigate the synergistic effect of sesame oil and tested extracts by dipping methods. Five concentrations (4%, 6%, 8%, 10% and 12%) were used for each plant extract in a Completely Randomized Design. Tubers ethanolic extract of *C. rotundus* at 12% concentration caused 90% mortality after 72 hrs of application, whereas 12% concentration of seeds ethanolic extract of *D. stramonium* generate only 70% mortality after 72 hrs of exposure.

When sesame oil was added to each concentration of ethanolic extract of *C. rotundus* and *D. stramonium* it exhibited a synergistic effect. In fact, the 10% concentration scored 53.3% and 76.7% mortality, however, when mixed with sesame oil the mortality increased significantly to 83.3% and 100% for *D. stramonium* and *C. rotundus* respectively after 72 hrs of exposure.

This study clearly demonstrates that both tested plants have a lethal effect on the larvae of the African bollworm. However, tubers ethanolic extract of *C. rotundus* seems to be significantly more toxic than the seeds ethanolic extract of *D. stramonium*. This study also revealed that sesame oil has a synergistic effect when added to these plant extracts.

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Introduction

African bollworm *Helicoverpa armigera* is a worldwide-spread species that shows a high degree of polyphagia and it is considered as one of the main agricultural pests in the world. Its presence has been reported in diverse continents, such as Africa, Europe, Asia and Oceania [1]. in addition to feeding on high value crops it is an extremely dangerous pest because its reproduction rate is extremely high and it can migrate over a long distance [2]. It has been reported on 67 host families, including Asteraceae, Fabaceae, Malvaceae, Poaceae and Solanaceae and this pest has caused losses to economically important crops such as cotton, legumes, sorghum, maize, tomato, soybean, ornamental plants, and fruit trees [3]. It develops resistance to broad spectrum of insecticides due to exposure of successive generations while moving from one crop to another which made this pest highly resistant to many pesticides such as cyclodiene, pyrethroids, organophosphates, carbamates etc [4].

Because of the hazards of synthetic insecticides, recently the pesticidal effects of botanical extracts have been investigated by several researchers worldwide [5 ,7].

Nut-grass *C. rotundus* is growing commonly and considered as noxious weed but at the same time it contains tremendous important medicinal as well as pharmacological properties. The most effective parts of this perennial herb are rhizomes and tubers. *C. rotundus* contains many secondary metabolites such as sesquiterpenes, quinones, flavonoids, saponins, alkaloids, phenolic acids, coumarins and steroids (steroidal glycoside, sitosteryl-(6'-hentriacontanoyl)-β-dgalactopyranoside [8].

Jimsonweed *Datura stramonium* is a cosmopolitan weed of cultivated fields. It is characterized by its narcotic, hallucinogenic, and medicinal properties, as well as its effects in human poisonings [9]. The phytochemical screening revealed that it contains phenols, flavonoids, tannins, saponins, alkaloids, steroids and glycosides. All parts of the plant are toxic, but the ripe seeds contained the highest amount of alkaloids [7] .

Insecticide synergists have been used not only to monitor the insecticide resistance mechanisms but also as an admixture in these insecticides for the control of many insects. They contribute significantly to the improvement of insecticides efficacy, particularly when problems of resistance need to be addressed. Piperonyl butoxide which is isolated from sesame oil has been used as a synergist with many organophosphates and pyrethroid insecticides to control various pests [10].

In the last four decades many botanical formulations have proven to be potent and effective as many as conventional synthetic pesticides even at low concentrations. In fact, botanical insecticides have drawn great attention as major control agents in organic farming. Higher plants are extremely abundant with biologically active secondary metabolites. Over 80% of all known alkaloids, phenols and other secondary metabolites were produced by higher plants[11]. Stoll [12] demonstrated that the use of plant extracts to control destructive insects is not new, rotenone, nicotine and pyrethrin have been used for a considerable time in small scale subsistence and also commercial agriculture. The objectives of this study were to evaluate the lethal effect of *C. rotundus* and *D. stramonium* on the H*. amigera* and to investigate the synergistic effect of sesame oil and tested extracts.

Materials and Methods

The experiments were conducted in the Research Laboratory, College of Agricultural Studies (Shambat), Sudan University of Science and Technology (SUST), during February-March, 2021, where the temperature was between 25-32°C.

Larval instars of *H. armigera* were collected from unsprayed tomato plants grown in Gamouaia Agricultural irrigated scheme Southern Khartoum and brought to the laboratory for rearing. Early instar were reared in groups of 100 larvae in plastic cages 19 cm in diameter covered with muslin cloth and fed on okra fruits, whereas 4^{th}_ instars were reared separately in plastic cubs 5 cm in diameter and 7 cm in height to avoid cannibalism the bottom of each cubs was filled with sand for pupations. Upon emergence the adults were transferred to plastic cages 31x20x19 cm covered with muslin cloth and fed on 10% sugar solution[13], cotton stripes were hung on the margins of the cages for eggs laying and were replaced daily with new stripes while newly hatching

larvae were transferred to the larval rearing cages. The rearing process continued until a sufficient number of homogenous population of larvae was collected for the experiments.

Tubers of *C. rotundus* were collected from Arashkool scheme White Nile State and seeds of *D. stramonium* were collected from river bank, Omdurman area and brought to the laboratory where they were shade-dried. After complete dryness the plant samples were crushed separately by an electronic blender, 120g of prepared seeds powder were extracted with absolute ethanol using soxhlet apparatus, extraction continued for six hours, and the ethanol solvent was removed off the crude extract by rotary evaporator[10].

Five concentrations (4%, 6%, 8%, 10% and 12%) were prepared by dilution, water plus 00.01% soap were used to make emulsion.

Bioassay Procedure

Second larval instar were used in this study. Fruits dipping method [14] was followed, fresh okra fruits were cut in small pieces and were dipped for 30 seconds in different concentrations and left to dry under laboratory condition for 10 minutes. One hour pre starved larvae were used for each treatment (10 larvae/ treatment) and each treatment was replicated three times. Three replicates were treated with *C. rotundus* and *D. stramonium* plus sesame oil at ratio of 1:1.

Three replicates were also used as a control in which water plus 00.01% soap was administered, in addition to thirty larvae treated with sesame oil. This experiment was set in a completely randomized design. All treated larvae were kept in petri-dishes 9 cm in diameter at temperature of 25±1°C. During treatment period the feeds were replaced as required. The mortality counts were recorded 24, 48, 72 and 96 hrs after application.

Statistical Analysis

The obtained data were statistically analyzed according to analysis of variance (ANOVA); Duncan's Multiple Range Test was used for means separation using Genstat version 12.1 also the data were subjected to Probit analysis using SPSS 16.0 software.

Results and Discussion

Obtained data in (table 1) shows that all concentrations of the ethanolic extract of *C. rotundus* and *D. stramonium* scored a significantly higher mortality percentage than the control after 24hrs of exposure. The mortality percent increased with the increase of both concentration and exposure period. Tubers ethanolic extract of *C. rotundus* at 4% and 12% concentrations caused 46.7% and 90% mortality respectively after 72 hrs of application, whereas 12% concentration of seeds ethanolic extract *D. stramonium* induced only 70% mortality after 72 hrs of exposure.

The results exhibited in (table 1 & table 2) showed that each concentration of the tubers ethanolic extract of *C. rotundus* and seeds ethanolic extract of *D. stramonium* mixed with sesame oil gave significantly higher mortality percentage than its counterpart alone after 24hrs of exposure. Meanwhile after 48 hrs of application the lowest concentration of *C. rotundus* mixed with sesame oil caused 63.3% mortality which were not significantly different from that caused by the highest concentration (12%) of its counterpart of *D. stramonium* that cause 73.3% mortality. It should be noted that there is no significant differences among the highest concentrations (8%, 10% and 12%) of both plant extract after 96 hrs of exposure as indicated in (table 2).

The results exhibited in (table 3) clearly demonstrated that tubers ethanolic extract of *C. rotundus* are significantly more toxic than the seeds ethanolic extract of *D. stramonium* (LC_{50} were 4.2% for *C. rotundus* and 7% for *D. stramonium*)

The obtained results revealed that all concentrations of the tubers ethanolic extract of *C. rotundus* generated significantly higher mortality percent than control throughout the experimental period. This clearly demonstrates that the tubers ethanolic extract of *C. rotundus* has a lethal effect against the *H. armigera*. Similar results were obtained by Imam, *et al*. [5] who found that the rhizomes of *C. rotundus* exhibited larvicidal activity against *Aedes aegypti* larvae and the mortality were in a dose- dependant manner. Sharma and Gupta [15] noted that methanolic extract of *C. rotundus* tubers strongly

Table 1. Lethal effect of *C. rotundus* and *D. stramonium* on the mortality of second larval instars of the African bollworm (Shambat-Khartoum-Sudan2021).

Plant extract	Conc. (%)	Means mortality (%)			
		Exposure time (hrs.)			
		24	48	72	96
Cyperus rotundus	4	36.7(6.1)cde	43.3(6.6)d	46.7 (6.9)def	50.0(7.1)d
	6	43.3(6.6)cd	53.3(7.3)c	60.0(7.8)c	60.0(7.8)c
	8	56.7(7.6)b	70.0(8.4)b	73.3(8.6)b	73.3(8.6)b
	10	60.0(7.8)b	73.3(8.6)b	76.7(8.8)b	80.0(9.0)b
	12	76.7(8.8)a	86.7(9.3)a	90.0(9.5)a	90.0(9.5)a
Datura stramonium	4	23.3(4.9)f	30.0(5.5)e	40.0(6.4)f	40.0(6.4)e
	6	26.7(5.2)ef	33.3(5.8)e	43.3(6.6)ef	43.3(6.6)e
	8	30.0(5.5)ef	46.7(6.9)cd	50.0(7.1)de	53.3(7.3)cd
	10	33.3(5.8)def	50.0(7.1)cd	53.3(7.3)cd	56.7(7.6)cd
	12	46.7(6.9)bc	63.3(8.0)b	70.0(8.4)b	76.7(8.8)b
Control	-	0.0(0.7)g	0.0(0.7)f	0.0(0.7)g	0.0(0.7)f
SE±	-	0.5	0.4	0.3	0.3
C.V. (%)	-	8.8	5.6	4.7	3.5

Means within column followed by the same letter (s) are not significantly different at ($p < .001$).

Means between brackets are transformed according to $\sqrt{(X+0.5)}$

Table 2. Effect of ethanolic extract of *C. rotundus* and *D. stramonium* mixed with sesame oil on the mortality of second larval instars of the African bollwom (Shambat-Khartoum-Sudan2021).

Plant extract	Conc.(%)	Means mortality (%)			
		Exposure time (hrs.)			
		24	48	72	96
Cyperus rotundus + Sesame oil	4	56.7 (7.6)bcd	63.3 (8.0)cd	66.7 (8.2)de	66.7 (8.2)c
	6	66.7 (8.2)b	76.7 (8.8)b	83.3 (9.2)abc	83.3(9.2)ab
	8	83.3 (9.2)a	90.0 (9.5)a	93.3 (9.7)ab	96.7 (9.9)a
	10	96.7 (9.9)a	100.0(10.0)a	100.0(10.0)a	100.0(10.0)a
	12	100.0(10.0)a	100.0(10.0)a	100.0(10.0)a	100.0(10.0)a
Datura stramonium Sesame oil	4	33.3(5.8)e	43.3 (6.6)e	60.0 (7.8)e	63.3(8.0)c
	6	43.3 (6.6)de	60.0 (7.8)d	70.0 (8.4)cde	73.3 (8.6)bc
	8	50.0 (7.1)cd	66.7 (8.2)bcd	76.7 (8.8)bcd	90.0 (9.5)a
	10	56.7 (7.6)bcd	70.0 (8.4)bcd	83.3 (9.2)abc	93.3 (9.7)a
	12	60.0 (7.8)bc	73.3(8.6)bc	93.3 (9.7)ab	93.3(9.7)a
Sesame oil	-	16.7 (4.1)f	20.0 (4.5)f	23.3 (4.8)f	23.3(4.8)d
Control	-	0.0 (0.7) g	0.0 (0.7) g	0.0 (0.7) g	0.0 (0.7) e
SE±	-	0.6	0.4	0.5	0.5
C.V. (%)	-	7.9	5.3	6.4	6.1

Means within column followed by the same letter (s) are not significantly different at (P< 0.05).

Means between brackets are transformed according to √(X+0.5)

Table 3. LC values of ethanolic extract of *C. rotundus* and *D. stramonium* mixed with sesame oil on the mortality of second larval instars of the African bollworm after 96 hrs (Shambat-Khartoum-Sudan2021

Plant extract	LC* values (%) and 95% Confidence limits (Lower – Upper)			
	LC_{50}	LC_{90}	Slope± SE	Chi- square χ2
C. rotundus	4.2 (0.5 – 5.8)	12.4 (10.3 – 18.1)	2.0 ± 0.6	0.6
C. rotundus + Sesame	3.4 (2.2 – 4.1)	6.2 (5.4 – 7.8)	4.9±1.1	1.40
D.*stramonium*	7.0 (3.8 – 9.0)	18.4 (14.0 – 38.4)	1.8± 0.6	1.8
D.*stramonium* + Sesame	3.1(1.3 – 4.3)	9.1 (7.3 – 14.7)	2.8±0.7	0.9

LC = Lethal Concentration SE = Standard Error

inhibit the activity of acetylcholinesterases (ache). Another finding reveal that the acetone leaves extracts of *C. rotundus* at 50% exhibited significant mortality percentage of 46.6% and 51.6% repellency against Rice grains weevils *Sitophilus oryzae* [16].

The present data also showed that the seeds ethanolic extract of *D. stramonium* scored a significantly higher mortality percentage than the control after 24 hrs of exposure and the mortality were dose and time dependent. Similar results were obtained by Karimzadeh and Rabiei [17]. They indicated that flower, seed, and root extracts of *D. stramonium* were highly toxic against diamondback moth *Plutella xylostella* larvae. Also, Abbasipour *et al*. [18] found that the *D. stramonium* extract had strong contact toxicity against *Callosobruchus maculatus* adults and the mortality increases with increase in the concentration and exposure period.

When sesame oil was added to each concentration of ethanolic extract of *C. rotundus* and *D. stramonium* it exhibited a synergistic effect. In fact, the lowest concentration (4%) of seeds ethanolic extract of *D. stramonium* gave only 40 % mortality after 96 hrs of exposure; however, when mixed with sesame oil it increased significantly to 63.3%. This may indicate that the detoxification mechanism in this insect involves mixed function oxidases which are known to be inhibited by sesame oil[19]. Similar result were obtained by Elnour [10] who indicated that the sesame oil have synergistic effect when mixed with *Cassia occidentalis* and *Conocarpus lancifolius* when tested against African melon lady bird beetle *Henosepilachna elaterii* (Rossi). Another finding demonstrate that the sesame oil was a synergist with cypermethrin that played more or less the same role as Piperonyl butoxide (PB) in monooxygenase inhibition against diamondback moth *Plutella xylostella* [20].

Conclusion and Recommendations

This study clearly demonstrates that both tested plants have a lethal effect on the larvae of the African bollworm. However, tubers ethanolic extract of *C. rotundus* are significantly more toxic than the seeds ethanolic extract of *D. stramonium*. In addition the study revealed that sesame oil has a synergistic effect when added to the these plant extracts.

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Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this article.

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