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Insecticidal and Preservative Potential of Selected Plant Extracts Against *Callosobruchus maculatus* and GC-MS Characterization of the Bioactive Constituents

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Abstract

Gas chromatography-mass spectrometry (GC-MS) characterization of phytocomponents, insecticidal and preservative activity of *Calotropis procera (Cp), Striga hermonthica (Sh) and, Hyptis suaveolens (Hs) against Callosobruchus maculatus*, a major post-harvest pest of stored cowpea, were evaluated using standard bioassay methods. The crude extracts yields ranged from 11.78 to 16.67%. The bioassay result revealed varying insecticidal and preservative efficacy among the exctracts. Hs and Sh exhibited 3.35 to 58.38% Abbott-corrected mortality, whereas Cp extract exhibited little or no insecticidal activity, with Abbott correct mortality ranging from -25.00 to 14.78%. Hs provided complete grain protection, maintaining zero grain damage, zero weevil perforated index, zero weight loss, and 100% seed viability for up to 120 days at 7.5 mg/ml. In comparison, preservative efficacy of Cp and Sh decreas after 60 days did not provide effective long-term protection comparable to Hs. The GC-MS analysis revealed that all three extracts were dominated by fatty acid methyl esters (FAMEs) with Hs containing the highest abundance of octadecenoic acid methyl ester (53.49%) which may have attributed to its higher bioactivity. These findings suggest that Hs crude extract is a promising eco-friendly alternatives to synthetic insecticides for the medium term preservation of stored cowpea.

Keywords: Bioassay activity, credue extracts, GC-MS characterization, *Callosobruchus maculatus*

Introduction

Postharvest insect pests remain one of the key constraints to food safety and sustainable grain storage, predominantly in tropical and subtropical regions where environmental conditions favors rapid pest reproduction [1]. Stored legumes, specifically cowpea, remain highly unprotected to

attack by *C. maculatus*, a destructive storage insect that causes seed perforation, weight loss, reduced germination, and deterioration in grain quality. These losses have serious implications for household food safety, market worth, and seed availability for subsequent planting. Recent studies have shown that storage pests can cause significant quantitative and qualitative losses in grains, making their control a critical component of postharvest management [2]. Synthetic insecticides have extensively been applied to control storage pests, but their constant use is with several limitations, including toxic residues on food grains, ecological contamination, hazards to human health, and the emergence of insect resistance. Evidence from toxicological and epidemiological studies indicates that chronic exposure to synthetic pesticides contributes to cancer, neurological disorders, endocrine disruption, reproductive abnormalities, immunotoxicity, and respiratory illness [3]. These alarms situations have increased interest in safer, inexpensive, and environmentally friendly alternatives for pest control in stored foods. Plant based products are among the most promising alternatives as they are biodegradable, locally accessible, and often rich in biologically active compounds that can affect insects through multiple mechanisms [4].

Phytochemical examination is highly necessary in this context hence it supports the identification the classes of compounds present in plant extracts and provides clues to the basis of their biological activity. Several plants comprise alkaloids, flavonoids, tannins, saponins, terpenoids, and fatty acid derivatives, which have been associated with insecticidal, repellent, antifeedant, and preservative [5]. Comparative evaluation of selected plant extracts is also necessary because the efficacy of botanical materials varies with plant species, extraction method, concentration, and target pest. Such comparison helps determine which extracts are most suitable for improvement into botanical insecticides.

This study is on the phytochemical investigation and comparative assessment of the insecticidal and preservative effectiveness of selected plant extracts against *C. maculatus*. The study is anticipated to provide evidence on the potential of these botanicals as sustainable substitutes to synthetic pesticides in stored-grain defense.

Material and Methods

Samples collection

Plant samples (*Calotropis procera, Striga hermonthica, and Hyptis suaveolens*) and cowpea grains were collected in clean sack bags to prevent contamination and ensure integrity. About 4 kg

of individual plant samples of *Calotropis procera, Striga hermonthica, Calotropis citratus and Hyptis suaveolens* were collected from the Nigerian Defence Academy. Approximately 30 kg of uninfected and 2 kg of infected cowpea were sourced from farms in Mando, Igabi Local Government Area, in Kaduna State, Nigeria. All plant leaves were subjected to authentication at the Herbarium, Department of Biological Sciences, Nigerian Defence Academy, Kaduna, Nigeria.

Samples preparation and preservation

The collected plant leaves were carefully washed with water and air-dried separately for two weeks. One kilogram of each dried plant sample was then pulverized into a fine powder using a mechanical grinder and sieved through a 0.25-inch mesh sieve. The cowpea samples were sorted out by removing undesirable materials, transferred into zip-lock bags, frozen 72 hours to eliminate any existing eggs or insects [6]. Subsequently frozen cowpea grains were exposed to air to get rid of residual moisture. After drying the uninfected cowpea samples were transferred into airtight storage sack bags to avoid exposure to dampness and pests before used. The individual plant powders were stored in labeled air tight bottles and kept at room temperature to prevent degradation of the bioactive compounds..

Rearing of insect for bioassay activity

About 600 g of infected cowpea collected from Mondo market in Igabi local government of Kaduba State Nigeria was weighed and placed in a 1000 ml plastic dish. Following a modified method by [7] , 50 pairs of adult Callosobruchus maculatus weevils were collected after 7 days and introduced into three different 500 ml plastic jar containing 300 g of uninfected cowpea for mass rearing. The experimental jars were covered with muslin cloth and tired with rubber band to allow airflow. The insects were removed (both dead and live) after 7 days oviposition by sieving and the cowpea grains were returned to the incubation jar under the same condition for emergence of the first generation (F1). The procedure was repeated using F1 for F2 generation used for this investigation.

Extraction of phytocompounds from plant samples

A total of 300 g of each individual pulverized plant sample were weighed and transferred into a 2-L maceration bottle, soaked using 1.5 L of 70% ethanol and 30% water, thoroughly mixed, covered , and shaken daily for one hour at 250 rpm using an orbital shaker for seven (7) days,

dependable with standardized maceration practices proposed for plant extraction [8]. On the seventh day, the extraction was carried out twice, and the mixture was soaked again for three (3) more days using the same amount of ethanol and water, making it a total of a ten (10) day extraction period for this study [9]. The filtrates obtained from each extraction were combined separately for individual plant extract, filtered with Whatman No. I filter paper, and concentrated at 40 °C via rotary evaporator. Finally, each sample extract was subjected to further drying to achieve a constant weight by leaving the concentrated crude extract at room temperature for three weeks to remove residual solvent and obtain semi-solid extract used for bioassay application

Insecticidal bioassay of crude extracts of the targeted plants against *Callosobruchus maculatus*

The insecticidal potential of crude extracts from *Calotropis procera*, *Sriga hermonthica*, and *Hyptis. suaveolens* was evaluated for its toxicity against the cowpea weevil, *C. maculatus*. Clean cowpea (100 g) was weighed into triplicate 250 mL plastic jars and treated with 2 mL extract concentrations at 5, 7.5, 10, 12.5, and 15 mg/mL [10], which established a total of forty-five (45) experimental setups of the targeted plants. Grains were manually stirred for uniform coating, air-dried for 1 h at room temperature to evaporate solvents, then infested with 10 adults of *C. maculatus*. The jars were covered with muslin cloth held with rubber bands to allow proper ventilation while preventing from insect escaping. Ngative (-ve) DMSO and positive (+ve control) dichlorvos were used for comparison. Insect mortality was assessed at 24, 48, and 72 h post-treatment, while oviposition was evaluated 7 days after infestation. Further observations were conducted monthly form 30 to 120 days to determine grain damage (WD%), weevil perforation index (WPI%), weight loss (W%), and germination potential (G%), as described by [11].

GC-MS characterization

The phytochemical constituents of the selected plant crude extracts were analyzed using an Agilent 7890B gas chromatograph coupled with an Agilent 5977A Mass Selective Detector (MSD). Chromatographic separation was done on an HP-5MS Ultra Inert capillary column (30 m × 0.25 mm i. d. × 0.25 μm film thickness) with helium (99.999% purity) as the carrier gas at a constant flow rate of 1.13 mL min-1. Samples (1 μL) were injected in the splitless mode at an injector temperature of 260 °C. The oven temperature program started at 110 °C and was maintained for 2

min, then increased to 200 °C at 10 °C min^{-1}, and subsequently raised to 280 °C at 5 °C min^{-1}, with a final hold of 9 min. The transfer line temperature was kept at 260 °C. The mass spectrometer operated under electron ionization (EI) at 70 eV, with the ion source and quadrupole temperatures set at 230 °C and 150 °C, respectively. Mass spectra were recorded in full-scan mode over a mass range of m/z 50–650 following a solvent delay of 5 min.

The GC–MS system was autotuned to obtain optimal mass accuracy, sensitivity and overall instrument performance using the manufacturer's automatic tuning procedure (Agilent autotune) before sample analysis. Crude plant extracts were prepared by dissolving 0.5 g of each extract in ethanol and filtering before GC–MS analysis. A 1 μL aliquot of each prepared sample was injected into the GC–MS system. Phytochemical constituents were identified by comparison of the mass spectra obtained with the entries in the National Institute of Standards and Technology (NIST) Mass Spectral Library [13]. Identity was verified using previously developed analytical methods by GC-MS based on match of the mass spectrum, retention time, relative peak area and match quality to the library [12, 13].

Data analysis

Entirely experiments were carried out in triplicates, and results are presented as mean ± standard deviation (SD). The significance of the differences between the means of the samples were established by the analysis of variance ANOVA($p<0.05$)

Results and Discussion

The results of the evaluation of the insecticidal and preservatives efficacy of targeted crude extracts against *C. maculatus* are shown in Tables 1 to 7

Table 1: Insecticidal activitiy of the targeted plants extracts against *C. maculatus.*

	Mean mortality of crude extracts			
Plant extracts	Conc. mg/ml and% v/v	No of 24 h Mortality (Mean ±SD)	No of 48 h (Mean±SD)	No of 72 h (Mean±SD)
Sh	7.5 mg/ml	1.67 ± 1.53	1.00 ± 1.00	2.33 ± 1.53
Hs	7.5	3.33 ± 1.15	4.33 ± 0.58	0.33 ± 0.58
Cp	7.5	1.67 ± 2.08	1.33 ± 1.15	1.00 ± 1.00
Sh	10	3.33 ± 0.58	0.67 ± 0.58	0.67 ± 1.15
Hs	10	3.67 ± 1.53	1.67 ± 0.58	3.00 ± 1.00
Cp	10	2.67 ± 1.53	1.00 ± 1.00	1.33 ± 1.15
Sh	12.5	4.00 ± 1.00	0.67 ± 0.58	1.67 ± 1.53
Hs	12.5	**6.**00 ± 1.73	2.00 ± 0.00	1.67 ± 1.15
Cp	12.5	0.00 ± 0.00	1.33 ± 1.15	2.00 ± 0.00

Sh	15	1.33 ± 1.53	0.33 ± 0.58	2.67 ± 1.15
Hs	15	6.67 ± 1.53	1.00 ± 0.00	1.33 ± 1.15
Cp	15	0.33 ± 0.58	1.00 ± 1.00	2.33 ± 0.58
Sh	20	2.33 ± 1.53	2.33 ± 1.53	1.00 ± 1.00
Hs	20	2.67 ± 2.08	1.00 ± 1.00	1.33 ± 1.15
Cp	20	2.00 ± 1.00	1.00 ± 1.00	1.33 ± 1.53
+ve	-	10.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
-ve	-	2.00 ± 1.00	0.33 ± 0.58	1.00 ± 1.00

Table 2: Percentage mortality (M%) and Abbott's Corrected mortality of *C. maculatus* exposed to crude extracts.

Treatment	Conc.mg/ml and % v/v	24 h M%	48 h M%	72 h M%	24 h Abbott %	48 h Abbott %	72 h Abbott%
Sh	7.5 mg/ml	16.7	10.0	23.3	-4.13	6.93	14.78
Hs	7.5	33.3	43.3	3.3	16.63	41.42	-7.44
Cp	7.5	16.7	13.3	10.0	-4.13	10.36	0.00
Sh	10	33.3	6.7	6.7	16.63	3.52	-3.67
Hs	10	36.7	16.7	30.0	20.88	13.83	22.22
Cp	10	26.7	10.0	13.3	8.38	6.93	3.67
Sh	12.5	40.0	6.7	16.7	25.00	3.52	7.44
Hs	12.5	60.0	20.0	16.7	50.00	17.22	7.44
Cp	12.5	0.0	13.3	20.0	-25.00	10.36	11.11
Sh	15	13.3	3.3	26.7	-8.38	0.00	18.56
H s	15	66.7	10.0	13.3	58.38	6.93	3.67
Cp	15	3.3	10.0	23.3	-20.88	6.93	14.78
Sh	20	23.3	23.3	10.0	4.13	20.62	0.00
Hs	20	26.7	10.0	13.3	8.38	6.93	3.67
Cp	20	20.0	10.0	13.3	0.00	6.93	3.67
+ve	-	100.0	0.00	0.00	100.00	Na	Na
-ve	-	20.0	3.3	10.0	0.00	0.00	0.00

Na = not applicable (because at 48 and 72 h, no insect to kill).

Table 3: Oviposition deterrence response of *C. maculatus* on the targeted plant extracts

Treatment	Conc.	No of Ovipostion (Mean± SD)	OD%
Sh	7.5 mg/ml	0.267 ± 0.153	52.91
Hs	7.5	0.300 ± 0.100	47.07
Cp	7.5	0.500 ± 0.400	11.82
Sh	10	0.267 ± 0.153	52.91
Hs	10	0.333 ± 0.058	41.22
Cp	10	0.467 ± 0.115	17.64
Sh	12.5	0.400 ± 0.289	29.45
Hs	12.5	0.200 ± 0.100	64.71
Cp	12.5	0.667 ± 0.208	-17.64
Sh	15	0.500 ± 0.265	11.82

Hs	15	0.400 ± 0.173	29.45
Cp	15	0.767 ± 0.379	-35.29
Sh	20	0.600 ± 0.265	-5.82
Hs	20	0.367 ± 0.289	35.26
Cp	20	0.633 ± 0.416	-11.64
+ve	—	0.070 ± 0.260	-
-ve	—	0.567 ± 0.115	-

Table 4: Preservative activity of plant extracts after 30 days of storage

30 days Treatment	Conc.mg/mL and%v/v	Nu (Mean ±SD)	Nd(Mean ±SD)	Wu (Mean±SD)	Wd (Mean ±SD)	GD%	WPI%	W%
S*h*	7.5 mg/mL	97 ± 1.00	3 ± 1,00	30.77 ± 0.46	0.56 ± 0.15	3.00	6.12	1.23
Hs	7.5	95.3 ± 3.78	3.66 ± 3.78	29.07 ± 1.08	1.05± 1.23	3.66	7.37	0.22
Cp	7.5	96.6 ± 2.30	3.33 ± 2.30	29.55 ± 1.10	1.42 ± 0.69	3.33	6.75	-1.31
Sh	10	96.6 ± 2.08	3.33 ± 1.52	28.79 ± 2.11	0.8 ± 0.42	3.33	6.75	0.65
Hs	10	94.6 ± 2.51	5.33 ± 2.51	29.89 ± 0.58	2.6 ± 0.75	5.33	10.38	-2.89
Cp	10	96.6± 4.16	3.33 ± 4.16	28.91 ± 2.86	0.76 ± 0.97	3.33	6.75	0.79
Sh	12.5	98.3 ± 1.52	1.66 ± 1.52	98.33 ± 1.26	0.48 ± 0.50	1.66	3.48	1.18
Hs	12.5	97.3 ± 2.30	2.66 ± 2.30	97.33 ± 1.87	0.88 ± 0.76	2.66	5.45	1.78
Cp	12.5	94.3± 3.78	5.33 ± 3.78	28.04 ± 1.88	1.39 ± 1.05	5.33	10.38	0.65
Sh	15	97.0 ± 4.72	3.00 ± 4.72	30.69 ± 3.09	0.64 ± 0.88	3.00	6.12	0.97
Hs	15	95.3 ± 3. 05	4.66 ± 3. 05	29.50 ± 0.23	1.18 ± 0.83	4.66	9.19	0.84
Cp	15	92.6 ± 2.30	7.33 ± 2.30	27.50 ± 1.31	2.13 ± 0.42	7.33	13.74	0.15
Sh	20	96.3± 4.72	3.66 ± 4.72	30.57 ± 2.47	0.81 ± 1.05	3.66	7.37	1.10
Hs	20	96.3 ± 0.57	3.66 3 ± 0.57	27.61 ± 2.49	0.74 ± 0.21	3.66	7.37	1.07
Cp	20	96.0 ± 0	4 ± 0	29.88 ±1.68	0.93 ± 0.06	4.00	8.00	1.01
+ve	–	100 ± 0	0± 0	33.16 ± 3.04	0.00 ± 0.00	0.00	00.00	0.00
-ve	–	54± 2.82	46± 2.82	27.5 ± 0.70	1.53 ± 0.71	46.00	-	-

Table 5: The results of preservative activity of plant extracts after 60 days of storage

60 days Treatment	Conc	Nu (Mean ± SD)	Nd (Mean± SD)	Wu (Mean± SD)	Wd (Mean ± SD)	GD%	WPI%	W%
Sh	7.5mg/ml	14.33 ± 9.07	85.67 ± 9.07	4.42 ± 1.92	21.91 ± 6.91	85.67	46.14	14.64
Hs	7.5	91.67 ± 6.03	8.33 ± 6.03	27.52 ± 1.27	2.33 ± 1.78	8.33	7.689	0.57
Cp	7.5	7.33 ± 7.09	92.67 ± 7.09	3.31 ± 3.86	23.35 ± 3.83	92.67	48.09	40.96
Sh	10	1.67 ± 2.89	98.33 ± 2.89	0.58 ± 1.00	24.36 ± 2.48	98.33	49.57	28.19
Hs	10	71.00 ± 20.07	29.00 ± 20.07	22.18 ± 8.31	8.80 ± 5.87	29.0	22.48	0.83
Cp	10	32.67 ± 47.45	67.33 ± 47.45	9.80 ± 15.49	16.14 ± 12.55	67.33	40.23	13.52
Sh	12.5	40.00 ± 42.43	60.00 ± 42.43	10.86 ± 11.27	16.08 ± 11.13	60.00	37.5	0.77
Hs	12.5	80.67 ± 33.64	19.33 ± 33.64	23.31 ± 9.94	5.29 ± 9.17	19.33	44.65	1.02
Cp	12.5	4.00 ± 6.93	96.00 ± 6.93	1.25 ± 2.16	20.73 ± 1.87	96.00	48.97	29.66
Sh	15	29.33 ± 37.23	70.67 ± 37.23	8.36 ± 10.43	18.37 ± 10.01	70.67	41.17	6.22
Hs	15	41.67 ± 34.47	58.33 ± 34.47	12.26 ± 11.83	14.68 ± 9.01	58.33	36.84	8.43
Cp	15	0.00 ± 0.00	100.00 ± 0.00	0.00 ± 0.00	23.91 ± 1.20	100.0	50	100
Sh	20	16.00 ± 10.00	84.00 ± 10.00	6.15 ± 3.41	21.17 ± 1.89	84.00	45.65	28.92
Hs	20	0.67 ± 1.15	99.33 ± 1.15	0.29 ± 0.49	27.27 ± 2.17	99.33	49.83	36.33
-ve	–	0.00 ± 0.00	100.00 ± 0.00	0.00 ± 0.00	21.95 ± 1.11	100.0	-	-
+ve	–	100.00 ± 0.00	0.00 ± 0.00	32.14 ± 2.52	0.00 ± 0.00	0.00	-	-

Table 6: The result of preservative activity of plant extracts on GD% , W%, and WPI% after 90 to 210 days of storage.

90 days	Conc. mg/ml)	Nu (Mean ± SD)	Nd (Mean ± SD)	Wu (Mean ± SD)	Wd (Mean ± SD)	GD%	WPI%	W%
Hs	7.5	100.00 ± 0.00	0.00 ± 0.00	31.610 ± 0.000	0.000 ± 0.000	0.00	0.00	0.00
Hs	12.5	95.00 ± 0.00	5.00 ± 0.00	27.290 ± 0.000	1.360 ± 0.000	5.00	4.76	0.72
120 days								
Hs	7.5	100	0	31.40 ± 0.00	0.00 ± 0.00	0.00	0.00	0.00
Hs	12.5	49	51	14.50 ± 0.00	13.50 ± 0.00	51.00	50.00	3.45
150days								
Hs	12.5	40 ± 00	60 ± 00	11.24 ± 00	12.93 ± 00	60.0	37.50	13.98
180 days								
Hs	12.5	39 ± –	56 ± –	9.68 ± –	13.80 ± –	56.67	100	-5.11
210 days								
Hs	12.5	35 ± –	55 ± –	8.68 ± –	14.80 ± –	55.00	100.00	-5.19
+ve	–	100 ± 0.00	0 ± 0.00	29.82 ± 1.07	0 ± 0.00	0.00	0.00	0.00

Table 7: Gas chromatography-mass spectrometry analysis of *He, Cc* and *St*

Hs	Rt (min)	Area%	Qual	Compound	Class	Molecular Formula
5	13.327	53.49	98	Octadecenoic acid methyl ester	Fatty acid methyl ester(oleate-type)	$C_{19}H_{36}O_2$
4	11.559	17.64	93	Pentadecanoic acid methyl ester	Fatty acid methyl ester	$C_{16}H_{30}O_2$
3	11.440	9.54	95	Hexadecanoic acid methyl ester,	Fatty acid Ester	$C_{17}H_{34}O_2$
Cp						
6	13.072	37.65	96	Octadecenoic acid methyl ester	Fatty acid methyl ester	$C_{19}H_{36}O_2$
3	11.225	17.49	95	Hexadecanoic acid methyl ester	Fatty acid methyl ester	$C_{17}H_{34}O_2$
8	13.429	16.33	64	9-Hexadecenoic acid methyl ester	Fatty acid methyl ester	$C_{17}H_{32}O_2$
18	30.612	1.08	43	3,7,11-Trimethyldodeca-2,6,10-trien-1-yl palmitate	Terpenoid ester / long-chain lipid ester	$C_{30}H_{56}O_2$

Sh						
9	13.057	44.22	95	13-Octadecenoic acid	Fatty acid methyl ester	$C_{19}H_{36}O_2$
5	11.227	17.75	96	Pentadecanoic acid,, methyl ester	Saturated fatty acid methyl ester	$C_{17}H_{34}O_2$
10	13.439	13.93	83	Heptadecanoic acid methyl ester	Fatty acid methyl ester	$C_{19}H_{38}O_2$
14	18.774	3.33	49	Heptadecanolide methyl ester.	Lactone (fatty acid–derived bioactive compound	$C_{17}H_{32}O_2$
18	23.207	0.48	53	2,6,10-Dodecatrien-1-ol, 3,7,11-trimethyl-(Farnesol isomer)	Sesquiterpenoid alcohol	$C_{15}H_{26}O$

The insecticidal effects of the crude extracts of *Calotropis procera*, *Striga hermonthica and Hyptis suaveolens against Callosobruchus maculatus* are presented in Tables 1 and 2. Table 1 shows the mean mortality values at 24, 48 and 72 h, while Table 2 presents the percentage mortality and Abbott's corrected mortality values. The result revealed a considerable variation in the insecticidal toxic efficacy of the three plant extracts.

Among the extract tested, *Hyptis suaveolens* demonstrated the highest insecticidal activity, followed by *Striga hermonthica* while *Calotropis procera* showed the lowest and most inconsistent activity. The positive (+ve) control caused 100% mortality within 24 h, confirming the susceptibility of the tested insects to insecticidal treatment, while the negative (-ve) untreated control recorded little or no mortality, indicating that the observed effects were attributed to the plant extracts**.** *Hyptis* suaveolens showed the highest insecticidal activity at 24 h exposure with mean mortality of 6.00 ± 1.73 and 6.67 ± 1.53 insects at 12.5 and 15 mg/mL concentrations respectively. On the other hand, Striga hermonthica showed moderate insecticidal effect with mean mortality of 3.33 ± 0.58 to 4.00 ± 1.00 insects. *Calotropis procera* was the least toxic with mortalities ranging from 0.00 ± 0.00 to 2.67 ± 1.53 insects.

Mortality rate generally decreased at the 48 and 72 h exposure period in all treatments indicating that the extracts had a rapid toxic effect shortly after application rather than a prolonged residual action. The trend was further confirmed by the percentage and Abbott's corrected mortality values. At 24 h, *Hyptis suaveolens* recorded the highest mortality, reaching 60.0% and 66.7% at 12.5 and 15 mg/mL, respectively, with corresponding Abbott's corrected mortalities of

50.00% and 58.38%. These results indicate a relatively strong mortality effect during the early stages of exposure. *Striga hermonthica* also exhibited moderate activity, recording 40.0% mortality and 25.00% corrected mortality at 12.5 mg/mL. Conversely, *Calotropis procera* recorded consistently low mortality values and even negative values of Abbott's corrected mortality, indicating little or no insecticidal effect relative to the untreated control. The decrease in mortality observed at 48 and 72 hours can be attributed to the volatilization, degradation or lower persistence of the bioactive compounds present in the crude extracts [14]. However, *Hyptist suaveolens* showed relatively superior activity as compared to other extracts during the exposure period.

The negative Abbott's corrected mortality values were observed in some treatments, including *Calotropis procera* at 12.5 mg/mL (-25.00%), *Striga hermonthica* at 15 mg/mL (-8.38%), and *Hyptis suaveolens* at 7.5 mg/mL (-7.44%). This values indicate that mortality in those treatment was lower than that observed in the untreate control. This findings suggest negligible insecticidal action and emphasize the significance of interpreting percentage mortality values alongside with corrected mortality.

The results presented in Table 3 additional confirmed that *Striga hermonthica* and *Hyptis suaveolens* were more effective in inhibiting oviposition than *Calotropis procera* against *C. maculatus.* The highest inhibition was recorded for *Hyptis suaveolens* at 12.5 mg/mL (64.71%), whereas *Striga hermonthica* recorded a maxium inhibition of 52.91% at 7.5 and 10 mg/mL. The oviposition deterrent observed in *Striga hermonthica* and *Hyptis suaveolens* may be associated the fatty acid methyl esters (FAMEs) and other bioactive phytocomponents identified by GC-MS analysis and phytochemical screening.

The botanical active component are known to influence insect behaviour through antifeedant, repellency, and oviposition deterrence, with their mode of action varying from plant species, concentration, and targeted insect species [15]. In contrast, *Calotropis procera* demonstrated weak or negative oviposition deterrence at higher concentrations, indicating low suppression of egg laying *by C. maculatus*.The preserving efficacy of the tested crude extracts varied with concentration and storage period. After 30 days of storage (Table 4), all the three extracts provided sustantial protection against *C. maculatus* infestation when compared with the untreated control. This protective efficacy was reflected in higher numbers of undamaged grains

(Nu), lower damaged grains (Nd), reduce grain damage (GD%), low weevil perforation indices (WPI), and low weight loss (W%) in the stored samples.

Among the extracts, *Striga hermonthica* showed the highest preservative activity during the short-term storage (30 days), with highest number of undamaged grains (Nu = 98.3 ± 1.52), lowest grain damage (GD =1.66%), and a low weevil perforation index (WPI =3.48%), signifying effective protection of stored cowpea. *Hyptis suaveolens* also exhibited considerable effectiveness, mainly at 12.5 mg/ml, thereby recording low GD (2.66%) and WPI (5.45%) in the stored cowpea. Although *Calotropis procera* presented some protective properties across all the tested concentrations, its performance was generally lower than that of *Hyptis suaveolens* and *Striga hermonthica* as shown with its higher GD% and WPI%**.** The protective action observed after 30 days may be associated with the active constituents identified by GC-MS characterization which may have contributed to grain preservation through repellency, feeding deterrent, oviposition deterrence or direct toxcicity to against *C. maculatus,* thereby reducing infestation [16]. After 60 days of storage (Table 5), clearer differences in residual action were observed between the extracts. *Hyptis suaveolens* maintained the highest protection at concentrations of 7.5 mg/ml, where it recorded releatively high number of Nu (91.67 ±6.03), low GD (8.33%), low WPI (7.689%) and low (0.57%). The sustained efficacy of *Hyptis suaveolens* may be associated to the persistence of its bioactive consitutent which continued to inhibit infestation during stroge **[**17]. In comparision the preservative efficacy of *Striga hermonthica* and *Calotropis procera* b decreased after 60 days, as shown by increase in GD%, WPI and W% which may be attributed to the gradual degradation of the active phytochemical constituents over time.

Deterioration in the effectiveness of botanical insecticides during extended storage are often attributed to environmental exposure, volatilization or chemical instability of the active compounds [18]. As presented in table 6, only *Hyptis suaveolens* crude extract showed significant preservative efficacy over the long storage period. After 90 days of storage, only one replicate each of *Hyptis suaveolens* at 7.5 mg/mL and 12.5 mg/mL remained active. Consequently, no variation could be determined, and the standard deviation was recorded as zero (±0.00). The extract at the concentration of 7.5 mg/mL provided 100% Nu, zero GD, zero WPI and zero W% and at 90 days of storage which implies complete protection. This protective effect was preserved until 120 days of storage, demonstrating the higher residual activity of *Hyptis suaveolens* compared to

the other extracts tested. However, the protective effect was not sustained beyond 120 days. At 150 days of storage, the 7.5 mg/mL treatment showed complete deterioration of efficacy, indicating that the active constituents were no longer capable of suppressing infestation and grain damage. This sharp deterioration suggests that the bioactive coponents responsible for protection may have undergone degradation, volatilization, or loss of biological activity during prolonged storage. The sustained protective activity of *Hyptis suaveolens* during the 30 to 120 can be attributed to the phytochemical profile of the extract particularly the presence of octadecenoic acid methyl ester (FAME) and other active compounets identified by GC-MS analysis. In contrast, the 12.5 mg/mL treatment exhibited a different pattern of activity. Although it did not achieve the complete protection observed at 7.5 mg/mL, it maintained measurable preservative effects beyond 120 days of storage. At 90 days, the treatment remained highly effective, recording 95 undamaged grains, low 5% GD, low 4.76% WPI and low 0.72% W. Subsequently, its efficacy gradually dropped, as reflected by high GD%, high WPI and high W% at 120, 150, 180, and 210 days of storage. Nevertheless, the persistence of some protective activity throughout the storage period suggests a longer residual effect than that observed for the 7.5 mg/mL treatment after 120 days.

These findings suggest that *Hyptis suaveolens* possesses considerable potential as a botanical grain protectant for medium-term storage of cowpea. However, further studies on formulation improvement may be required to enhance the stability and persistence of its bioactive compounds during extended storage periods.

The GC-MS analysis (Table 7) revealed that the crude extracts of *Hyptis suaveolens*, *Calotropis procera* and *Striga hermonthica* consisted mainly of fatty acid methyl esters and terpenoid-related compounds were only detected in a very small amount in *Calotropis procera* and *Striga hermonthica*. In *Hyptis suaveolens* extract, GC-MS identified octadecenoic acid methyl ester (53.49%) as major constituent followed by pentadecanoic acid methyl ester (17.64%) and hexadecanoic acid methyl ester (9.54%). These findings are in agreement with previous studies, which reported fatty acids and its derivatives as major components of *Hyptis suaveolens* extracts [19, 20].

Collectively, these studies support prvious that the major phytochemical constituents of *Hyptis suaveolens* are fatty acid derivatives and they could be important contributors to its biological activity. GC-MS analysis of *Calotropis procera* similarly identified octadecenoic acid

methyl ester (37.65%) , hexadecanoic acid methyl ester (17.49%) and 9-hexadecenoic acid methyl ester (16.33%) as the major phyto-consitutents. These results are consistent with the report of [21] who identified fatty acid esters as major components of ethanolic extract of *Calotropis procera*. In *Striga hermonthica*, major phytochemical constituents were detected with relatively higher amounts of octadecenoic acid methyl ester (44.22%), pentadecanoic acid methyl ester (17.75%) and heptadecanoic acid methyl ester (13.93%). These compounds may be significant in causing insect mortality by disrupting cellular membranes, interfering with physiological processes or acting as feeding deterrents. In comparision, *Hyptis suaveolens* demonstrated the highest level of octadecenoic acid methyl ester, This finding suggested that the observed bioactivity (insecticidal and preservative activity) in *Hyptis suaveolens* may have resulted from the combined or synergistic effects of multiple phytochemicals rather than a single constituent.

Figures 1- 3 show GC-MS total ion chromatograms (TIC) of ethanolic extracts of Calotropis procera, Hyptis suaveolens and Striga hermonthica respectively.

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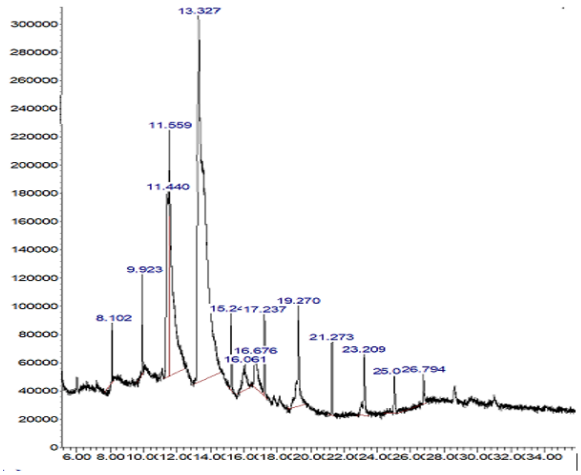

Figure 1. Total ion chromatogram of the ethanolic extract of *Hyptis suaveolens* obtained by GC–MS analysis

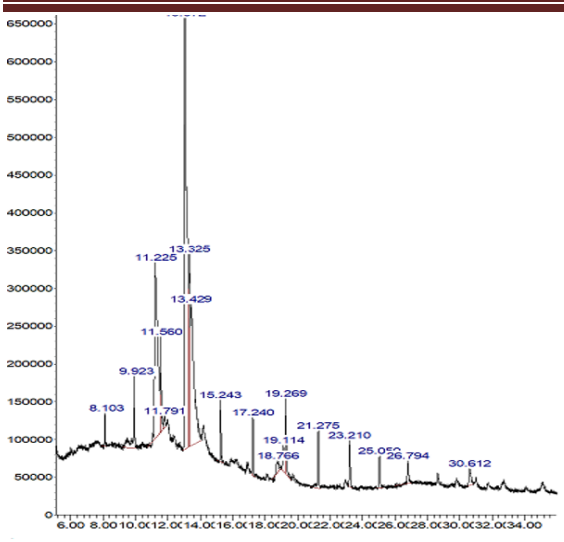

Figure 2. Total ion chromatogram of the ethanolic extract of *Calotropis procera* obtained by GC-MS analysis.

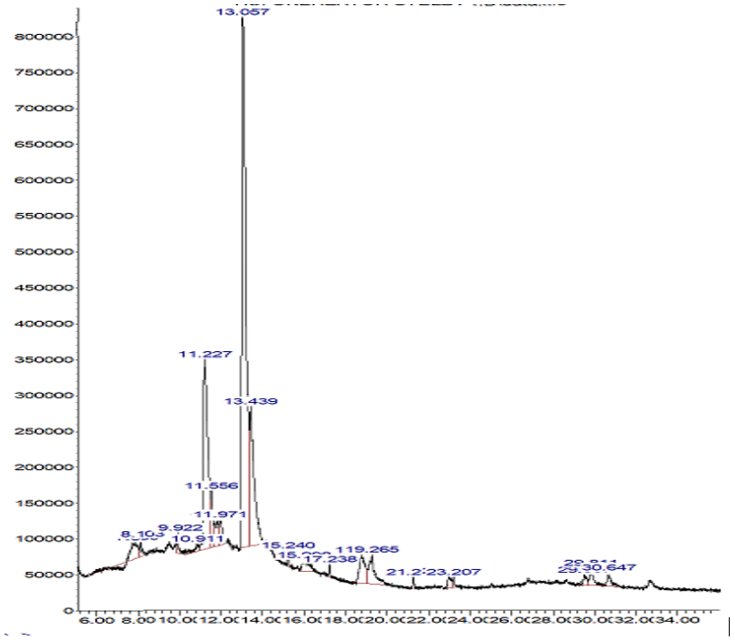

Figure 3. Total ion chromatogram of the ethanolic extract of *Striga hermonthica* obtained by GC–MS analysis**.**

The chromatograms demonstrated multiple peaks at different retention times, which suggested that the extracts contained several phytochemical components. The differences in the chemical composition of the three botanical species are indicated by the variations in the peak number,

intensity and retention times among the chromatograms. The chromatographic profiles showed a complexity that suggests that the biological activity observed is possibly the result of the combination or synergism of several bioactive constituents and not of a single compound. The present study was not intended for detailed isolation and characterization of individual phytochemical constituents and such work is recommended for further studies.

Conclusion

The present study was conducted to determine the insecticidal and grain preservative effect of crude extracts of *Hyptis suaveolens, Striga hermonthica* and *Calotropis procera* against *Callosobruchus maculatus*. Of the botanicals tested, *Hyptis suaveolens* showed the highest insecticidal activity and gave complete protection of stored cowpea grains for 120 days. Future studies should be directed towards isolation, purification and characterization of the individual bioactive constituents responsible for the observed biological activity and elucidation of their mechanisms of action and possible synergistic interactions.

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