

Chemical And Medicinal Properties of *XylopiA AethiopiCA* Harvested from The South Eastern Nigeria

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ABSTRACT

The characterization of chemical and medicinal properties of the leaf of *XylopiA aethiopiCA* was carried out with the aid of Shimazu Japan Gas Chromatography 5890-11 and GC-MS QP 2010 Plus Shimazu Japan. The chromatogram revealed 34 peaks which yielded 34 compounds with their molecular weights, structures and formulars elucidated. Initial phytochemical examination revealed the presence of alkaloid 4.3%, flavonoids 5.1%, saponins 4.89%, tannin 2.2%, cardiac glycosides and steriods. Antimicrobial determination revealed activity against selected human pathogens: *Pseudomonas aeruginosa* 10mm, *Staphylococcus aureus* 20mm, *Streptococcus specie* 22mm, *Aspergillus niger* 16mm, *Mucor specie* 18mm and but was resistant to *Klebsiella pneumonia*. Most of the compounds identified in the leaf extract such as phytol, Vitamin E, Squalene, Caryophyllene oxide and Isoaromadendrene epoxide has proven anti-cancer, anti-tumors, anti-inflammatory antioxidant and anti-arthritis properties.

Keywords

XylopiA aethiopiCA, Anticancer, Antitumour, Anti-inflammation, Antioxidant. Pathogens.

Introduction

XylopiA aethiopiCA (Annonaceae), commonly called black pepper, African pepper, Guinea pepper and spice tree, is an ever green aromatic tree growing up to 15-30 m high. It is a native to the low land rain forests and moist fringe forests in the savanna zones and coastal regions of Africa. *XylopiA aethiopiCA* is a vastly used medicinal plant and has a lot of chemical compounds [1]. Some researchers have examined the effects of *XylopiA aethiopiCA* aqueous extract on the antioxidants of matured tomato fruits at red stage. They observed that there was statistically significant differences in ascorbic acid content of stored tomato fruits [2]. Similarly hot water extracts of the leaf and seed of (*XylopiA aethiopiCA*) and Ginger (*Zinigiber officinale*), has been reported to be fungi toxic as the extracts suppressed the growth of *Fusarium oxysporum*, *Aspergillus niger* and *Aspergillus flavus* and reduced

rot development in yam tubers [3]. The plant has been claimed to be useful as abortifacients, ecobolics as well as in the treatment of diarrhea, dysentery; stomach disorder, menstrual disorder, naos-pharyngeal infections, arthritis, rheumatism, infections [1]. There has been reports that the fruit extract could be a potential therapeutic agent against cancer since it inhibits cell proliferation, and induces apoptosis [4]. Other researchers had worked on the hepatoprotective effects of aqueous extract of stem bark on carbon tetrachloride (CCl₄) induced liver damage in Sprague dawley rats. They suggested that aqueous extract of *XylopiA. aethiopiCA* could relieve the liver injuries by its antioxidative effect, hence eliminating the deleterious effect of toxic metabolites from the CCl₄ [5]. In most parts of Africa, it is used in the treatment of cough, rheumatism, dysentery, malaria, uterine fibroid, boils, and wounds among others. The methanolic extract of *XylopiA aethiopiCA* possessed hepatoreparative property especially in acetaminophen-induced hepatotoxicity, however, its toxicity needs to be examined [6,7]. It has been reported that there was reduction in induced intraocular pressure upon administration of *XylopiA aethiopiCA* extract on rats [8].

The seed extract has been shown to have good hypoglycemic effects by lowering the plasma sugar level and also had some beneficial and reduction effects on cardiovascular risk factors [9]. Similarly, the seed extract has been reported to be useful as a pesticide. The extract has been integrated in the weevil management scheme as the extract was strongly repellent to *S. zeamais* [10]. Aqueous and ethanolic extracts of *xylopia aethiopica* has been reported to have marked activity against *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus fecalis*, *Pseudomonas aeruginosa* and *Lactobacilli*, suggesting that the anti-microbial activity of the extract concentrates in the aqueous fraction and also indicate that very low concentration is required to achieve antimicrobial effects [11]. There has been other reports that the oil extracted from the seed of *xylopia aethiopica* affects cell membrane stability and prostaglandin synthetase activity. Similarly, the methanol extract of *Xylopia. aethiopica* stabilized the red blood cells against the haemolytic action of distilled water. The plant extract enhances the healing of wounds, inflammatory disorders and treatment of post-natal pains [12]. The plants extracts has been shown to exhibits significant increase in ameliorating diabetes complication and blood glucose control compared to glibenclamide, a synthetic antidiabetic drug [13]. Phytochemical screening of the fruit of *Xylopia aethiopica* has been reported as the fruit is said to contain Saponin, Saponin glycoside, Tannin and Cardiac glycoside. The aqueous and petroleum ether extracts has been reported to have shown growth inhibitory effects on *Staphylococcus aureus* and *Escherichia coli* but *Pseudomonas aeruginosa* and *Saccharomyces cerevisiae* were resistant to the fruit extract [14]. Similar reports has been made about the seed as it is said to contain alkaloids, saponins, tannins, reducing sugar, anthraquinones, steroids, flavonoids, and glycosides [15]. The antioxidant and antimicrobial activities of Nigerian *X. aethiopica*, has been reported though the extract was reported not to have caused toxic effects to the animals' liver and kidney [16]. The fruit extract is been prospected to have a potential therapeutic activity against cancer since it inhibits cell proliferation, and induces apoptosis [4]. *Xylopia aesthiopica parts* are used in folk medicine for arresting bleeding among women that put to bed as well as treating cardiovascular and diabetic diseases. Also, this plant may be a good factor in the maintenance of electrolyte balance [17]. The antifungal properties of water, ethanol and chloroform extracts of the fruits of *Xylopia aethiopica* has been reported as the extracts showed activities against *Candida albicans* and *Aspergillus niger* [18]. The extract of the seed showed significant increase in serum testosterone and luteinizing hormone levels which clearly reveal androgenic activity of the extract and its effects on hypothalamic pituitary gonadal axis [19]. The dried fruits showed various degrees of activity against gram positive bacteria *Staphylococcus aureus* and gram negative bacterial *Escherichia coli* isolated from gastroenteritic patient [20]. The plant extract has anti proliferative activity against a panel of cancer cell lines and the active natural product been ent-15-oxokaur-16-en-19-oic acid [21].

Sample collection and preparation

Fresh leaves of *Xylopia aethiopica* plant was harvested from federal university of Technology, Owerri, Imo state in July, 2019.

The plant was identify by Mr Iwueze Francis of forest and wild life department, federal university of Technology, owerri. The leaves were selected and dried under room temperature for 60 days and were grinded into fine particles to increase its surface area for better result. The sample was separated into different parts for different analysis.

Test for Flavonoids

5g of the sample was soaked with 50cm³ of water for 2hours and then filtered and to the filtrate was added drops of ammonia and 3cm³ of concentrated H₂SO₄ was added. A yellow precipitate which disappears on storage indicates the presence of flavonoids [22].

Test for Alkaloids

5g of the sample was extracted using 20% acetic acid in ethanol .5cm³ of the extract was treated with Wagner's reagent (iodine crystals and KI). A yellowish brown precipitate indicates the presence of alkaloids [22].

Test for Tannins

5g of the leaf sample was weighed into a beaker and 50cm³ of water was added and allowed to soak properly for two hours and extracted. The extract was treated with drops of ferric chloride. A blue-black precipitate indicates the presence of tannins [22].

Test for Steroids

5cm³ of the water extract was treated with concentrated H₂SO₄ in acetic anhydride. The formation of a blue-green color indicates the presence of steroids [22].

Test for Glycosides

20cm³ of the water extract was treated with Fehling solutions of A and B in equal amount and boiled. A brownish red precipitate indicates the presence of glycoside [22].

Alkaloid determination

5 g of the sample was weighed into a 250 cm³ beaker and 200 cm³ of 20 % acetic acid in ethanol was added and covered to stand for 6hrs. This was filtered and the extract was concentrated using a water bath to one quarter of the original volume. The alkaloid was precipitated out using concentrated ammonium hydroxide which was added drop by drop until precipitation was complete. The solution was allowed to settle and the precipitation was collected by filtration using whatman filter paper, the precipitate was dried and weighed [23,24].

Saponins Determination

20g of the sample was weighed into a 250cm³ beaker and 200cm³ of 20 % ethanol was added and stirred using glass rod. The mixture was heated over water bath for 4hrs with continuous stirring while the temperature was maintained at 55°C. The mixture was extracted and the residue was extracted with 200 cm³ of 20% ethanol. The combined extract was reduced to 40cm³ over water bath at 90°C. The concentrated extract was transferred into a 250cm³ separation funnel and 20cm³ of diethyl ether was added

and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. This process was repeated thrice. 60 cm³ of n-butanol was added. The mixture was washed twice with a 10 cm³ of 5 % sodium chloride. The remaining solution was heated over water bath and the residue dried to constant weight. The saponin content was calculated in percentages [23,24].

Flavonoid Determination

10g of the defatted sample were extracted repeatedly with 100cm³ of 80% of aqueous methanol at room temperature. The solution obtained was filtered with whatman filter paper no 45. The filtrates were later transferred into a crucible and evaporated to dryness over a water bath and weighed [24,25].

Quantification of Tannins

20g of the sample measured into a beaker and 150cm³ of water was added. The sample stirred & was allowed to stay for 4hrs before filtration; using a whatman filter paper. Few drops of conc. HCl was added to the clear solution to the acidified it, then followed by the addition of ethyl acetate. This solution was properly mix and separated using a separating funnel. This was repeated thrice the aqueous solution obtained while the ethyl acetates layer was discarded, was heated to dryness and tannins was obtained and weighed [22].

Preparation of Samples for GC-MS Analysis

Two hundred grams of the sample was repeatedly extracted with ethanol using soxhlet extractor, another 200g of sample was soaked in ethanol for 48 hour and extracted. The extracts from the soxhlet extracts and that obtained from cold extracts for the sample were combined and re-extracted using chloroform to obtain chloroform soluble extract. This was centrifuged at 10,000rpm for 20 minutes and the clear supernatant oil was subjected to GC and GC-MS analysis.

GC-MS Experimental Procedures

GC- analysis was carried out with SHIMAZU Japan Gas Chromatography 5890-11 with a fused GC column OV 101 coated with polymethyl silicon (0.25 mm x 50 m) and the conditions are as follows: Temperature programming from 80 – 200°C held at 80°C for 1 minute, the rate is 5°C/min and at 200°C for 20 minutes. FID Temperature of 300°C, injection temperature of 250°C, carrier gas is Nitrogen at a flow rate of 1 cm³/min and split ratio of 1: 75. GC-MS Gas chromatography Mass spectrum analysis were conducted using GC-MS QP 2010 Plus Shimazu Japan with injector Temperature at 230°C and carrier gas pressure of 100kpa. The column length was 30 m with a diameter of 0.25 mm and the flow rate of 50 ml/min. The eluents were automatically passed into the Mass Spectrometer with a detector voltage set at 1.5 KV and sampling rate of 0.2seconds. The Mass Spectrometer was also equipped with a computer fed Mass Spectra data bank, HERMCE Z 233 M-Z centrifuge Germany was used. Reagents and solvents such as Ethanol, Chloroform, Diethyl ether, hexane all of analytics grade was obtained from Merck Germany [22].

Antimicrobial Analysis

Collection of isolates of Bacteria and fungi isolates and identification

Clinical bacterial isolates were obtained from federal medical Centre Owerri Microbiology laboratory. The bacteria includes *Escherichia coli*, *Klebsiella pneumonia*, *Staphylococcus aureus* and *Streptococcus pneumonia*. Each of this bacteria were sub-cultured on a sterile plate of nutrient agar, Mac-Conkey agar, blood agar and chocolate agar according to the type of bacteria. The identification of the bacteria was confirmed using the using the following gram staining and biochemical test such as methyl red test, motility test and indole test.

Mycological Examination of Fungal Isolates

Two fungal isolates were sub-cultured on sterile plates of sabouraud dextrose agar. The two fungi includes *Aspergillus Niger* and *Mucor specie*. The identity of the fungus was determined using the needle mouth technique. With the help of a sterile inoculating needle, small portion of each fungal structure was collected and dropped at the center of a sterile grease free glass slide. With the help of Pasteur's pipette, a drop of ethanol was made on the fungal structure .the ethanol was allowed to evaporate. A drop of lacto phenol cotton blue stain was made on the fungal structure using Pasteur's pipette. Two inoculating needles were used to tease the fungal structure. The preparation was carefully covered with cover slip. It was examined using low and high power magnification, the fungal isolates were identified using types of conidia, chlamydosperm and hyphae.

Evaluation of Antimicrobial Activity

Preparation of Bacterial and Fungal Suspensions

2mls of normal saline was aseptically poured into sterile 5ml test tube. The test tube. The test tubes were labeled with the names of each isolate; bacteria and fungi. Two gram positive bacteria (*Streptococcus and Staphylococcus*), two gram negative bacteria (*Klebsiella and Escherichia coli*), two fungi specie *Aspergillus Niger* and *Mucors specie*. These six test tubes were set up on test tube rack. With the help of a sterile wire loop, each fungal and bacteria were transferred to each tube bearing each isolates name according to the previous labeling of the test tubes. The test tubes were swirled after each inoculation until the isolate suspension becomes turbid. The color of each tube was marched with that of a 5% Marc Farlard standard. Bacterial suspension had four test tubes while the fungal suspensions had two test tubes.

Dilution of the plant extract

Four sterile test tubes were set up on a test tube rack and labelled accordingly. 1 cm³ of the plant extract was added to the first test tube that is the stock. 1 cm³ of distilled water was added into each test tube from test tube 2-4. 1cm³ of plant extract. 1ml of plant extract was added into test tube 2 [1,20]. The 1cm³ distilled water and 1 ml extract were carefully shaken to obtain homogenous mixture. 1cm³ of this mixture was transferred to the third test tube and well shaken. With a new pipette 1 cm³ of the content of test tube 3 was transferred to test tube 4 and carefully shaken. 1ml of its content was discarded using another sterile pipette. With

this method four different concentrations of plant extracts were obtained in each test tube.

Antibacterial activity using well-in- agar method

4 plates of sterile Muller Hinton agar plates were prepared according to the manufacturer's instruction. The plates were allowed to cool and solidify. With the help of a pipette 0.1cm³. *Klebsiella specie* suspension was introduced on the surface of the plate of prepared Muller Hinton agar. In like manner, equal volume of other bacterial suspensions were made on the remaining agar plates. Using a sterile 6mm cork borer, four wells were made on each agar plate. The wells were labelled according to the dilution of plant extract made (stock, 1/10, 1/20, 1/40). With different pipette, each strength of the plant extract was filled in each well as labelled until each well is filled to the brim. This was left for 1 hour to ensure that the extract has been absorbed by the agar. The plates were incubated at 37°C for 24 hours. Zones of inhibition of the different the plant extract on each bacteria were observed and recorded. The process was repeated with each of the bacteria isolates.

Antifungal Activity Using Well In Agar Method

Two plates of sabouraud dextrose agar were freshly prepared. Two plates of the agar were smeared with *Aspergillus Niger* while the other two plate were also smeared with *Mucor specie*, four well agar were also bored on each plate. Each well containing different dilution of each extract just as was done to the bacteria isolates earlier. The plates were incubated at 37°C temperature for 48 hours. Plates were observed and zones of inhibition of different plant extracts on the two fungi plates were recorded.

Results And Discussion

Table 1: Result of phytochemical screening of the *X.aethiopica*.

| Phytochemical | Inference |
|-------------------|-----------|
| Alkaloid | ++ |
| Saponin | ++ |
| Flavonoid | ++ |
| Steroid | ++ |
| Tannin | ++ |
| Cardiac glycoside | ++ |
| Key ++ present | |

Table 2: Result of determination of Phytochemicals.

| Phytochemical | % yield |
|---------------|---------|
| Alkaloid | 4.8 |
| Flavonoid | 5.1 |
| Saponin | 4.89 |
| Tannins | 2.2 |

The results reveal the presence of alkaloids 0.92%, saponins 0.12%, flavonoids 0.15% and cardiac glycosides, while tannins and steroids were not detected.

Alkaloids have reported to be present in most green leafy plants and they have many therapeutic properties. Most alkaloids have found their way in medicinal application as anti-malarial, antimicrobial, antibacterial, antifungal and anti-parasitic activities [26]. Reported that alkaloids in plant extract has a lot of medical

potential in curing and preventing ailments like malaria, typhoid, and jaundice. Their result could only be justified because of the presence of alkaloids most samples containing alkaloid are used in Nigeria for the treatment of malaria and fever [24] some alkaloids are may be considered toxic but others are used medicine. Many alkaloids can be used for medical purposes [27,28]. Some act on the central nervous system while many others are anti-malarial drugs

Flavonoids are distributed group of polycyclic compounds characterized by a common Benzo pyrone ring structure that has been reported to act as antioxidants in many biological systems. The family encompasses flavonoids, flavones, chalcones, catchins, anthocyanidins and isoflavonoids [29]. In addition to their free radical scavenging activities, Flavonoids have multiple biological activities including – vasodilator, anti-carcinogenic, anti-allergic, antiviral, estrogenic effects as well as being inhibitors of phospholipase H2, cyclooxygenase, glutathione reductase and xanthine oxidase. Flavonoids prevent oxidative cell damage, have strong anti-cancer activity and protects against all stages of cancer development [30]. Flavonoids are important antioxidants have many health benefits as they have antiviral, anticancer, anti-inflammatory and anti-allergic properties [27,31], they support lactogenesis. These properties therefore support the use of plant extract in cancer therapy [32]. Flavonoids in intestinal tracks lower the risk of heart diseases. As anti-oxidant, flavonoids provide anti-inflammatory actions.

Saponins was found to be available in the leaf of the plant Some of the general characteristic of saponins includes; formation of forms in aqueous solutions, hemolytic activity and cholesterol binding properties [33,28]. Saponin has the natural tendency to ward off microbes and this makes them good candidates for treating fungal and yeast infections they also serve as hormones and contraceptives. These compounds serve as natural antibiotic, helping the body to fight infections and microbial invasion [24]. Saponins have anti-inflammatory and anti-cancer properties and can lower the risk of human cancers by preventing cancer cells from growing. [15] Cardiac glycosides are useful in treating heart related diseases and also for the treatment of high blood pressure [14].

Table 3: Results of antimicrobial analysis

| Conc | <i>Streptococcus specie</i> | <i>Staphylococcus aureus</i> | <i>Klebsiella pneumonia</i> | <i>Pseudomonas aeruginosa</i> | <i>Aspergillus niger</i> | <i>Mucor specie</i> |
|-------|-----------------------------|------------------------------|-----------------------------|-------------------------------|--------------------------|---------------------|
| Stock | 22 | 20 | R | 10 | 16 | 18 |
| 1/10 | 15 | 14 | R | 7 | 13 | 10 |
| 1/20 | 10 | 7 | R | 5 | 10 | 6 |
| 1/40 | 10 | R | R | 5 | 10 | R |

The plant extract showed marked inhibitory potentials against certain selected human pathogens, the main stock inhibited the growth of *Staphylococcus aureus* by a diameter of 20mm, similarly, other micro-organisms were not left out with inhibitions recorded against *Pseudomonas aeruginosa* 10 mm, *Aspergillus niger* 16mm and *Mucor specie* 18 mm and *Streptococcus specie* 22 mm. Most of these pathogens have been implicated to be the main causes of some human ailments. *Staphylococcus aureus* is a gram positive coccus that causes skin infection such as; pimples, impetigo,

boils, cellulitis, folliculitis, carbuncles, scalded skin syndrome, abscesses, pneumonia, toxic shock syndrome, bacteremia and sepsis [22]. It has been reported that extracts from this plant has activity against gram positive bacteria *Staphylococcus aureus* and gram negative bacterial [20].

Pseudomonas aeruginosa is a gram negative gamma-proteobacteria which belong to the family *Pseudomonaceae*. It causes bacteremia, pneumonia, folliculitis, swimmer ear which is an ear infection accompanied with swelling, ear pus. Itching, discharge and difficulty in hearing, eye inflammation with associated pains, pus, swelling redness and impaired vision. Klebsiella, a non-motile gram negative, oxidase rod shaped bacteria which causes infectious wounds, pneumonia, blood stream infections and urinary tract infections [22,34].

GC-MS Results

Gas Chromatography Mass Spectrophotometer result

The result of the GC-MS analysis of *Xylopia Aethiopica* is shown in figure 1 above. 36 peaks were obtained representing 36 compounds table 4. Peak 1 occurred at M/Z 128, with molecular formula, $C_{10}H_8$ and identified as Naphthalene, Peak 2 occurred at M/Z 194, with molecular formula $C_{12}H_{18}O_2$ and is identified as Bicyclo [3.1.1] hept-3-ene-2,4-(1',3'-dioxane-7,7-dimethyl. Their structures are contained in fig 2 below. Similarly. Peak 3 occurred at M/Z 138 with molecular formula, $C_8H_{10}O_2$, named Ethanol -2-

phenoxy. Peak 4, M/Z 146, molecular formula $C_{11}H_{14}$, name, benzene-2-ethenyl-1,3,5-trimethyl- Peak 5, m/z 142, formula $C_{11}H_{10}$ name Naphthalene-2-methyl. Peak 6 m/z 142, formula $C_{10}H_{11}$, name, Benzocycloheptatriene. Peak 7, m/z 190, formula $C_{12}H_{18}O$, name ,Oxacyclotetradeca-4,11-diene, peak 8 m/z 204, formula $C_{15}H_{24}$, name, Cyclohexane,1-ethenyl-1-1-methyl-2,4-bis(1- . Peak 9 m/z 204, formula $C_{15}H_{24}$ name Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl-2-(1-methylethenyl)-. Peak 10 m/z, 224, Formula $C_{16}H_{32}$, name, 1-Hexadecene . Peak 11, M/Z 220, Formula $C_{15}H_{24}O$, name, trans-Z- α -Bisabolene epoxide. Peak 12, M/Z, 220, Formula $C_{15}H_{24}O$, name, Caryophyllene oxide. Peak 13, M/Z 220, Formula $C_{15}H_{24}O$, name, Isoaromadendrene epoxide. Peak 14, m/z 266. Formula $C_{16}H_{26}O_3$ Name 2-Dodecen-1-y (-) succinic anhydride. Peak 15, m/z 228, Formula $C_{15}H_{36}O$, name,1-Dodecanol,1,3,7,11-trimethyl- .Peak 16, M/Z 296, Formula $C_{20}H_{40}O$, name, 3,7,11,15-Tetramethyl-2-hexadecen-2-ol. Peak 17, M/Z 256, Formula $C_{16}H_{32}O$, name, n-Hexadecanoic acid. Peak 18 M/Z, 272, Formula $C_{20}H_{32}$ name, Kaur-15-ene .Peak 19 M/Z, 272, Formula $C_{20}H_{32}$, name, Kaur-16-ene. Peak 20, M/Z, 296, Formula $C_{20}H_{40}O$, name Phytol. Peak 21 M/Z, 282, Formula $C_{18}H_{34}O$, Trans-13-Octadecanoic acid. Peak 22 M/Z 290, Formula $C_{20}H_{34}O$, 16 β H-Kauran-16-ol. Peak 23, M/Z 302, Formula $C_{20}H_{30}O_2$, name, Icosapent. Peak 24, M/Z 316, Formula $C_{20}H_{32}O_2$, name, Kaur-16-en-18-oic acid methyl ester, (4 β) - . Peak 25, M/Z 302, Formula $C_{20}H_{30}O_2$, name ,Abietic acid .Peak

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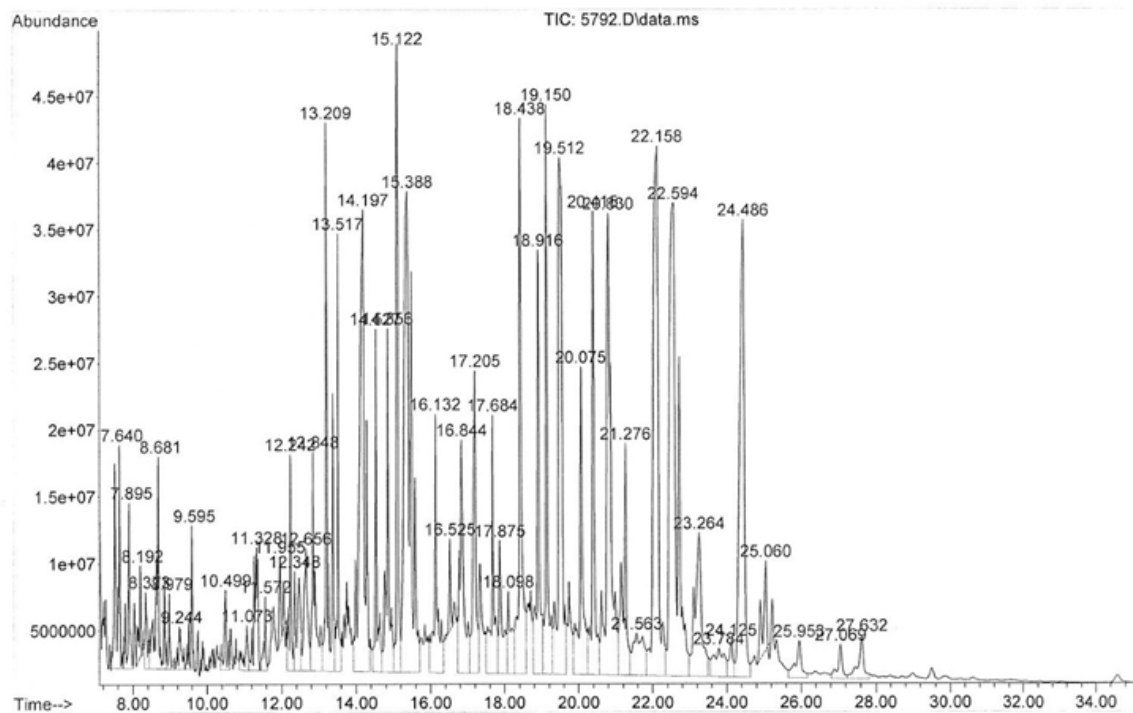


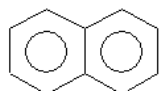
Figure 1: The result after GC-MS analysis was carried out are shown having 36 peaks and compound obtained are listed below in table 4.

26, M/Z 390, Formular $C_{24}H_{38}O_4$, name, 1,2-Benzenedicarboxylic acid, diisooctyl ester .Peak 27, M/Z, 364, Formular $C_{26}H_{52}$, name, 1-Hexacosene. Peak 28, M/Z 396, Formular $C_{27}H_{56}$, name, 1-Heptacosanol. Peak 29, M/Z 410, Formular $C_{20}H_{50}$, name, Squalene .Peak 30, M/Z, 394. Formular, $C_{28}H_{58}$, name, Octacosane. Peak 31, M/Z 396, Formular $C_{27}H_{56}O$, name 1-Heptacosanol peak 32, M/Z 286, formular $C_{36}H_{74}$, name, Octadecanal .Peak 33, M/Z 506, Formular $C_{36}H_{74}$, name, Hexatriacontane .Peak 34, M/Z 430, Formular $C_{29}H_{50}O_2$. Name. Vitamin E. Peak 35. M/Z 454. Formular $C_{30}H_{62}O_2$, name, 1, 30-Triacontanediol. Peak 36, M/Z 450, Formular $C_{31}H_{62}O$, name, 18-Hentriacontanone.

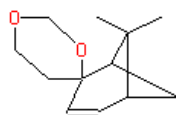
Table 3: Gas Chromatography Mass Spectrophotometer result from

| s/n | Name | MWT | Formula |
|-----|-----------------------------------------------------------------------------|-----|-------------------|
| 1 | Naphthalene | 128 | $C_{10}H_8$ |
| 2 | Bicyclo [3.1.1] hept-3-ene-spiro-2,4'-(1',3'-dioxane), 7,7-dimethyl- | 194 | $C_{12}H_{18}O_2$ |
| 3 | Ethanol-2-phenoxy- | 138 | $C_8H_{10}O_2$ |
| 4 | Benzene, 2-ethenyl-1,3,5-trimethyl- | 146 | $C_{11}H_{14}$ |
| 5 | Naphthalene 2-methyl- | 142 | $C_{11}H_{10}$ |
| 6 | Benzocycloheptatriene | 142 | $C_{11}H_{10}$ |
| 7 | Oxacyclotetradeca-4,11-diyne | 190 | $C_{12}H_{18}O$ |
| 8 | Cyclohexane, 1-ethenyl-1-1-methyl-2,4-bis(1-methylethenyl)- | 204 | $C_{15}H_{24}$ |
| 9 | Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl-2-(1-methylethenyl)- | 204 | $C_{15}H_{24}$ |
| 10 | 1-Hexadecene | 224 | $C_{16}H_{32}$ |
| 11 | trans-Z- α -Bisabolene epoxide | 220 | $C_{15}H_{24}O$ |
| 12 | Caryophyllene oxide | 220 | $C_{15}H_{24}O$ |
| 13 | Isoaromadendrene epoxide | 220 | $C_{15}H_{24}O$ |
| 14 | 2-Dodecen-1-y(-)succinic anhydride | 266 | $C_{16}H_{26}O_3$ |
| 15 | 1-Dodecanol,1,3,7,11-trimethyl- | 228 | $C_{15}H_{32}O$ |
| 16 | 3,7,11,15-Tetramethyl-2-hexadecen-2-ol | 296 | $C_{20}H_{40}O$ |
| 17 | n-Hexadecanoic acid | 256 | $C_{16}H_{32}O_2$ |
| 18 | Kaur-15-ene | 272 | $C_{20}H_{32}$ |
| 19 | Kaur-16-ene | 272 | $C_{20}H_{32}$ |
| 20 | Phytol | 296 | $C_{20}H_{40}O$ |
| 21 | Trans-13-Octadecanoic acid | 282 | $C_{18}H_{34}O_2$ |
| 22 | 16 β H-Kauran-16-ol | 290 | $C_{20}H_{34}O$ |
| 23 | Icosapent | 302 | $C_{20}H_{30}O_2$ |
| 24 | Kaur-16-en-18-oic acid, methyl ester,(4 β)- | 316 | $C_{20}H_{32}O_2$ |
| 25 | Abietic acid | 302 | $C_{20}H_{30}O_2$ |
| 26 | 1,2-Benzenedicarboxylic acid, diisooctyl ester | 390 | $C_{24}H_{38}O_4$ |
| 27 | 1-Hexacosene | 364 | $C_{26}H_{52}$ |
| 28 | 1-Heptacosanol | 396 | $C_{27}H_{56}O$ |
| 29 | Squalene | 410 | $C_{30}H_{50}$ |
| 30 | Octacosane | 394 | $C_{28}H_{58}$ |
| 31 | 1-Heptacosanol | 396 | $C_{27}H_{56}O$ |
| 32 | Octadecanal | 268 | $C_{18}H_{36}O$ |
| 33 | Hexatriacontane | 506 | $C_{36}H_{74}$ |
| 34 | Vitamin E | 430 | $C_{29}H_{50}O_2$ |
| 35 | 1,30-Triacontanediol | 454 | $C_{30}H_{62}O_2$ |
| 36 | 18-Hentriacontanone | 450 | $C_{31}H_{62}O$ |

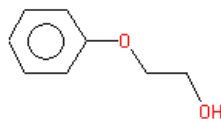
Figure 2: Structures of listed compounds.



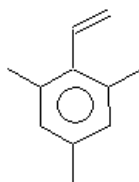
(i) Naphthalene



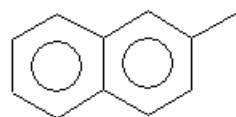
(ii) Bicyclo[3.1.1]hept-3-ene-spiro-2,4'-(1',3'-dioxane), 7,7-dimethyl



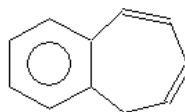
(iii) Ethanol, 2-phenoxy



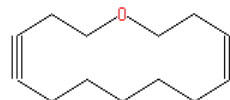
(iv) Benzene, 2-ethenyl-1,3,5-trimethyl-



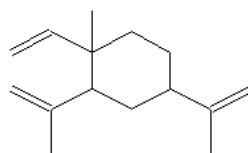
(v) Naphthalene 2-methyl



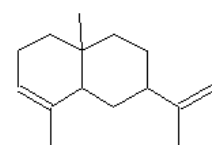
(vi) Benzocycloheptatriene



(vii) Oxacyclotetradeca-4,11-diyne



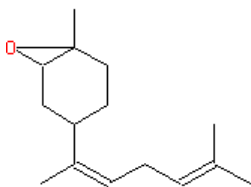
(viii) Cyclohexane, 1-ethenyl-1-1-methyl-2,4-bis(1-methylethenyl)-



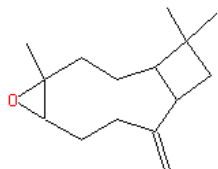
(ix) Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl-2-(1-methylethenyl)-



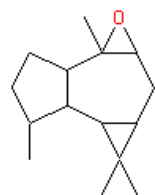
(x) 1-Hexadecene



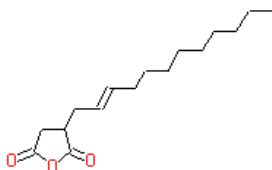
(xi) trans-Z- α -Bisabolene epoxide



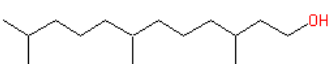
(xii) Caryophyllene oxide



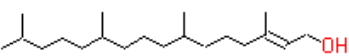
(xiii) Isoaromadendrene epoxide



(xiv) 2-Dodecen-1-y(-)succinic anhydride



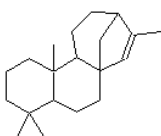
(xv) 1-Dodecanol,1,3,7,11-trimethyl-



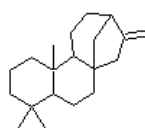
(xvi) 3,7,11,15-Tetramethyl-2-hexadecen-2-ol



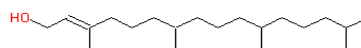
(xvii) n-Hexadecanoic acid



(xviii) Kaur-15-ene



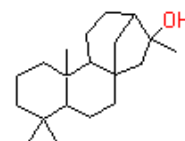
(xix) Kaur-16-ene



(xx) Phytol



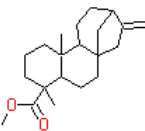
(xxi) trans-13-Octadecanoic acid



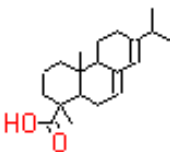
(xxii) 16 β H-Kauran-16-ol



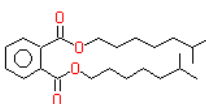
(xxiii) Icosapent



(xxiv) Kaur-16-en-18-oic acid, methyl ester,(4 β)-



(xxv) Abietic acid



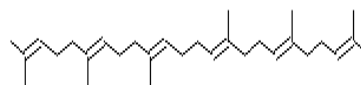
(xxvi) 1,2-Benzenedicarboxylic acid, disooctyl ester



(xxvii) 1-Hexacosene



(xxviii) 1-Heptacosanol



(xxix) Squalene



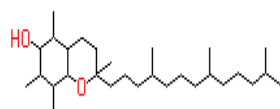
(xxx) Octacosane



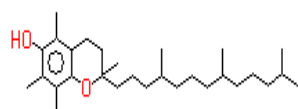
(xxxii) Octadecanal



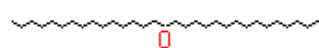
(xxxiii) Hexatriacontane



(xxxiii) Hexatriacontane



(xxxiiii) Vitamin E



(xxxiv) 30-triacontanone



(xxxv) 18-Hentracotanal

Some of the compounds so identified include Squalene is a natural product that reduces skin damage by ultra violet radiation, it also reduces the cholesterol level in the blood and prevents cardiovascular diseases, having antitumor, anticancer against ovarian, breast, lungs and colon cancer [35]. Phytol is a precursor of vitamin K1 and E. It is a schistosomicide drug. It is used in making synthetic vitamin E and K. It has antimicrobial, antioxidant anti-inflammatory cytotoxic effects. Vitamin E was detected in the extract. Vitamin E is a group of eight fat soluble compounds that include four tocopherols and four tocotrienols. Vitamin E functions for strong immunity and healthy skin and eyes. It is an antioxidant is believed to help in healing and reduction of scarring when applied to the skin. It is a lipid soluble component in the cell antioxidant defense system, it can serve to prevent ageing, cancer, arthritis and cataracts, it helps to reduce prostaglandins such as thromboxane which causes clumping of platelets. [36]. Caryophyllene oxide is a terpene found in cloves, basil, hops and pepper as well as cannabis, it has antioxidant effect and strong cholinesterase inhibitory properties and anticancer activities [37]. Aromadendrene is a 5, 10-cycloaromadendrene sesquiterpenoids which possess antimicrobial activity and occurs mostly in essential oils.

Conclusion

The research work has both confirmed certain compounds found in *Xylopi aethiopia* and also revealed certain new compounds yet unidentified in the plant, Evidence of the potent use of the

plant in lactogenicity has been laid to the many phytochemicals especially its saponins which also acts as hormone and hormone modulators, the flavonoids the fights cancer and tumors. Most of the compounds identified in the extract have anti-cancer, anti-tumor, anti-inflammation activities the plant can be used in the treatment of cancer, tumor and inflammations.

References

1. Earnest O E, Goodies E M. *Xylopi aethiopia*: A Review of its Ethnomedicinal, Chemical and Pharmacological Properties. *Am J Pharm Tech Res.* 2014; 4: 22-37.
2. Babarinde G O, Adegoke G O. Effect of *Xylopi aethiopia* aqueous extract on antioxidant properties of refrigerated Roma tomato variety packaged in low density polyethylene bags. *J Food Sci Technol.* 2015; 52: 1790-1795.
3. Okigbo R N, Nmeko I A. Control of yam tuber rot with leaf extracts of *Xylopi aethiopia* and *Zingiber officinale*. *African Journal of Biotechnology.* 2005; 4: 804-807.
4. Oluwatosin AA, Jayanta S, Neetu S, et al. Antiproliferative action of *Xylopi aethiopia* fruit extract on human cervical cancer cells. *Phytother Res.* 2011; 25: 1558-1563.
5. Adewale OB, Adekeye AO, Akintayo CO, et al. Carbon tetrachloride (CCl₄)-induced hepatic damage in experimental Sprague Dawley rats: Antioxidant potential of *Xylopi aethiopia*. *Journal of Phytopharmacology.* 2014; 3: 118-123.
6. Folorunso OS, Ogungbe BF, Ikanone C E O. Methanolic Extract of *Xylopi aethiopia* Ameliorates Acetaminophen-Induced Liver Damage in Male Wistar Rats. *Annual Review & Research in Biology.* 2013; 3: 1074-1090.
7. Fleischer T C, Mensah M L K, Mensah A Y, et al. Antimicrobial Activity of Essential Oils of *Xylopi aethiopia*. *Afr J Tradit Complement Altern Med.* 2008; 5: 391-393.
8. Uzodike E B, Onuoha I N. The Effect of (uda) on Intraocular Pressure. *Journal of the Nigerian Optometric Association.* 2010; 16: 21-25.
9. Ogbonnia S, Adekunle A A, Bosa M K, et al. Evaluation of acute and subacute toxicity of *Alstonia congensis* Engler (Apocynaceae) bark and *Xylopi aethiopia* (Dunal) A. Rich (Annonaceae) fruits mixtures used in the treatment of diabetes. *African Journal of Biotechnology.* 2008; 7: 701-705.
10. Babarinde S A, Adebayo M A, Oduyemi K. Integrating varietal resistance with *Xylopi aethiopia* (Dunal) A. Richard seed extract for the management of *Sitophilus zeamais* Motschulsky in stored maize. *African Journal of Biotechnology.* 2008; 7: 1187-1191.
11. Ijeh I I, Omodamiro O D, Nwanna I J. Antimicrobial effects of aqueous and ethanolic fractions of two spices, *Ocimum gratissimum* and *Xylopi aethiopia*. *African Journal of Biotechnology.* 2005; 4: 953-956.
12. Ezekwesili C N, Nwodo F C, Ogbunugafor A H. Investigation of the Chemical Composition and biological activity of *Xylopi aethiopia* Dunal Annonaceae. *African Journal of Biotechnology.* 2010; 9: 7352-7356.

13. Victor E O, Bayim P B, Margaret O. Comparative Effects of Some Medicinal Plants: *Anacardium occidentale*, *Eucalyptus globulus*, *Psidium guajava*, and *Xylopiya aethiopic*a Extracts in Alloxan-Induced Diabetic Male Wistar Albino Rats. *Biochemistry Research International*. 2014; 2014:203051.
14. John-Dewole O O, Agunbiade S O, Alao O.O, et al. Phytochemical and antimicrobial studies of extract of the fruit of *Xylopiya aethiopic*a for medicinal importance. *Journal of Biotechnology and Pharmaceutical Research*. 2012; 3: 118-122.
15. Aguoru C U, Pilla C, Olasan J O. Phytochemical screening of *Xylopiya aethiopic*a with emphasis on its medicinally active principles. *Journal of Medicinal Plants Research*. 2016; 10: 306-309.
16. Abubakar A Y, Bashir L, Mumeen A Y, et al. Free Radical Scavenging, Antimicrobial Activities and Effect of Sub-Acute Exposure to Nigerian *Xylopiya aethiopic*a Seed Extract on Liver and Kidney Functional Indices of Albino Rat Iranian. *Journal of Toxicology*. 2018; 12: No 3.
17. Nnodim J, Emejulu A, Amaechi A, et al. Influence of *Xylopiya Aethiopic*a Fruits on Some Hematological and Biochemical Profile. *J Med Sci*. 2011; 4: 191-196.
18. Kanife U C, Doherty F, Nwakanma N M C, et al. Antifungal Activity of *Xylopiya aethiopic*a on Some Clinical Organisms in Nigeria. *Hamdard Medicus*. 2012; 55: No.1.
19. Woode E, Alhassan A, Chrissie S, et al. Effect of ethanolic fruit extract of *Xylopiya aethiopic*a on reproductive function of male rats. *Int J Pharm Biomed Res*. 2011; 2: 161-165.
20. Ikeyi A P, Ogbonna A O, Ibekwe R O, et al. antimicrobial activity of *xylopiya aethiopic*a (uda) on *escherichia coli* and *staphylococcus aureus* isolates from gastroenteritic patients. *International journal of life science Biotechnology and Pharma Research*. 2013; 2: 3.
21. Aphrodite T C, Mathieu D, Stefan C, et al. Characterization of the antiproliferative activity *Xylopiya aethiopic*a. *Cell Division*. 2012; 7: 1-8.
22. Iwu I C, Onu U, Ukaoma A A, et al. Characterization of the Volatile Components of the Leaf of *Starchytarpheta cayennensis* (Rich) Vahl. *International Journal of Herbs, Spices and Medicinal Plants*. 2019; 4: 041-049.
23. Obadoni B O, Ochuko P O. Phytochemical studies and comparative efficacy of the crude extract of some homeostatic plants in Edo state and Delta state of Nigeria. *Global journal of Pure and Applied Sciences*. 2001; 8: 203-208.
24. Iwu I C, Onu U L, Chijioke-okere M, et al. GC-MS, Phytochemical and Antibacterial Analysis of *Pentaclethra macrophylla* Leaf. *The International Journal of Science and Technology*. 2016; 4: 151-159.
25. Boham B A, Kocipai A C. Flavonoids and condensed tannins from leaves of *Hawaina Vaccinium* and *V.calycinium*. *Pacific Sci*. 1994; 48: 458-463.
26. Fapojuwomi O A, Asinwa I O. Assessment of Medicinal Values of *Rauwolfia vomitoria* (Afzel) in Ibadan Municipality. *Greener Journal of Medical Sciences*. 2013; 3: 037-041.
27. Fleischer T C. *Xylopiya aethiopic*a A Rich.: A chemical and biological perspective. *J. Univ. Sci. Technol*. 2003; 23: 24-31.
28. Keita B, Sidibe L, Figneredo G, et al. Chemical Composition of the Essential oil of *Xylopiya aethiopic*a (Dunal) A. ch. from Mali. *J. Essent. Oil Res*. 2003; 15: 267-269.
29. Okwu D E, Aluwuo C J. Studies on the phytochemical composition and fermentation of the seed of African oil bean tree *Pentaclethra Macrophylla* Benth. *Int. journal of chemical societies*. 2008; 6: 773-788.
30. Aguoru C U, Ameh S J, Olasan O. Comparative Phytochemical Studies on the presence and quantification of various bioactive compounds in the three major organs of Okoho plant (*Cissus Popunea* Guill & Perr) in Benue State, North Central Nigeria, West Africa. *Eur. J. Adv. Res. Biol. Life Sci*. 2014; 2: 16-22.
31. Okwu D E. Phytochemicals and mineral content of indigenous spices south of eastern Nigeria. *Journal of sustainable Agriculture and Environment*. 2004; 6: 30-37.
32. Asoegwu S N, Ohanyere S O, Kanu O P, et al. Physical properties of African oil Bean Seed (*Pentaclethra macrophylla*). *Agricultural Engineering international*. 2006.
33. Okwu D E. Phytochemicals, Vitamins and Mineral Contents of Two Nigerian Medicinal Plants. *International. Int. J.Mol. Med. Adv. Sci*. 2005; 1: 375-381.
34. Okoronkwo NE, Echeme JO, Onwuchekwa EC. Cholinesterase and bacterial inhibitory activities of *Stachytarpheta cayennensis*. *Academic Research International*. 2012; 2: 209.
35. Azalia LG, Shela G, Eduardo E R, et al. Plant Sources, Extraction Methods, and Uses of squalene. *International Journal of Agronomy*. 2018; 2018: Article ID1829160.
36. Saliha R, Syed T R, Farzana M, et al. The Role of Vitamin E in Human Health and some Diseases. *Sultan Qaboos Univ Med J*. 2014; 14: 157-165.
37. Songul K, Serder V Y, Ozlem O, et al. C.B.K2020 Acaryophyllene oxide and other potential anticholinesterase and anticancer agent in *Salvia Verticillata* .subsp.amasiaca Freyn and Bornm) Bornm. (Lamiaceae). *Journal of Essential oil Research* 2020; 32: 512-525.