

Chemical Composition, Antioxidant, and Anticholine Esterase Activities of Essential Oil of *Xylopiya aethiopic* Seeds

Lateef Adegboyega Sulaimon, Rahmat Adetutu Adisa¹, Efere Martins Obuotor², Modinat Olasunkanmi Lawal, Abdullahi I. Moshood, Nura H. Muhammad

Department of Chemical Sciences, Crescent University, Abeokuta, ¹Department of Biochemistry, University of Lagos, Lagos, ²Department of Biochemistry, Obafemi Awolowo University, Ile-Ife, Nigeria

ABSTRACT

Background: *Xylopiya aethiopic* is well known to treat neurodegenerative diseases in traditional medicine and there is no scientific evidence for this claim. **Objective:** The current research aimed at investigating chemical characterization, antioxidant, and anticholine esterase activity of the essential oil from *X. aethiopic*. **Materials and Methods:** Essential oil extraction was carried out by the use of the steam distillation method in a modified Clevenger-type apparatus. The chemical composition of the essential oil from seeds of *X. aethiopic* (African pepper) was determined using gas chromatography coupled with mass spectrometry (GC-MS) and its potentials as antioxidant and anticholine esterase were evaluated for the first time. **Results:** The oil yield was 5.2% (v/w) in *X. aethiopic*. The GC-MS analysis identified a total of 52 compounds corresponding to 100% of the total oil in *X. aethiopic*. The major constituents of *X. aethiopic* essential oil are terpinen-4-ol (11.88%), α -terpineol (5.93%), cyclohexane methanol (4.79%), and β -copaene (4.74%). The most abundant classes of compounds from the essential oil were oxygenated monoterpenes (MT) which amounted to (37.6%), followed by oxygenated sesquiterpenes (29.61%), sesquiterpenes (14.67%), oxygenated diterpenes (9.05%), nonterpenoid aliphatic and aromatic hydrocarbons (4.81%), diterpenes (3.8%), and MT (0.47%) of all the identified constituents. A significant antioxidant (IC₅₀ value of 2,2-diphenyl-1-picrylhydrazyl = 2.19 ± 0.09 mg/mL) and anticholine esterase activity (IC₅₀ = 1.21 ± 0.06 mg/mL) was obtained for the essential oil of *X. aethiopic*. **Conclusion:** The study established the chemical composition, antioxidant, and anticholine esterase activities of the essential oil of the plant seeds.

Key words: Anticholine esterase, antioxidant, monoterpenes, sesquiterpenes, *Xylopiya aethiopic*

SUMMARY

- Steam distillation method was used to extract essential oil from the seeds of *Xylopiya aethiopic* with the yield of 5.2% (V/W)
- Fifty two (52) compounds were identified in *X. aethiopic* essential oil. Terpinen-4-ol, α -terpineol, cyclohexane methanol and β -copaene are the major constituents in *X. aethiopic* essential oil
- Essential oil of *X. aethiopic* seeds showed a significant antioxidant and Anticholine esterase activity.

Abbreviations Used: ROS: Reactive oxygen species, AD: Alzheimer's disease, DPPH: 2,2-diphenyl-1-picrylhydrazyl, AChE: Acetyl choline esterase, BChE: Butyryl choline esterase, DTNB: 5, 5'-Dithiobis-(2-nitrobenzoic acid), GC: Gas chromatography, GC-MS: Gas chromatography-mass spectrometry, FRAP: Ferric-reducing antioxidant power, FID: Flame ionizing detector, TAC: Total antioxidant capacity, MT: Monoterpenes, OM: Oxygenated monoterpenes, ST: Sesquiterpenes, OS: Oxygenated sesquiterpenes, NH: Nonterpenoid hydrocarbons, DT: Diterpene, OD: Oxygenated diterpenes, OST: Oxygenated sesterterpene, NS: Not significant, SD: Standard deviation, AAE: Ascorbic acid equivalent, RT: Retention time, RI: Retention index.

Correspondence:

Mr. Lateef Adegboyega Sulaimon,
Department of Chemical Sciences, Crescent
University, Abeokuta, Nigeria.
E-mail: adlat4best@yahoo.com
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INTRODUCTION

A brief history of medicine demonstrates the use of herbal medicine for the effective treatment of various ailments. Herbal medicine has been used since long in various forms including the decoction, powdered sample, oleoresins, crude extracts, fixed oil, essential oil, etc.^[1] Various plants have been used in multiple types of food items for preservation and therapeutic effects.^[2] In this regard, essential oils have been manifested by several reporters to play a major role. Essential oils have the property to attenuate the effects of free radicals, for example, reactive oxygen species (ROS) which are derived from the metabolism of oxygen and exogenous agents.^[3] ROS are responsible for wide variety of diseased conditions including oxidative stress and nervous disorders.^[4] Essential oils are well known for their radicals scavenging properties and amelioration of various cognitive disorders. Among the cognitive disorders, Alzheimer's disease (AD) is the most common in elderly people.^[5] One of the best therapeutic approaches for AD is to increase the concentration of the neurotransmitter (Acetylcholine) by inhibiting the enzyme (acetyl cholinesterase) responsible for its breakdown. Various drugs originated either from natural or synthetic sources are being used for the management of AD and other nervous disorders.^[6] Similarly, it has also been reported that oxidative stress are responsible for wide variety of mental diseases due to neuronal

degeneration and other factors. Oxidative stress is mainly developed due to increase in concentration of free radicals within the body. The free radicals have been reported by numerous researchers to possess multiple destructive properties, and the research interest has been focused on scavenging the free radicals and avoiding their deteriorating effects.^[7] In this context, investigators are trying to explore more and more sources of natural and synthetic bioactive principles.^[8] The natural drugs are being preferred over the synthetic due to their negligible harmful and deleterious effects.^[9] That is why researchers are trying to explore novel sources of natural medicine.^[10-18] Among the natural sources, herbal medicines have been shown promising results due to the presence of numerous secondary metabolites and essential

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oil. Essential oils isolated from various plants have been reported to possess marked acetyl cholinesterase inhibitory and radicals scavenging potentials.^[19-21] Traditional knowledge also demonstrates the use of essential oil for various nervous system disorders.^[22]

Xylopia aethiopica is a deciduous tree, belongs to the family Annonaceae. It is popularly known as “African pepper,” “Ethiopian pepper” or “Guinea pepper.” It is a small tree which can reach 20 m in height. Its fruits are used as spice and stimulate the appetite. It is widely distributed in the West African rainforest from Senegal to Sudan in Eastern Africa and down to Angola in Southern Africa.^[23] Its organs are used in African traditional medicine, only or in association with other plants for the treatment of the skin infections, cholera, dysentery, and the hernia.^[24]

X. aethiopica is a medicinal plant of great repute in West Africa and contains a variety of complex chemical compounds.^[25] Almost every part of the plant is used in traditional medicine for managing various ailments, including skin infections, candidiasis, dyspepsia, neurodegenerative disease, cough, and fever.^[26] A major advantage of using *X. aethiopica* as a food preservative is that foods preserved by this spice may qualify as a functional food since it has many health benefits such as anti-tumor, anti-asthmatic, anti-inflammatory, antioxidant, antimicrobial,^[27] hypotensive, and coronary vasodilators effects.^[28] The seeds contain bitter principles, alkaloids, terpenes, glycosides, saponins, tannins, sterols, carbohydrate, protein and free fatty acid, mucilage, and acidic compounds; some of which might be responsible for its reported uses.^[29] To date, the chemical composition of essential oil of *X. aethiopica* seeds has not been reported or evaluated for any pharmacological activity. Based on the literature survey and medicinal importance of *X. aethiopica*, the current investigational study was arranged to isolate the essential oil, analyze the chemical composition, and to evaluate for the anticholinesterase and antioxidant potentials, which may be a possible remedy for oxidative stress and nervous system disorders.

MATERIALS AND METHODS

Plant material

Dried fruits of *X. aethiopica* were purchased from the Itoku market, Abeokuta, Ogun State, Nigeria. The fruits were identified and authenticated at the Biological Sciences Department, Crescent University Abeokuta with a voucher number CUH 1220.

Chemicals

DPPH (Sigma Aldrich, Germany), $K_2S_2O_4$ (Riedel-de Haen Germany), Folin Ciocalteu reagent (Merck Co. Germany), acetylcholinesterase (AChE) (Electric eel type-VI-S, Sigma-Aldrich Germany), butyrylcholinesterase (BChE) (Equine serum Lyophilized Sigma-Aldrich), Acetylthiocholine iodide (Sigma-Aldrich Germany), Butyrylthiocholine Iodide (Sigma-Aldrich), DTNB (Sigma-Aldrich Germany). The entire chemicals used were of analytical grade.

Essential oil extraction

Extraction of the essential oil was carried out in the Department of Biochemistry and Molecular Biology, Obafemi Awolowo University, Ile-Ife, Osun State. The seeds were separated from the shaft and grinded to a coarse powder using a manual blender. Two kilogram (2 kg) of the coarse powder was weighed and loaded into a flat-bottomed distillation tank that formed part of the modified Clevenger-type apparatus.^[30] Four liters of water was poured into the tank and the lid secured tightly. The powdered seeds were then subjected to steam distillation with the collection of the oil starting after a heating time of 50 min and continued until no more essential oil was obtained (5–8 h). The volatile oil was collected from the top of the hydrosol and dried over anhydrous sodium sulfate (Na_2SO_4). The oil was filtered using Whatman filter paper (No. 1), weighed and

collected into 3 ml airtight glass vials. The essential oil was then stored at -20°C in a freezer until when required for chemical analysis.

Gas chromatography analysis

The gas chromatography (GC) analysis of essential oil was carried out through gas chromatograph Agilent USB-393752 (Agilent Technologies, Palo Alto, CA, USA) with HHP-5MS 5% phenyl-methyl siloxane capillary column ($30\text{ m} \times 0.25\text{ mm} \times 0.25\text{ }\mu\text{m}$ film thickness; Restek, Bellefonte, PA, USA) connected with flame ionizing detector. The oven was set at temperature of 50°C for 1 min and then increased to 100°C at the rate of $5^\circ\text{C}/\text{min}$ for 10 min and lastly to 280°C at the rate of $10^\circ\text{C}/\text{min}$ for 20 min. The temperature of injector and detector were maintained at 220°C and 290°C correspondingly. The flow rate of carrier gas, i.e., helium was 1 ml/min and the diluted samples (1/1000 inn-pentane, v/v) of 1 μl were manually injected in the split-less mode.

Gas chromatography–mass spectrometry analysis

The gas chromatography–mass spectrometry (GC/MS) of the essential oil was performed through USB-393752 gas chromatograph (Agilent Technologies, Palo Alto, CA, USA) with an HHP-5MS 5% phenyl-methyl siloxane capillary column ($30\text{ m} \times 0.25\text{ mm} \times 0.25\text{ }\mu\text{m}$ film thickness; Restek, Bellefonte, PA, USA) outfitted with an Agilent HP-5973 mass selective detector in the electron impact mode (Ionization energy: 70 eV) working under the experimental conditions as those maintained for GC.

Identification of components

The recognition of all the major constituents of oil was performed by comparing their retention times with the authentic compounds in the literature. Identification of compounds was further processed through the spectral data obtained from the Wiley and NIST libraries as well as fragmentation patterns comparisons of the mass spectra with data reported in literature or with those of mass spectra from literature.^[31,32]

Anticholinesterase assay

Anticholinesterase (AChE and BChE inhibitions) activity was performed for the essential oil of *X. aethiopica* by spectrophotometric analysis following the method of Ellman's assay.^[33] The substrates used were acetylthiocholine iodide and butyrylthiocholine iodide. Briefly, 5 μL of 0.03 U/mL AChE and 0.01 U/mL BChE were taken in a cuvette and 205 μL of essential oil having concentration of (156.25–5000) $\mu\text{g}/\text{mL}$ were transferred to them using micropipette. Similarly, 100 μL of DTNB was also added to this afterward. The mixtures obtained were kept in water bath for 15 min at the temperature of 30°C . After incubation, 200 μL of the Substrates were added to the mixture to optimize the reaction. A double beam spectrophotometer was used to measure the reaction time at 412 nm through a double beam spectrophotometer (Thermo electron corporation USA). Absorption values were obtained for 4 min. Meanwhile, the yellow color mixtures indicated the formation of 5-thio-2-nitrobenzoate anion as a reaction product of thiocholines and DTNB. White assay was also performed without enzymes and plant samples to check the non-enzymatic hydrolysis of the substrate. The mixture which contained all the components excluding essential oil was marked as control. Percentage inhibition was recorded as follows:

$$\% \text{ inhibition} = (E - S)/E \times 100.$$

Where *E* is the activity of the enzyme without sample and *S* is the activity of enzyme with the test sample.

2,2-Diphenyl-1-picrylhydrazyl radical scavenging assay

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging potential was evaluated for essential oil of *X. aethiopica* following

the previously described procedure.^[34] DPPH solution (0.004%) was prepared in methanol to get a deep violet color solution. Similarly, stock solution of essential oil was prepared in ethanol having concentration of 1 mg/mL. The stock solution was serially diluted to get the concentrations of 156.25–5000 µg/mL. Afterward, 0.1 mL of each concentration was added to the 3 mL of DPPH solution. The mixture obtained was incubated at 23°C for 30 min in dark. After incubation, the absorbance of each sample was recorded at the wavelength of 517 nm using double beam spectrophotometer. Ascorbic acid was used as the positive control. All the samples were processed in triplicates and the percent activity was recorded as mean ± standard error of the mean. The percent radical scavenging potential was figured out using the following formula;

$$([A_c - A_0]/A_c) \times 100; \text{ where } A_c = \text{Absorbance of control and } A_0 = \text{Absorbance of sample.}$$

Estimation of IC₅₀ values

The median inhibitory concentration, i.e., IC₅₀ values of AChE, BChE, and DPPH were determined by a linear regression analysis of the percent inhibition versus the concentrations of test samples through MS Excel program.

Ferric-reducing antioxidant power assay

Ferric-reducing antioxidant power (FRAP) assay of extracts was carried out according to method.^[35] The FRAP reagent consists of 300 mM acetate buffer pH 3.6, 10 mM TPTZ (2, 4, 6-tripyridyl-s-triazine), and 20 mM FeCl₃.6H₂O solution. A volume of 150 µl plant extracts were mixed to FRAP reagent, allowed to stand for 6 min and absorbance was noted at 593 nm in a microplate reader. The antioxidant activity of the extract was expressed as the number of mg equivalents to ascorbic acid (AAE)/g extract. All the experiments were carried out in triplicates.

Total antioxidant capacity

Total iron-reducing power assay was carried out by taking 100 µl of extract (2 mg/ml) in individual test tubes added 2.5 ml buffer, 1.5 ml potassium ferrocyanide (1%), and 100 µl FeCl₃ then the reaction mixtures were incubated for 30 min at RT. The absorbance was noted at 700 nm in a microplate reader.^[35] The antioxidant activity of the extract was expressed as the number of mg AAE/g extract. All the experiments were carried out in triplicates.

Statistical data analysis

All the tests were conducted in triplicate and the values were tabulated as mean ± standard deviation.

RESULTS AND DISCUSSION

In the current investigational study, the essential oil obtained by steam distillation from seeds of *X. aethiopica* yielded 5.2% (v/w) on dry weight. This result is comparable to those previously obtained by Tegang *et al.*^[36] who found extraction yield of 4.2% for the essential oil of dried fruits of *X. aethiopica* from Cameroon. Bakarnga-Via *et al.*^[37] earlier reported the yields of 3.57% and 4.68%, for dried fruits of *X. aethiopica* from Chad and Cameroon, respectively. However, the extraction yields of the essential oil of dried fruits of *X. aethiopica* obtained from the local market in Keffi, Nasarawa State, Nigeria, was lower 1.2%.^[38]

Antioxidants play a crucial role in the management and treatment of neurodegenerative conditions, as the role of free radicals in such conditions has been well established since brain cells consume a high amount of oxygen.^[39] The DPPH radical scavenging activity, total antioxidant capacity (TAC) and FRAP assays exhibit the antioxidant property of the essential oil used in this study. The stable organic radical

DPPH has been widely employed in studies of the antioxidant capacity of essential oil.^[40] The DPPH (1, 1-diphenyl-2-picrylhydrazyl) test measures the hydrogen atom or electron donor capacity of the essential oil to the stable radical DPPH formed in solution.^[41] It also measures the ability of the essential oil to scavenge free radicals in solution.

The radical scavenging potential of volatile oil was studied based on spectrophotometric analysis. The source of free radicals employed was DPPH, which has maximum absorbance value at 517 nm. After getting scavenged by antioxidant compounds, the color of DPPH (violet) solution changes into yellow. Change in the color results in decrease of absorbance values, which is directly proportional to the amount of radical scavenging compounds in the solution.^[42,43] In our current investigational study, the free radicals scavenging assay of essential oil of *X. aethiopica* against DPPH (IC₅₀ value = 2.19 ± 0.09 mg/ml) was significant and almost comparable with the positive control (IC₅₀ value = 0.013 mg/ml). From Table 1, it is clear that essential oil exhibited marked antioxidant potential with TAC and FRAP values of 10.92 ± 0.63 mg AAE/g and 10.03 ± 0.25 mg AAE/g, respectively. Our findings are in contrary to the study of Adefegha *et al.*,^[44] who reported high IC₅₀ values of 37.3 µL/L and 62.5 µL/L for DPPH and nitric oxide radical scavenging activities of *X. aethiopica*, respectively.

The available literatures on etiology of diseases demonstrate multiple causative agents responsible for specific disease.^[45] In the context of AD, numerous investigators have reported the role of various causative agents along with various successful approaches.^[46] Like all neurodegenerative disorders, the free radicals have a prominent role in the induction and progression of AD.^[47] By avoiding or attenuating the causative agents, one can hinder the progression of a specific disease. In the case of neurodegenerative disorders, the scavenging of free radicals can be a vital target. Various researchers have demonstrated the effective role of natural antioxidants, especially the essential oil to combat the free radicals.^[48] Similarly, one of the most widely employed treatment strategies for AD, i.e., the inhibition of AChE to increase the concentration of neurotransmitter is highly recommended.^[49] The inhibition of AChE and BChE, however, has been accepted as an effective approach toward the treatment and management of AD.^[50,51] The anticholinesterase and antioxidant potentials of essential oil of *X. aethiopica* seeds are summarized in Table 1. Based on the IC₅₀ values for the concentration-dependent inhibition of AChE and BChE, the essential oil of *X. aethiopica* seeds showed high AChE (IC₅₀ = 1.21 ± 0.06 mg/mL) and BChE (IC₅₀ = 4.39 ± 1.38 mg/mL) inhibitory effects. Adeniyi *et al.*^[44] previously reported similar results of the inhibitory effects of *X. aethiopica* on AChE (IC₅₀ = 18.5 µL/L) and BChE (IC₅₀ = 26.4 µL/L). It is worth noting that the essential oil of *X. aethiopica* seeds showed an AChE inhibition stronger than the BChE inhibition. Therefore, the oil may be of immense therapeutic importance in that they may enhance nerve cell communication, improve cognitive effects and manage AD.^[52,53] The essential oil obtained from various plants possesses marked anti-Alzheimer's potential due to the presence of wide variety of valuable compounds in them.^[54,55] The anticholinesterase activity of essential oil

Table 1: Anticholine esterase and antioxidant activity of essential oil of *Xylopia aethiopica*

Assays	Units	Mean±SD
Acetylcholine esterase inhibition	IC ₅₀ (mg/ml)	1.21±0.06
Butyrylcholine esterase inhibition	IC ₅₀ (mg/ml)	4.39±1.38
DPPH radical scavenging activity	IC ₅₀ (mg/ml)	2.19±0.09
TAC	mgAAE/g	10.92±0.63
FRAP	mgAAE/g	10.03±0.25

TAC: Total antioxidant capacity; DPPH: 1, 1 -diphenyl-2 picrylhydrazyl; FRAP: Ferric-reducing antioxidant power; SD: Standard deviation; IC₅₀: Half maximal inhibitory concentration

of *X. aethiopica* might be due to its hydrophobic nature because of the good affinity of hydrophobic active site of AChE.^[56,57]

The GC-MS analysis of essential oil of *X. aethiopica* demonstrates a total of 52 components as shown in Table 2. The ion chromatogram indicating 62 phyto-components in the essential oil of *X. aethiopica* (with 62 peaks) is shown in Figure 1. However, some of the components appeared more than once with different retention time and this accounts for fifty-two different components of *X. aethiopica* essential oil in the present study. Some of the most common components of essential oil, i.e., terpinen-4-ol, carveol, eugenol, α -copaene, α -terpineol, Caryophyllene, α -muurolene,

guaiol, thunbergol, geraniol, farnesol acetate, spathulenol, and isolongifolol have been found in the essential oil of *X. aethiopica* seeds. Ayedoun *et al.*^[58] identified elemol and guaial (among other terpenes) in the essential oil of the fruit of *X. aethiopica* from the Republic of Benin. Previous investigations on the essential oil of the fruits of *X. aethiopica* from various areas revealed that they mainly consist of α - and β -pinene, myrcene, *p*-cymene, limonene, linalool, terpinen-4-ol, α -terpineol, and 1,8-cineole.^[59-62] Cumine aldehyde and sabinene have been identified in the fruit essential oil of *X. aethiopica* from Nigeria^[63] in contrast to our study, where both are practically absent. Furthermore, β -pinene, which

Table 2: List of components of essential oil of *Xylopia aethiopica*

Number	RT (min)	Compound name	RI	Class	Concentration (%)
1	5.149	4-Isopropyl-1-methyl-2-cyclohexen-1-ol	1109	OM	1.50
2	5.341	Pinocarveol	1131	OM	4.39
3	5.455	Dihydrocarveol	1196	OM	1.88
4	5.817	Terpin-4-ol	1137	OM	11.88
5	6.063	α -Terpineol	1143	OM	5.93
6	6.184	Myrtenol	1191	OM	3.90
7	6.274	Piperitol	1175	OM	1.31
8	6.316	Carveol	1206	OM	0.85
9	6.403	3-Allyl-2,6,6-trimethyl-Bicycloheptane	1187	NH	3.13
10	6.933	Myrtenal	1136	OM	2.12
11	7.089	2H-Inden-2-one	1237	OM	0.33
12	7.168	Naphthalene	1185	NH	0.37
13	7.306	Perillic Alcohol	1261	OM	1.57
14	7.482	<i>p</i> -Ethylguaiacol	1303	NH	1.23
15	7.553	3-Isopropenyl-5-methyl-1-cyclohexene	990	MT	0.47
16	7.737	α -Copaene	1221	ST	2.95
17	7.997	Myrtenyl acetate	1314	OM	0.46
18	8.063	Eudesma-4 (14), 11-diene	1469	ST	1.45
19	8.280	β -Copaene	1216	ST	5.04
20	8.382	Eugenol	1392	OM	0.63
21	8.509	Caryophyllene	1494	ST	1.26
22	8.558	Cis-Isoeugenol	1410	OM	0.85
23	8.896	γ -Muurolene	1435	ST	1.52
24	8.938	1H-3a, 7-methanoazulene	1398	ST	0.79
25	9.128	α -Muurolene	1440	ST	0.49
26	9.357	Eremophila-1 (10), 11-diene	1474	ST	0.73
27	9.949	Aromadendrene oxide	1462	OS	0.90
28	10.038	(-)-Spathulenol	1536	OS	4.49
29	10.407	11- α -Hydroxy-pregn-4-ene-3,20-dione	2435	OD	1.91
30	10.582	Cyclohexane methanol	1522	OS	4.79
31	10.817	Aristolene epoxide	1293	OS	0.87
32	10.978	Guaial	1614	OS	1.47
33	11.042	Naphthalen-2-ol	1690	OS	2.47
34	11.211	Alloaromadendrene	1386	ST	0.44
35	11.550	γ -Eudesmol	1626	OS	2.21
36	11.676	Alloaromadendreneoxide	1386	OS	1.68
37	11.930	Aromadendrene	1462	OS	2.37
38	12.022	Cyclopropane carboxylic acid ester	2345	OD	1.03
39	12.145	α -Eudesmol	1598	OS	2.74
40	12.234	Eudesm-4 (14)-en-11-ol	1593	OS	2.40
41	12.331	12-Oxabicyclo[9.1.0] dodeca-3,7-diene	1592	OS	0.66
42	12.435	Bicyclo[5.3.1] undec-1-en-8-ol	1754	OS	2.41
43	12.892	Trans-Geranyl geraniol	2192	OD	0.85
44	14.739	Farnesol acetate	1834	OS	0.15
45	16.524	Labd-4-ene	1978	OD	4.14
46	17.167	Thunbergol	2211	OD	0.07
47	18.017	1-Phenanthrene carboxylic acid	2193	OD	0.15
48	19.127	Kauran-16-ol	1934	OD	0.71
49	19.370	Labd-14-en-3-one	2149	OD	0.19
50	20.033	Kaur-16-ene	1789	DT	3.80
51	21.268	Ethanol, 2-butoxy phosphate ester	0	NH	0.08
52	21.325	10-12-Pentacosadiynoic acid	2897	OST	NS

RT: Retention time; RI: Retention index; MT: Monoterpenes; ST: Sesquiterpenes; OM: Oxygenated MT; NH: Nonterpenoid hydrocarbon; OS: Oxygenated ST; OD: Oxygenated diterpenes; DT: Diterpene; OST: Oxygenated sesterterpene

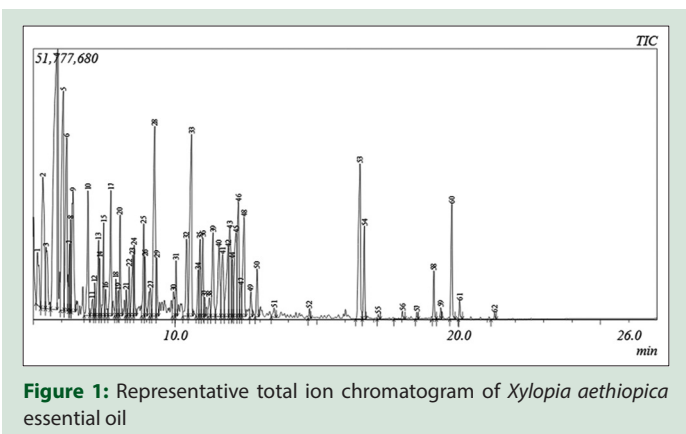


Figure 1: Representative total ion chromatogram of *Xylopi aethiopica* essential oil

is predominantly present in essential oil of *X. aethiopica* fruits from Ghana^[64] and Cameroon,^[36] is not found in essential oil of *X. aethiopica* seeds. Some of these components have been reported previously by other investigators to possess antioxidant and anticholinesterase potentials.^[65-73] Going to the detail of various components of essential oil of *X. aethiopica*, it is clear that the marked antioxidant and anticholinesterase activities shown by the essential oil is observed due to the presence of wide variety of compounds in it.

Fifty-two constituents, representing 100% of *X. aethiopica* essential oil, were identified in the present study [Table 2]. The major components of essential oil of *X. aethiopica* seeds are terpinen-4-ol (11.88%), α -terpineol (5.93%), β -copaene (5.04%), cyclohexane methanol (4.79%), (-)-spathulenol (4.49%), pinocarveol (4.39%), and labd-4-ene (4.14%). Karioti *et al.*,^[64] identified 93 compounds from essential oil of leaves, barks of the stem and root, fresh and dried fruits of *X. aethiopica* from Ghana with the predominant compounds such as β -pinene, trans-m-metha-1 (7), 8-diene, germacrene D. Whereas, Tegang *et al.*,^[36] identified seventy (70) components in the essential oil of *X. aethiopica* fruit from Cameroon which mainly consists of β -pinene, β -phellandrene, bisabolene, and α -pinene while Adefegha *et al.*,^[44] detected 30 phyto-compounds in the essential oil of *X. aethiopica* from Nigeria, having eugenol, terpinen-4-ol, germacrene D, (-) spathulenol, and limonene as its major constituents. Different parts of the plant and geographic origins may be responsible for variation in the chemicals and aromatic components of essential oil of *X. aethiopica* obtained from different regions.

Data from Table 3 indicated that eight classes of compounds have been detected from *X. aethiopica* essential oil like monoterpenes (MT), oxygenated monoterpenes (OM), sesquiterpenes (ST), oxygenated sesquiterpenes (OS), nonterpenoid hydrocarbons (NH), diterpene (DT), oxygenated diterpenes (OD), and oxygenated sesterterpene. The essential oil obtained from seeds of *X. aethiopica* has been found to be rich in OM which amounted to (37.6%), followed by OS (29.61%), ST (14.67%), OD (9.05%), NH (4.81%), DT (3.8%), and MT (0.47%). This is in contrast to previous studies that reported the absence/traces of diterpenes and aliphatic esters in the essential oil of *X. aethiopica*.^[36,44,64]

CONCLUSION

Essential oil isolated for the first time from the seeds of *X. aethiopica* and its chemical composition demonstrates that *X. aethiopica* is a source of valuable volatile components. Based on the anticholinesterase and antioxidant results of essential oil, it can be concluded that *X. aethiopica* seeds may be an effective source of compounds that may lead to possible palliative therapy and cure of neurodegenerative and oxidative stress-related diseases.

Table 3: Major class of compounds in essential oil of *Xylopi aethiopica*

Class of compounds	Concentration (%)
OM	37.60
MT	0.47
ST	14.67
OS	29.61
DT	3.80
OD	9.05
NH	4.81
OST	NS
Total	100

MT: Monoterpenes; ST: Sesquiterpenes; OM: Oxygenated MT; NH: Nonterpenoid hydrocarbon; OS: Oxygenated ST; OD: Oxygenated diterpenes; DT: Diterpene; OST: Oxygenated sesterterpene

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Conflicts of interest

There are no conflicts of interest.

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