ORIGINAL RESEARCH REPORT NIGER. J. PLT. PROT. 40(2):34-47 (2026)

IDENTIFICATION OF SECONDARY METABOLITES IN SELECTED INDIGENOUS PLANT MATERIALS AND THEIR POTENTIALS FOR STORAGE INSECT PEST MANAGEMENT

***[1]KASHERE, M.A., [2]DATTIJO, S.A.**
[1]Department of Agricultural Technology, Federal College of Horticulture P.M.B 108, Dadin Kowa, Gombe, Gombe State, Nigeria.
[2]Department of Pest Management Technology, Audu Bako College of Agriculture Dambatta, P.M.B. 3159, Kano, Kano State, Nigeria.
Corresponding Author: mkashere@gmail.com

SUMMARY

The study screened the phytochemical compounds in *Ocimum basilicum* L. whole plant, *Senna siamea* (Lam.) and *Ficus polita* Vahl stembark and provided information on their potentials for storage insect pest management. The plant materials were sourced locally in Gombe and environs subsequently identified at the Biological Sciences Department, Faculty of Science, Federal University of Kashere. Gas Chromatography-Mass Spectrometry (GC-MS) using three (3) different extraction solvents (Ethanol, Methanol and Water) was employed for the laboratory screening. Ethanol extracts of *O. basilicum* revealed 68.28% Fatty acids, 10.37% Esters and 5.74% Terpenoid. Methanol extracts showed 63.61% Fatty acids, 5.15% Terpenes and 3.75% Carboxylic acids. Water extracts revealed 20.59% Siloxanes, 5.61% Steroids and 4.65% Ethers. *S. siamea* ethanol extracts revealed 17.94% Esters, 16.44% Fatty acids, 13.18% Acetates and 12.88% Alcohols. Methanol extracts revealed 26.01% Siloxanes, 15.72% Fatty acids and 9.41% Alcohols. Water extracts revealed 42.11% Fatty acids and 6.67% Heterocyclic in *S. siamea* stembark. *Ficus. polita* ethanol extracts showed 43.07% Terpenoids, 29.34% Sesquiterpenes and 4.91% Phenols. The methanol extracts revealed 56.47% Terpenoids, 10.04% Amines, 9.10% Heterocycles and 7.62% Sterols, while water extracts revealed 43.12% Steroids, 23.90% Esters and 11.85% Terpenoids in *F. polita* stembark. Plant secondary metabolites are linked to insecticidal activities of plants. Many plant materials contained insecticidal properties such as repellant, anti-feedant, oviposition deterrent, growth inhibition either as contact toxicity or fumigant. Studies have indicated terpenoids as major examples of insecticide substances, hence, plants secondary metabolites could serve as good alternatives to chemical insecticides.

Keywords: Phytochemical, Repellant, Fumigant, Screening, stembark and solvents.

INTRODUCTION

Biopesticides of plant origin have been studied in many recent investigations, [1];[2];[3]. The use of plant material for grain preservation is considered as a promising alternative to synthetic insecticides for several decades [4]. They are biodegradable, environmentally friendly, and can also be used in insecticide resistance management programs [5]. Agricultural raw and processed commodities must be protected from insects, rodents, birds, mites, and microorganisms throughout the value chain. The mobility and the ability of many stored-product insects to find and reproduce on many different foods throughout the value chain make them

particularly difficult to manage. The use of plants in the fight against insects has been long known. Many investigations have been carried out with known and still to be utilized species of plants having pesticide characteristics [2].

Ocimum basilicum L. (African Wild Basil) known as basil, has long been acclaimed for their insecticidal potentials by different researchers [6];[7];[8];[9]. [10], found out that *O. basilicum* leaf powder caused up to 90% mortality of maize weevils within two weeks when applied at 2 g powder per 100 g of maize grains.

The insecticide activity of *Cassia siamea* extracts and pure compounds had been reported recently [11];[12]. Fresh leaves from *S. siamea* was used for repelling or killing insects such as termites, bed bugs and mosquitoes [13]. In India, *Cassia* species is used as a natural pesticide in organic farms [14]. The demonstrated larvicidal activity of *C. siamea* may also be due to presence of flavonoids, phenols, glycosides and tannins, which is in agreement with [15].

Several studies have provided baseline information on many *Ficus* spp potential validity in the treatment and management of insect pests associated with stored grains [16];[17];[18];[19]. The leaves and stem of *F. sarmentosa* var. henryi have been reported to possessed insecticide activity [20]. Application of leaves and bark extracts of *F. exasperata* was effective in the control of insect pest infestation when compared with control [16]. Fatty acids methyl esters have also been screened in *Ficus* species by many authors and reported to have larvicidal activity against the vector *C. quinquefasciatus* [21]. It has been reported that fatty acids mixture has toxicity and repellence against insecticide susceptible and resistant strains of the major malaria vector *Anopheles funestus* [22].

A number of secondary metabolites are linked to pesticides activities of plants in which they occur [23]. Monoterpenes such as eugenol, limonene, camphor and thymol commonly found in basil have strong repellent activity against insects [24]. Other studies have also indicated terpenoids as Arthropod-repellent compounds [23].

The study aimed to screen phytochemical compounds in *O. basilicum* L. (whole plant), *S. siamea* L., and *F. polita* Vahl (stembark) found in Gombe and environs and their potentials for insect pest control during storage.

MATERIALS AND METHODS

Collection and Authentication of Plant Materials

Fresh plant materials of *Ocimum basilicum* whole plant, *Sena siamea* and *F. polita* stembark were locally collected in Gombe and suburb subsequently, identified at the herbarium of Biological Sciences Department (specimen voucher numbers 089; 090 and 091), Faculty of Science, Federal University of Kashere (FUK), Gombe State, Nigeria.

Plant Materials Processing

The entire plant materials were washed thoroughly with running tap water to remove dust and soil, thereafter, sterile with distilled water. The plant materials was then grounded separately using a laboratory electric blender to get the crude extracts

Extraction of Crude Extracts of the Plant Materials

A 600g portions of the grounded plant parts were soaked separately in 1.8 litres of 70% aqueous ethanol, 1.8 litres of 70% aqueous methanol and 1.8 litres of distilled water for 24 hours to get the crude extracts. After 24hours, the crude extracts were sieved with a muslin cloth and stored in a refrigerator when not in use.

Phytochemical Screening of the Crude Extract

The processed crude extracts were aseptically carried using Laboratory sample bottles (to avoid contamination on transit) to the National Research Institute for Chemical Technology (NARICT) Basawa, Zaria, kaduna State where laboratory tests was conducted on the crude extracts of the plants specimens using standard procedures (Untargeted Gas Chromatography–Mass Spectrometry (GC-MS)).

GC - MS Experimental Conditions

The analysis was performed using Agilent Gas chromatography couple to the mass spectrometer system (model GC Agilent S/N 7890A and 5975A). HP 5ms 5% phenyl Methyl) siloxane Capillary Colum (30M x 250m) was used under the following conditions: Oven temperature 70°C for 1 min, then increase to 280°C by 10°C/min for 10 min and Injector temperature of 260°C. Helium gas was used as the carrier gas with flow rate of 1.9m/min, the volume of the injected sample was 1uL of diluted extract in ethanol, methanol and distilled water. Split injection techniques was used during sample injection with ionization energy 70ev in the electron ionization (EI) mode, ion source temperature 230°C scan mass range of M/Z 50-500.

Identification of Constituents

The constituents of the plant materials were identified base on the result obtained from the Library search and mass spectra of most of the compound with data generated under identical experimental conditions by applying a search algorithm considering the retention index as well as mass spectra similar with those of authentic compounds available in [25];[26].

RESULTS

The results in Table 1, 2 and 3 was limited to only the first four compounds with the highest percentage area (peaks) and their chromatogram in figures 1 – 9 screened from each plant material. The Compounds names, Molecular Formula, Molecular Weight, Retention Time (Min), Area (%) and Organic compounds class are presented in each of the Tables.

Table 1 shows that, the ethanol extracts of *Ocimum basilicum* whole plant revealed the highest peak area % of 68.28% from Fatty acids class of organic compounds, followed by 10.37% from Esters class and 5.74% from Terpenoid class. The methanol extracts revealed the highest peak area % of 63.61% from Fatty acids class, followed by 5.15% from Terpenes class and 3.75% from Carboxylic acids class. The water extracts revealed the highest peak area % of 20.59% from Siloxanes class, followed by 5.61% from Steroids class and 4.65% from Ethers class of organic compounds. Figures 1 – 3 shows the chromatogram for the methanol, ethanol and water extracts of *O. basilicum.*

Results of *S. siamea* Stembark extracts (Table 2) revealed the presence of various classes of organic compounds. The ethanol extracts of *S.siamea* stembark revealed the highest peak area % of 17.94% from Esters class, followed by 16.44% from Fatty acids class, 13.18% from Acetates class and 12.88% from Alcohols class of organic compounds; methanol extracts revealed the highest peak area % of 26.01% from Siloxanes class, followed by 15.72% from Fatty acids class and 9.41% from Alcohols class of organic compounds. The water extracts revealed the highest peak area % of 42.11% from Fatty acids class followed by 6.67% from Heterocyclic class of organic compounds. Figures 4 – 6 shows the chromatogram for the methanol, ethanol and water extracts of *S. siamea.*

The ethanol extracts of *F. polita* stembark (Table 3) showed the highest peak area %

of 43.07% from Terpenoids class, followed by 29.34% from Sesquiterpenes class and 4.91% from Phenols class; 56.47% from Terpenoids class, 10.04% from Amines class, 9.10% from Heterocycles class and 7.62% from Sterols class were revealed with the highest peak area % in methanol extracts. The water extracts revealed the highest peak area % of 43.12% from Steroids class, 23.90% from Esters class and 11.85% from Terpenoids class of organic compounds. Figures 7 – 9 shows the chromatogram for the methanol, ethanol and water extracts of *F. polita* stembark.

Table 1: Composition of Ethanolic, Methanolic and Distilled Water Extracts of *O. basilicum* L. Whole Plant Powders (GC – MS Analysis results)

Extraction solvent	Peak	Compounds	Molecular Formula	Molecular Weight	Retention Time (Min)	Area (%)	Organic Compounds Class
Ethanol	1	Terpinen-4-ol	C10H18O	154	5.519	1.63	Terpenoid
	2	2,2-Bis(butoxy) propionic acid, butyl ester	C15H30O4	274	10.371	10.37	Esters
	3	9,12-Octadecadienoic acid (Z,Z)-	C18H32O2	280	15.412	68.28	Fatty acids
	4	Squalene	C30H50	410	20.413	4.11	Terpenes
Methanol	1	Terpinen-4-ol	C10H18O	154	5.507	2.20	Terpene alcohol
	2	n-Hexadecanoic acid	C16H32O2	256	14.250	3.75	Carboxylic acids
	3	9,12-Octadecadienoic acid (Z,Z)-	C18H32O2	280	15.441	63.61	Fatty acids
	4	Squalene	C30H50	410	20.419	2.95	Terpenes
Distilled water	1	Thiocyanic acid, 5.alpha.-cholestan-3.beta.-yl ester	C28H47NS	429	14.977	5.61	Steroids
	2	1-Methoxy-3-(2-hydroxyethyl)nonane	C12H26O2	202	16.648	4.65	Ethers
	3	Cyclodecasiloxane, eicosamethyl-	C20H60O10Si10	740	16.865	9.55	Siloxanes
	4	Hexasiloxane, tetradecamethyl-	C14H42O5Si6	458	17.798	11.04	Siloxanes

Table 2 Composition of Ethanolic, Methanolic and Distilled Water Extracts of *S. siamea* Stembark Powders (GC – MS Analysis results)

Extraction solvent	Peak	Compounds	Molecular Formula	Molecular Weight	Retention Time (Min)	Area (%)	Organic Compounds Class
Ethanol	1	Octanoic acid, 4,6-dimethyl-, methyl ester, (4S,6S)-(+)-	C11H22O2	186	14.519	17.84	Esters
	2	cis-Vaccenic acid	C19H36O2	296	15.938	16.44	Fatty acids
	3	Z,Z-10,12-Hexadecadien-1-ol acetate	C18H32O2	280	17.088	13.18	Acetates
	4	2-Methyl-Z,Z-3,13-octadecadienol	C19H36O	280	17.678	12.88	Alcohols
Methanol	1	Cyclononasiloxane, octadecamethyl-	C18H54O9Si9	666	14.268	13.72	Siloxanes
	2	2-Methyl-Z,Z-3,13-octadecadienol	C19H36O	280	15.732	9.41	Alcohols
	3	Oleic Acid	C18H34O2	282	17.088	15.72	Fatty acids
	4	Cyclodecasiloxane, eicosamethyl-	C20H60O10Si10	740	18.359	12.29	Siloxanes
Distilled water	1	11-Octadecenoic acid, methyl ester	C19H36O2	296	14.525	5.41	Fatty acids
	2	cis-Vaccenic acid	C19H36O2	296	15.498	30.15	Fatty acids
	3	Oleic Acid	C18H34O2	282	17.695	6.55	Fatty acids
	4	1,2-Benzisothiazole, 3-(hexahydro-1H-azepin-1-yl)-, 1,1-dioxide	C13H16N2O2S	246	18.153	6.67	Heterocyclic

Table 3 Composition of Ethanolic, Methanolic and Distilled Water Extracts of *F. polita* Stembark Powders (GC – MS Analysis results)

Extraction solvent	Peak	Compounds	Molecular Formula	Molecular Weight	Retention Time (Min)	Area (%)	Organic Compounds Class
Ethanol	1	Phenol, 2-propyl-	C9H12O	136	9.885	4.91	Phenols
	2	Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl-2-(1-methylethenyl)-, [2R-(2.alpha.,4a.alpha.,8a.beta.)]-	C15H24	204	17.031	43.07	Terpenoids
	3	Cedran-diol, (8S,14)-	C15H26O2	238	17.701	20.63	Sesquiterpenes
	4	Longifolenaldehyde	C15H24O	220	19.246	8.71	Sesquiterpenes
Methanol	1	7-Oxabicyclo[4.1.0]heptane, 2,2,6-trimethyl-1-(3-methyl-1,3-butadienyl)-5-methylene-	C15H22O	218	17.043	56.47	Terpenoids
	2	Lanosterol	C30H50O	426	19.114	7.62	Sterols
	3	Bornylamine, N-cyclopropylcarbonyl	C14H23NO	221	20.413	10.04	Amines
	4	Pyrido[1,2-a]pyrimidin-4(5H)-one,9-hydroxy-2-methyl-8-nitro-	C9H7N3O4	221	20.813	9.10	Heterocycles
Distilled water	1	D-Homoandrostane, (5.alpha.,13.alpha.)-	C20H34	274	17.043	24.97	Steroids
	2	(2-Pent-2-enylcyclopentylideneamino oxy)acetic acid, methyl ester	C13H21NO3	239	17.701	23.90	Esters
	3	Lanosterol	C30H50O	426	19.183	11.85	Terpenoids
	4	9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-,(3.beta.,4.alpha.,5.alpha.)-	C30H50O	426	20.991	18.15	Sterols

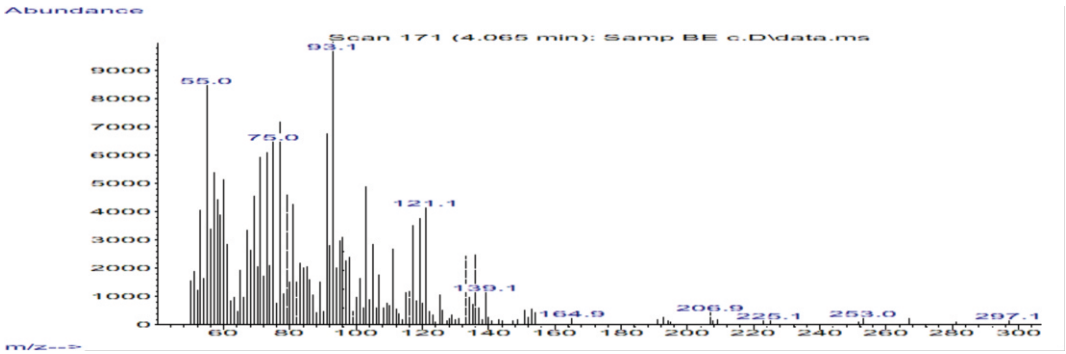


Figure 1: GC-MS Combine Chromatogram of Ethanolic Extract of *O.basilicum* Whole Plant

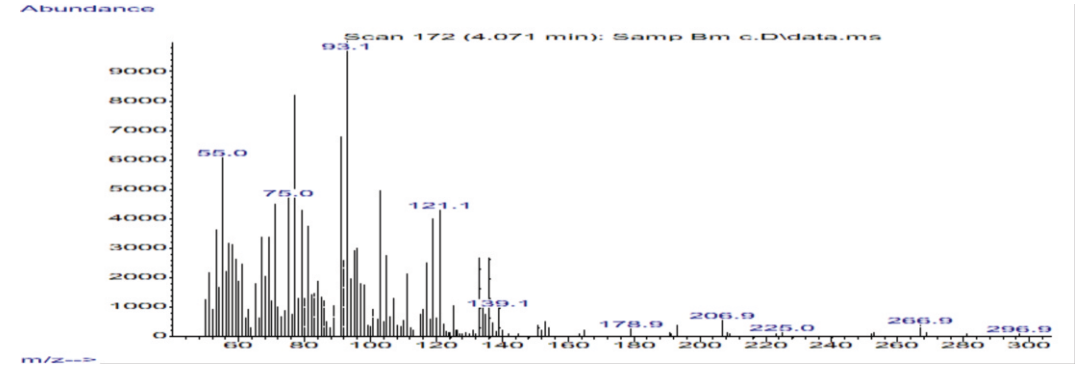


Figure 2: GC-MS Combine Chromatogram of Methanolic Extract of *O.basilicum* Whole Plant

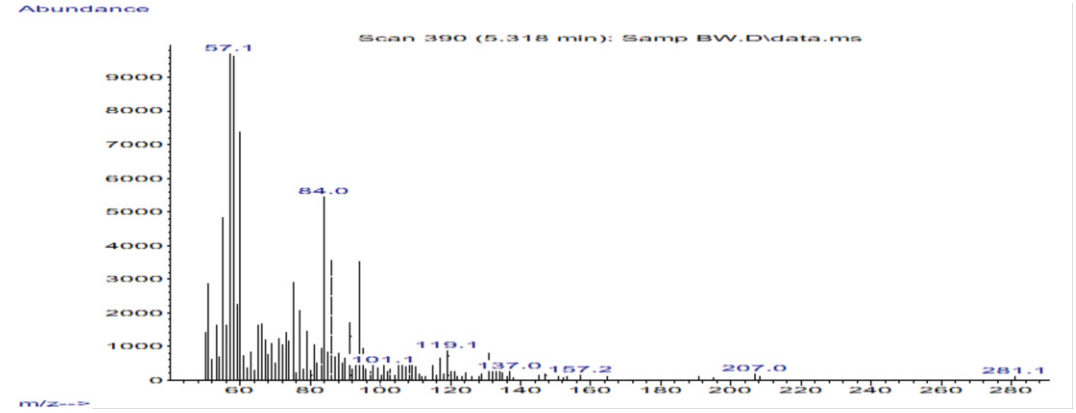


Figure 3: GC-MS Combine Chromatogram of Distilled Water Extract of *O.basilicum* Whole Plant

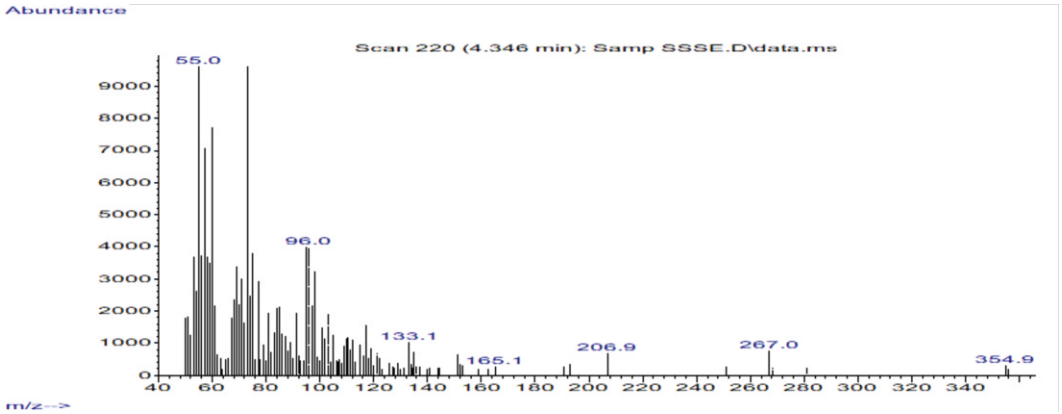


Figure 4: GC-MS Combine Chromatogram of Ethanolic Extract of *S. siamea* Stembark

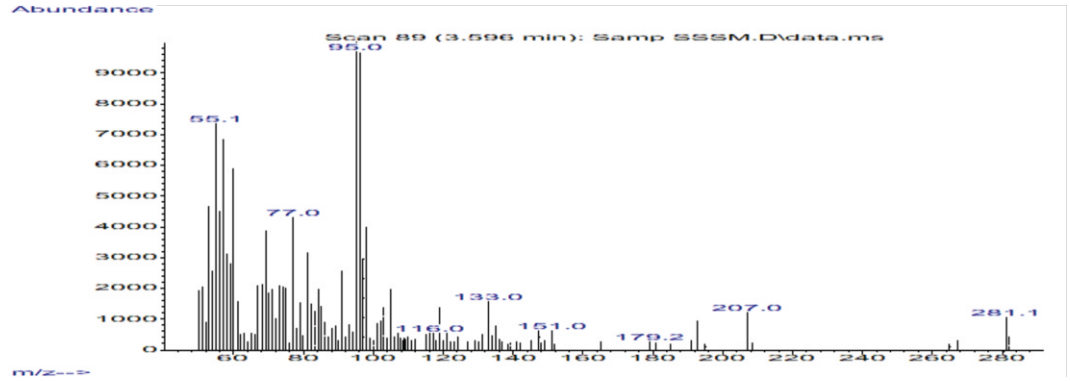


Figure 5: GC-MS Combine Chromatogram of Methanolic Extract of *S. siamea* Stembark

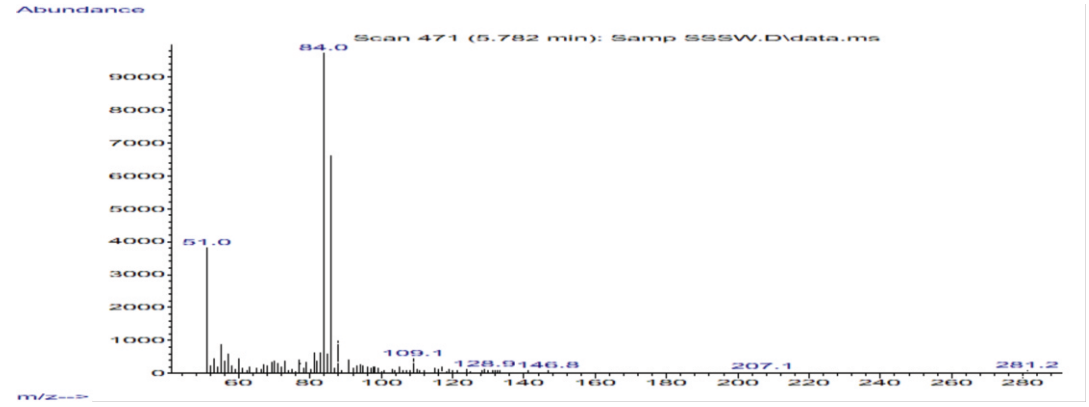


Figure 6: GC-MS Combine Chromatogram of Distilled Water Extract of *S. siamea* Stembark

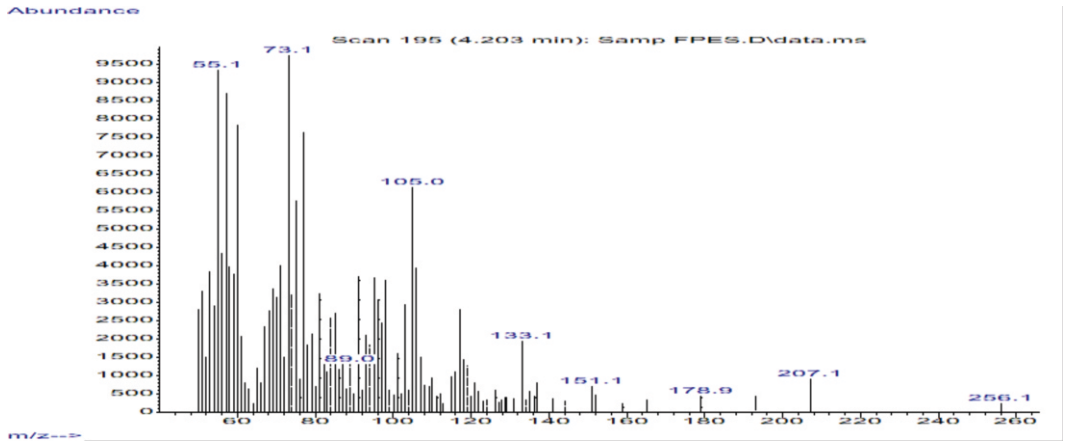


Figure 7: GC-MS Combine Chromatogram of Ethanolic Extract of *F. polita* Stembark

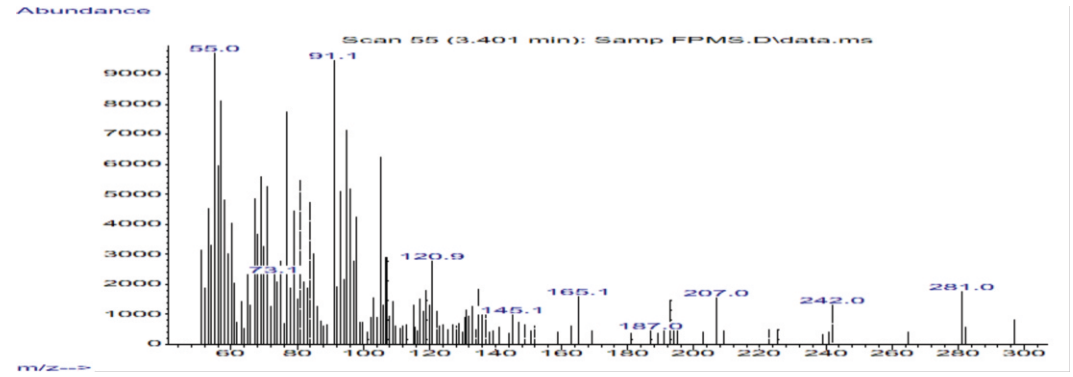


Figure 8: GC-MS Combine Chromatogram of Methanolic Extract of *F. polita* Stembark

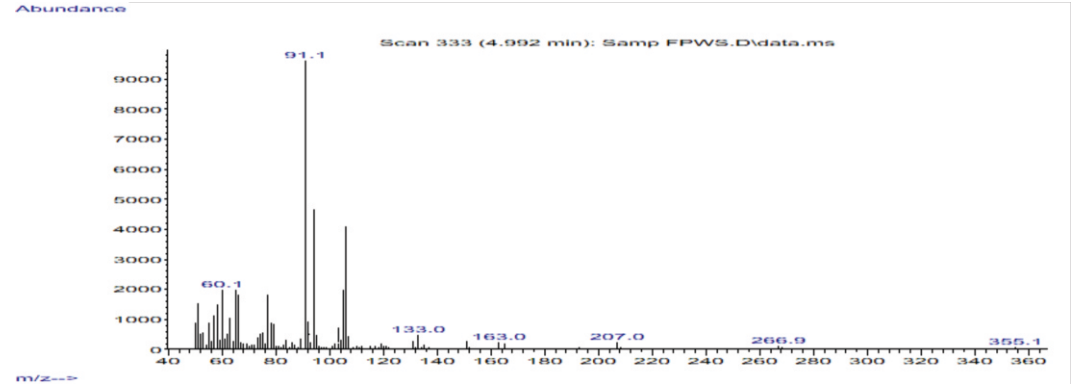


Figure 9: GC-MS Combine Chromatogram of Distilled Water Extract of *F. polita* Stembark

DISCUSSION

Results from all the three extraction solvents confirm the significant influence of solvent polarity on phytochemical extraction. **Ethanol** extracted a wide range of **moderately polar and non-polar compounds**, such as esters, alcohols, and fatty acids. **Methanol,** being more polar, yielded a **broader spectrum**, including terpenoids, phenolics, ketones, and glycosides. In contrast, **distilled water** favored **highly polar compounds**, such as aldehydes, ketones, amines, and carboxylic acids.

This variability is crucial for selecting plant extracts with targeted bioactivities in pest management. *Ocimum basilicum* L. (whole plant), *S. siamea* L., and *F. polita* Vahl (stembark) demonstrated potential for insecticidal applications, particularly due to the presence of terpenes, phenols, glycosides, and fatty acids, which are known to possess bioefficacy against stored-product pests [27];[28];[29].

The identified phytochemical classes, particularly terpenoids, fatty acids, and esters, are known for their insecticidal properties. Terpenoids, for instance, have been documented to exhibit toxic, repellent, and deterrent effects on insects [27]. They can interfere with insect neurotransmission by inhibiting acetylcholinesterase activity and modulating gamma-aminobutyric acid (GABA) receptors, leading to neurotoxicity in pests.

Fatty acids and esters also contribute to insecticidal activity Anti-inflammatory, larvicidal [28];[29]. Fatty acids can disrupt insect cell membranes, leading to cell lysis, while esters may act as feeding deterrents or oviposition inhibitors.

Extracts of the plant (*O.basilicum*) have been shown to contain biologically active constituents that are insecticidal, nematicidal, fungistatic, and antimicrobial [30];[31];[32]. [10], found out that *O. basilicum* leaf powder caused up to 90% mortality of maize weevils within two weeks when applied at 2 g powder per 100 g of maize grains. Other studies have also shown phenolic compounds of basil to have ovicidal, larvicidal and nymphicidal properties [33].

A number of authors isolated and identified several compounds from different *Cassia* species such as anthraquinones, anthracenes, polyphenols, fatty acids, sterols, polysaccharides, alkaloids, glycosides, coumarins, chromones, terpenenoids, tannin, saponins, sterols, polyphenols, flavonoids, volatile oils and some other miscellaneous compounds from different *Cassia* species [34];[35];[36]. The insecticide activity of *Cassia siamea* extracts and pure compounds has been reported recently. Fresh leaves from *S. siamea* is used for repelling or killing insects such as termites, bed bugs and mosquitoes [13]. In India, Cassia species is used as a natural pesticide in organic farms [14].

Phytochemical compounds extracted from members of the *Ficus* have received attention recently from applied entomologists due to their excellent properties as control agents for insects. [37], afforded the identification of 27 compounds in the leaf essential oil, 17 in the stem bark and 14 in the root. The leaf oil composition comprised of 32.3% monoterpenoid which are 21.0% monoterpene hydrocarbon and 11.3% oxygenated monoterpene. The sesquiterpenoid fraction (39.7%) was identified as 21.5% sesquiterpene hydrocarbons and 18.2% oxygenated sesquiterpenes. Other constituents were

characterized as 3.1% diterpene, 6.9% fatty acids and 8.4% simple aliphatic. Systematic investigations of members of the *Ficus* family for their insecticidal potential have been undertaken [38];[20];[39].

The presence of these compounds in the plants extracts suggests their potential utility in developing botanical insecticides for protecting stored products.

Several compounds identified in this study are known for their insecticidal properties:

i. Terpenoids: Function as repellents, feeding inhibitors, and growth regulators [40].
ii. Phenolics: Exhibit contact toxicity and fumigant action [41].
iii. Steroids and Androstanes: Impede hormonal functions in insects [42].
iv. Heterocyclic Compounds: Potential neurotoxic agents against insect pests [43].
v. Cyclopentadiene derivatives: Insecticidal, fragrance precursor [44].
vi. Phenols, tannins and flavonoids have shown to interface with many biological activities of insects [27].

The findings support the potential integration of these plant extracts into pest management strategies, particularly for storage pests [38];[20];[39]. Utilizing botanical insecticides derived from plants could offer environmentally friendly alternatives to synthetic chemicals, reducing the risk of resistance development in pests and minimizing environmental contamination. Their availability makes them inexpensive and hence they are easily incorporated into agricultural production systems [45]

CONCLUSION AND RECOMMENDATION

The phytochemical screening of *O. basilicum* L. (whole plant), *S. siamea* L., and *F. polita* Vahl (stembark) revealed a diverse spectrum of organic compound classes, varying significantly across extraction solvents. Ethanol and methanol were particularly effective in extracting moderately polar and polar bioactive compounds, including terpenoids, phenolics, esters, and glycosides, while distilled water was more selective for highly polar constituents such as aldehydes, amines, and carboxylic acids.

These phytochemicals are associated with various biological activities, including insecticidal, antimicrobial, and antioxidant properties. The identification of key secondary metabolites such as terpenes, fatty acids, phenols, and glycosides in *O. basilicum* L. (whole plant), *S. siamea* L., and *F. polita* Vahl (stembark) provides a scientific basis for their potential use in sustainable pest management strategies, especially for stored grain protection.

The findings also suggest that the identified secondary metabolites can be developed into eco-friendly biopesticides suitable for smallholder farmers.

From the findings of the study, it is recommended that Future work should focus on:

i. Developing bio-formulations from these extracts could enhance the applicability of botanical pesticides as eco-friendly alternatives to synthetic chemicals in agricultural systems.
ii. Assessing toxicity on non-target organisms and long-term efficacy in storage systems.

Conflicts of Interest: The author's declare no conflicts of interest.

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