DOI: 10.38061/idunas.1500241

| IDUNAS | NATURAL & APPLIED SCIENCES JOURNAL | 2024 Vol. 7 No. 2 (8-28) |
| --- | --- | --- |

# Ethnobotany and Bioactive Constituents of *Tabernaemontana pachysiphon* Stapf (Apocynaceae) Used in Indigenous Medicine Practices in Abia State, Nigeria

Research Article

**David N. Enyiukwu[1*], Anderson C. Amadioha[1], Inemesit N. Bassey[2]**

[1]Department of Plant Health Management, Michael Okpara University of Agriculture, Umudike, KM 10 Umuahia-Ikot Ekpene Road, PMB 7267 Umuahia, Abia State, Nigeria. Tel.: +234(0)09092309790

[2]Department of Botany and Ecological Studies, University of Uyo, PMB 1017 Uyo, Akwa-Ibom State, Nigeria

Author E-mails:
enyidave2003@gmail.com
amadioha4u@yahoo.com
inemesitnbassey@uniuyo.edu.ng
David N. Enyiukwu ORDID ID: 0000-0003-0595-0630
Anderson C. Amadioha ORDID ID: 0009-0003-5050-8697
Inemesit N. Bassey ORDID ID: 0009-0005-2677-2062

*Correspondence to: David N. Enyiukwu, Department of Plant Health Management, Michael Okpara University of Agriculture, Umudike , KM 10 Umuahia-Ikot Ekpene Road, PMB 7267 Umuahia, Abia State,

DOI: 10.38061/idunas.1500241

**Received: 12.06.2024; Accepted: 06.11.2024**

## Abstract

This work surveyed the ethno-botanical relevance of *Tabernaemontana pachysiphon* in the indigenous medicinal practices of the people of Ekebedi ancient Kingdom, Abia State, Nigeria. In rural tropical localities of Africa such as the Kingdom, modern western Medicare is scarcely available, and plants have provided substantial part of the medicines used by the natives over the years to treat both metabolic and pathological diseases as well as fractures amongst many other health conditions. Questionnaires and oral interviews were utilized to capture relevant data about the plant from 102 indigenous herbal doctors, traditional medicine vendors and natives of the Kingdom and its environs. Phytochemical analysis of the test extract was conducted using GC-MS fingerprinting. The results obtained from the survey indicated that preparations from seeds, leaves, stems and root barks of the plant are used by traditional medicine men and natives of the Kingdom to treat fevers, pains, diabetes, hypertension, stomach troubles, ringworms, malaria, gonorrhea, syphilis, venereal diseases, tumors and bouts of poisoning etc.

GC-MS profiling identified 32 bioactive principles in the methanol leaf extract of the plant, with 9, 12-octadecadienoic acid (Z, Z)-, methyl ester (16.35%); octadecenoic acid, methyl ester, (E)-, (19.17%); dodecanoic acid 1, 2, 3-propanetrieyl ester (29.69%) being the major ingredients. Amongst many others were hexadecenoic acid, methyl ester (2.91%); cyclopentadecanone, oxime (1.01%); Z,Z-8,10-hexadecadien-1-ol

(1.82); 9-octadecenoic acid, 1,2,3-propanetriyl ester, E (5.86%); 9-octadecenoic acid, 1,2,3-propanetriyl ester, (E- (1.72%); d-mannitol, 1-O-(22-hydroxydocosyl)- (6.00); and dodecanoic acid, 1,2,3-propanetriyl ester (6.73%). These principles may underpin the efficacy of *T. pachysiphon* in folk medicine and possibly explain the reasons for the medicinal uses of this plant by the natives of the Kingdom.

**Keywords:** Ethnobotanical relevance, Folk-medicine, *Tabernaemontana pachysiphon*, Bioactive constituents

## 1. INTRODUCTION

*Tabernaemontana pachysiphon* Stapf (Apocynaceae) (Synonyms: *Conopharyngia pachysiphon, T. angolensis* Stapf*, T. holstii* Schum.) is commonly called *Osi* in Eastern Nigeria (Personal communication, 2020). The genus is native to Africa and is distributed throughout tropical regions of the world [1-2]. *T. pachysiphon* is a small tree that grows up to 15 m in height [2]. The leaves of the plant are leathery, glossy, simple and elliptical in shape. It blooms twice yearly, producing sweet-scented, cup-shaped flowers that are predominantly white or pale yellow in colour [3].

Several members of this genus play roles as shade plants, windbreakers, and ornamentals in gardens and landscapes [1] and are good sources of timber [3]. Plants in this tropical genus serve diverse medicinal purposes for natives in traditional medicine practices across Asia, Americas and Africa [4-6]. Decoctions or infusions of leaves, roots, and stem bark of *T. pachysiphon* and its related species (4g/200 mL; 5 leaves/250 mL) in traditional medicine are effectively used to treat insanity, scabies, hypertension, pains, headache, stomach-ache, constipation, syphilis, ulcers and tumors [1,5,7]. Furthermore, preparations from the plants are used in diverse cultures of the tropics to treat dementia, fevers, and as styptic to control bleeding and facilitate wound healing [6,8]. They have also been used as anti-nociceptive agents to relieve breast inflammation in local settings of Nigeria [9].

Scientifically, the genus *Tabernaemontana* has demonstrated activities such as vasorelaxation, analgesic, anthelminthic, antitumor, antimicrobial and antiviral effects [5,10]. Methanol and hexane leaf and stem bark extracts of *Tabernaeontana* sp. effectively inhibited methicillin–resistant *Staphylococcus aureus*, *Escherichia coli, Pseudomonas aeruginosa, Bacillus subtilis and Candida* sp. [6]. Retardation of growth and development of *Entamoeba histolytica*, *Cryptococcus neoformans* and *Penicilium notatum* by extracts of *T. alternifolia* L., *T. stapfiana* Britten and *T. pachysiphon* have also been documented [1,7,9]. Pallant [13] reported that fractions and isolates from *T. pachysiphon* demonstrated strong antibacterial activities against stubborn clinical strains of *S. aureus* and *Mycobacterium tuberculosis,* causative agent of tuberculosis in humans.

Phytochemical analysis of extracts from different parts of *Tabernaemontana* species has revealed the presence of bioactive alkaloids, including ibogine, voacristine, cononaridine, and conolidine [1,7,9–12]. Additional alkaloids such as tubotaiwine, ovoacangine, ibogaline, 3R-hydroxyconopharyngine, 3S-hydroxyconopharyngine, and 11-demethylconoduramine have been isolated from the stem bark and leaves of *T. pachysiphon* in Kenya and Nigeria [13–14]. Flavonoids, polyphenols, and tannins have also been identified in *T. pachysiphon* and related species [10,15–18]. Fatty acids, including n-hexadecenoic acid, octadecanoic acid, tetracosanol-1, n-nonadecanol-1, α-linolenic acid, and pentadecanoic acid, have been detected in organic root and stem bark extracts of *Tabernaemontana* species [6,19–22].

Statistics show that 80% of the rural populace worldwide still depend on using medicinal plants to assuage disease conditions [23-24]. Owing to the challenge posed by the growing number of antimicrobial resistant strains of medically important pathogens which cause about 5 million deaths per annum around the world [25], scientists are turning their focus to secondary metabolites from higher plants with a view to finding alternative or complementary carbon skeleton leads for drug candidates [26]. These bioactive phyto-ingredients act in synergy and could bind on multiple target sites or elements of complex cellular pathways of disease-causing agents, and thus are less likely for pathogens to develop resistance to them than modern allopathic medicines [6,26]. All these give impetuses to elicit, and to document so as to preserve indigenous knowledge of medicinal plants [26].

Although *T. pachysiphon* is widely used as a medicinal plant by the natives of Ikwuano Local Government Area in Abia State, its ethnobotanical relevance has not yet been systematically documented in this region. Furthermore, while numerous alkaloids have been isolated from the plant, the bioactive fatty acid constituents in its leaves are, to the best of our knowledge, scantily documented or entirely uncharacterized.

Therefore, the ethnobotany and bioactive chemical constituents of *T. pachysiphon* used in the indigenous medicine practices of Ekebedi ancient Kingdom of Ikwuano Local Government Area, Abia State, Nigeria are presented in this work.

## 2. MATERIALS AND METHODS

### 2.1 Survey location

Ekebedi Oboro ancient Kingdom is located in Ikwuano Local Government Area (LGA) of Abia State, in the rainforest belt of Southeast Nigeria. The Kingdom lies on latitude 5° 24' 22"N, longitude 7° 34' 5"E with an elevation of 112 meters above sea level (Fig. 1). Estimates show that about 200 households make up the ancient Kingdom.

A modern community health center is conspicuously absent in the Kingdom, though there are pockets of patent medicine stores dotting its landscape. The majority of the people in the Kingdom practice mixed farming characterized by land rotation, fishing, traditional medicinal practices, lumbering, trading, hunting as well as bone-setting.

The warm and humid rainforest zone of the tropics is endemic to malaria and several other kinds of parasitological diseases as well as metabolic dysfunctions including diabetes. A large number of tropical natives usually administer herbal recipes against attacks of these diseases [6,23,27]. This work seeks hence to validate the use of *Tabernaemontana pachysiphon* by people of Ekebedi Oboro Ancient Kingdom in Abia State, Nigeria, in the treatment of some pathological and metabolic diseases.

### 2.2 Research design and collection of ethno-botanical data

A good research follows a systematic design and scientific protocol. In this study, a survey of the ethno-botanical relevance of *T. pachysiphon* by people of the ancient Ekebedi Kingdom was undertaken. A total population 102 indigenous people made up of 14 herbal practitioners and 26 indigenous herb vendors and 62 native herbal medicine users from the Kingdom and its environs were randomly selected for the study. The survey employed a house-to-house strategy which lasted over a period of 6 weeks (September 01, 2023 – October 07, 2023). Data collection involved interviews and structured questionnaires, as adopted by Mgbeahuruike [23,27]. The survey questions were tailored in such a way as to elicit responses from the target respondents on whether or not they use *T. pachysiphon* in traditional medicine, and if yes, why, when, and how do they use the plant as medicine in the traditional medicine practices in the ancient Kingdom.

### 2.3 Source of plant material

*Tabernaemontana pachysiphon* leaves were obtained from a courtyard in the Ancient Kingdom of Ekebedi Oboro. Fresh aerial parts of the plant were collected and authenticated by Prof. M. C. Dike from the

Department of Forestry, College of Natural Resources and Environmental Management (CNREM), Michael Okpara University of Agriculture, Umudike.

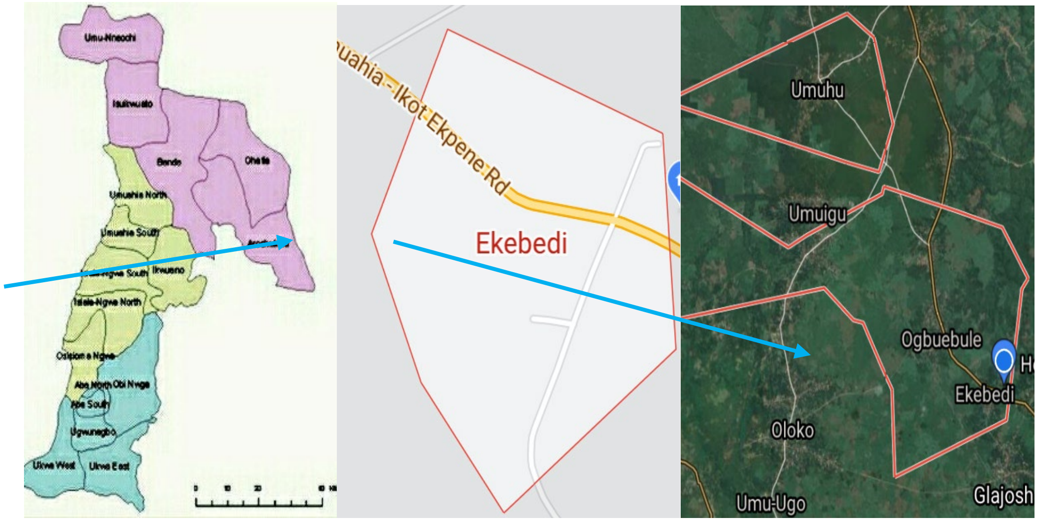



**Figure 1.** Map of the study area. Left: Map of Abia State, Nigeria showing the 17 Local Government Areas (L.G.A.) of the State. Center: Map of Ekebedi Kingdom in Ikwuano L.G.A. of Abia State, Nigeria. Right: Satellite view of the study location terrain

## 2.4 Methanol extraction of *T. pachysiphon* leaves

The leaves of the plant were washed with tap water, and dried on the laboratory bench for 21 days, enveloped and oven-dried at 40ºC for 30 minutes, and then milled into powder using a Thomas Wiley machine (Model: ED-5 USA). Then 500 g of the milled powder was packed separately into a 2-liter Soxhlet apparatus and extracted exhaustively with 1000 ml of analytical-grade methanol for 24 hours. The methanol leaf extract was concentrated using a rotary evaporator at 45ºC and left on the laboratory bench for two days to afford residue [28].

## 2.5 Gas chromatography-mass spectrometry (GC-MS) analysis of the plant residues

This was conducted according to the standard procedure as adopted by Gopinath [29] and Omotosho 30]. The residue *T. pachysiphon* was re-dissolved in methanol. One (1) ml aliquot of the solution was injected into the GC-MS equipment [Model: QP 2010 Plus Schmadzu, Tokyo, Japan) (AOC-20i auto-sampler, VF 5 MS fixed silica capillary column] The carrier gas was helium (99.99%), flow rate (1.58 ml/min), injector and mass transfer line temperature (250 and 200ºC respectively), injection volume of 1 µl was employed at a split ratio of 10:1. The MS ionization energy (70 eV), ion source temperature (200ºC), solvent cut time (2.5 min), and the interface temperature was 250ºC. The total running time of the machine was set to 1 hour.

The percentage of the extract was expressed as percentage with peak and normalization. The relative percentage amount of each phyto-component was calculated by comparing its average peak area to the total areas. The detection process utilized the National Institute of Standard and Technology (NIST) (Version 2.0, 2005) library. The compound prediction was based on the Phytochemical and Ethno-botanical Databases of USDA [31]. The interpretation of GC-MS was conducted using the NIST database having more than 62,000 structural patterns. The spectra of the unknown phyto-components were compared with those of known components stored in NIST library. The names, molecular weights and chemical structures of the extracts-derived compounds were ascertained from the National Institute of Standard and Technology (NIST) [32].

### 2.6 Data analysis

Data generated from this study was analyzed by simple descriptive statistical tools such as range, means and percentages using SPSS computer software version 25.

## 3. RESULTS AND DISCUSSION

### 3.1 Results

#### 3.1.1 Demographic characteristics of respondents in the survey

The results of the study presented in Fig. 2 indicate that in the ancient Kingdom both genders (male and female) of the natives were involved in traditional medicine practices and usage. However, women had a higher involvement, representing 62.01% of the respondents engaged in preparation, vending, and utilization of traditional medicines, compared to their male counterparts, who accounted for only 39.99% (Fig. 2A).

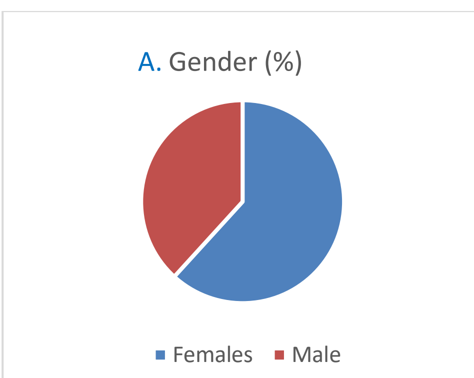
In terms of age, majority of the practitioners of indigenous traditional medicine in the Kingdom clustered around the 21–40-year-old age bracket (51.19%). This was closely followed by those between the ages of 41-70 years (27.33%); while practitioners above 71 years old represented only 3.44% (Fig. 2B).

Regarding education, a significant portion of the respondents had no Western education (42.56%) or had received limited Western education (39.82%). Conversely, those with post-secondary or tertiary education exposure accounted for just 1.67% of the respondents (Fig. 2C).

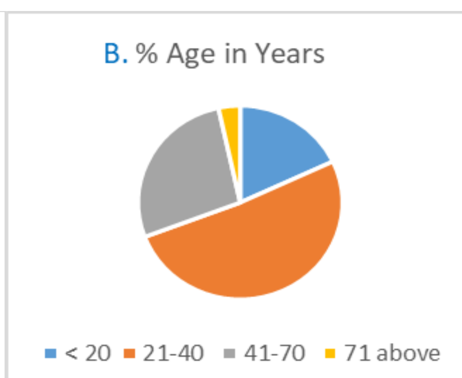
In terms of occupation, farmers (49.69%) and traders/artisans (35.00%) topped the chart of respondents using and vending *T. pachysiphon*-based herbal recipes whereas others who are engaged in elitist occupations were the least (Fig. 2D). Similarly, 47.50% of the herbal medicine users and dealers were of Christian faith, 31.25% others practice typical traditional African religion while 3.08% who were mostly of Yoruba extraction were Muslim faithful (Fig. 2E).

The majority of the indigenous dwellers of the Kingdom are Igbo and they accounted for 76.19% of the people involved in the traditional medicine system of the Kingdom. Next to them are practitioners from the neighbouring Ibibio tribe, which constituted 13.63% of the herbal medicine practitioners and users. The migrants from the Yoruba ethnic group of western Nigeria, however accounted for only 10.18% (Fig. 2F).

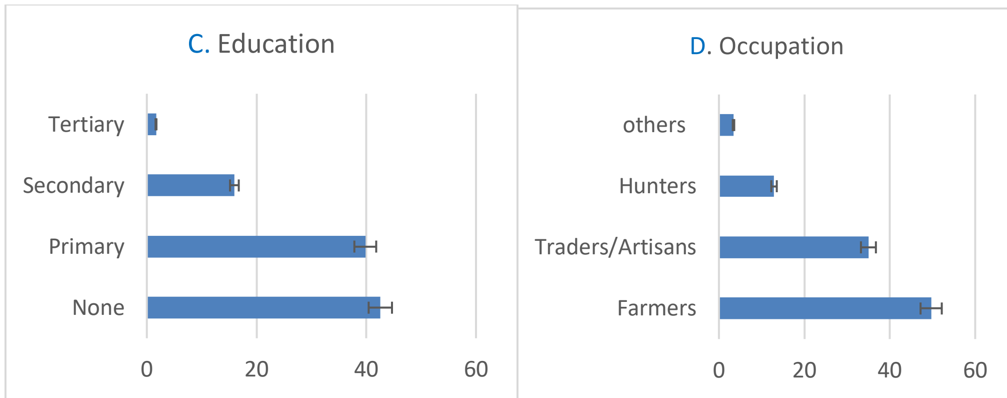
These plant parts are prepared as decoction or infused as teas or soaked in local gins (macerations/elixirs). In the case of poisons, the leaf juice is mixed with honey and given to the patient bitten by snakes, centipedes or scorpions to induce the patients to nausea, and vomiting the poisonous material (Fig. 3).



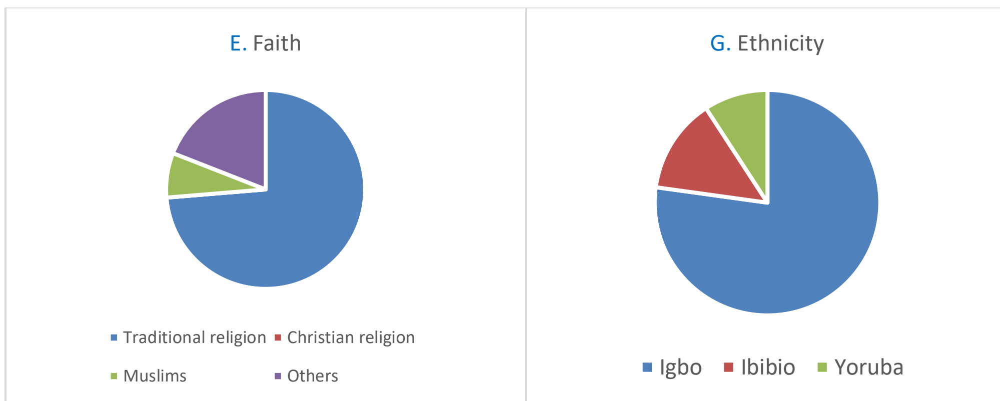

(Fig. 2A)




(Fig. 2B)




(Fig. 2C) (Fig. 2D)




(Fig. 2D) (Fig. 2E)

**Figure 2.** Demographic characteristics of respondents on medicinal significance of the test plant (a = gender, b = age, c = education, d = occupation, e = faith, g = ethnicity)

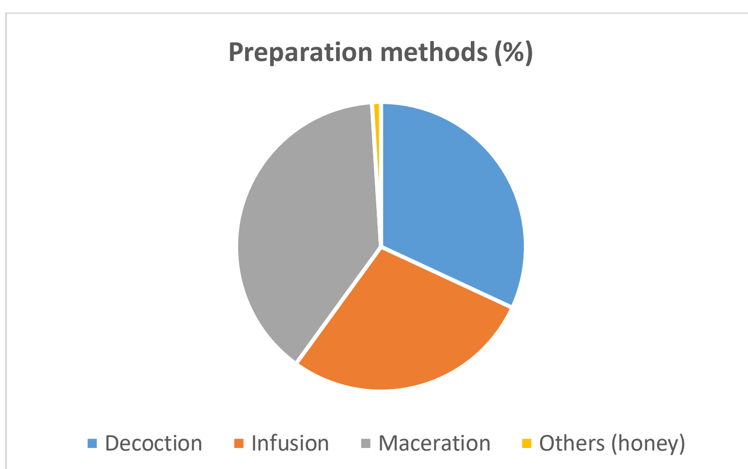



**Figure 3.** Methods of preparation of *T. pachysiphon* crude drug recipes

### 3.1.2 Medicinal relevance of *T. pachysiphon* in the traditional medicine system of Ekebedi ancient Kingdom

The ethnobotanical uses of *T. pachysiphon* in the study area are presented in Table 1. The results of the survey showed that the plant has varied ethno-botanical significance in the area. All respondents in the study agreed that the plant is used for treating cases of diabetes, stomach aches, menstrual pains, ringworms and venereal diseases.

About 80% of the respondents use different preparations of leaves of the plant for treating hypertension and gonorrhea, ringworms and poison, 60% of the respondents prescribe or use the plant for diverse purposes, while 70% others employ the root bark of the plant in treating cases of miscarriages, pelvic inflammatory diseases and venereal diseases (Table 1).

**Table 1.** Survey on medicinal uses and relevance of *T. pachysihon* in ethnobotany of Ekebedi Community and its surrounding villages in Ikwuano LGA, Abia State

| Common/ Local Name of The Plant | Number of Medicine Men Interviewed | Respondents Interviewed (%) | Parts Used for Treatment | Traditional Medicinal Uses and Relevance |
|---|---|---|---|---|
| Giant pin wheel flower | 102 | 80 | Leaf | Malaria fever, typhoid fever, diabetes, poison, dysmenorrhea, hypertension, gonorrhea, ringworms, lacerations, bruises, wounds, dementia, sores, tumors |
| *Osi* in Igbo | 102 | 50 | Stem bark | Fevers, diabetes, stomachache, malaria, ulcers, syphilis, abdominal troubles, gonorrhea, dementia |
| | 102 | 70 | Root bark | Malaria fever, miscarriage, pelvic inflammatory disease (PID), waist pains, venereal diseases, ulcers, sores |
| | 102 | 60 | Seed | Diabetes, menstrual pain, venereal diseases, ringworms |
| *Osi* in Igbo | 102 | 100 | All parts | Diabetes, stomachache, fevers, venereal diseases, hypertension, syphilis, gonorrhea, scabies, dementia, tumors, skin diseases, dysmenorrhea |

### 3.1.3 Mass Spectral data on *T. pachysiphon*

The mass spectral chromatogram of the GC-MS analysis of the volatile components of 2µl of the methanol leaf extract of *T. pachysiphon* is presented in Fig. 4. The data indicated the presence of a wide array of bioactive volatile principles in the plant leaf, and a total of 32 compounds were identified as shown by the spectral peaks (Table 2).

The relative abundance of the compounds ranged from as low as 0.01% recorded for 1-Tridecyn-4-ol to 29.69% obtained for dodecanoic acid 1, 2, 3-propanetrieyl ester. The peaks representing major volatile compounds in the chromatogram include peak 7 which had 9, 12-octadecadienoic acid (Z, Z) methyl ester (16.35%); and peak 8 which had octadecenoic acid methyl ester (E) (19.17%) while peak 30 recorded dodecanoic acid 1, 2, 3-propanetrieyl ester (29.69%). Other compounds identified in sizeable quantities were hexadecanoic acid methyl ester (2.91%) at peak 5, cyclopentadecanone oxime (1.01%) at peak 14 and

Z,Z-8,10-hexadecadien-1-ol (1.82%) at peak 20. Also, peaks 21 and 26 had 9-Octadecenoic acid, 1,2,3-propanetriyl ester E (5.86%) and 9-octadecenoic acid, 1,2,3-propanetriyl ester (E- (1.72%) respectively while d-mannitol 1-o-(22-hydroxydocosyl) (6.00%) and dodecanoic acid, 1,2,3-propanetriyl ester (6.73%) were recorded at Peaks 31 and 32 respectively. Some of the bioactive constituents which were detected in trace quantities (area %) in the methanol extract of *T. pachysiphon* as shown in Table 2 include: cyclononasiloxane, lauroyl peroxide, dichloroacetic acid, cyclopentadecanone oxime, undecanal, 1-octadecyne, hexadecanoic acid 2-methyl ester etc. The structures of some of the bioactive chemical compounds are shown in Fig. 5.

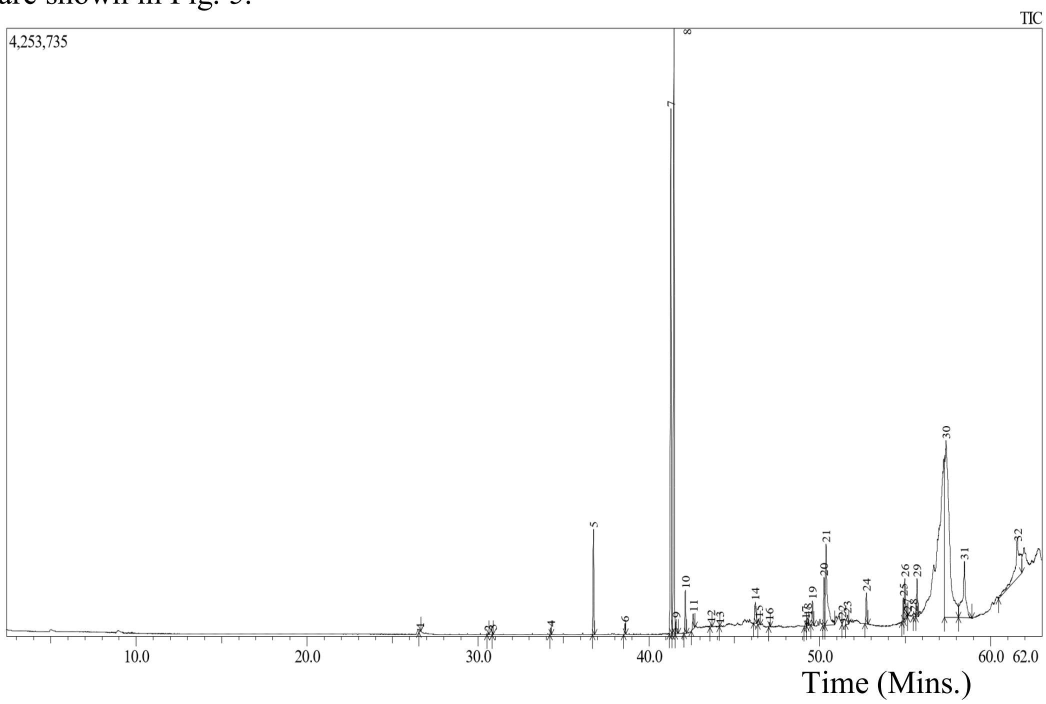



**Figure 4.** Chromatogram of volatile compounds of *T. pachysiphon* leaf

**Table 2:** Bioactive compounds in *T. pachysiphon* methanol leaf extract

| Peak Num. | Retention Time | Area | Peak Area (%) | Height | Height % | A/H | Name of Compound (Constituent) |
|---|---|---|---|---|---|---|---|
| 1 | 26.584 | 71163 | 0.09 | 16422 | 0.12 | 4.33 | Phthalic acid, ethyl pentyl ester |
| 2 | 30.595 | 31132 | 0.04 | 11914 | 0.09 | 2.61 | Propane |
| 3 | 30.851 | 37194 | 0.05 | 18011 | 0.13 | 2.07 | Pentanoic acid, 2-methyl- |
| 4 | 34.253 | 134521 | 0.17 | 47832 | 0.35 | 2.81 | 2-(2',4',4',6',6',8',8'-Heptamethyltetrasiloxan-2'-y |
| 5 | 36.754 | 2314114 | 2.91 | 737134 | 5.38 | 3.14 | Hexadecenoic acid, methyl ester |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 6 | 38.592 | 253355 | 0.32 | 74813 | 0.55 | 3.39 | 2-(2',4',4',6',6',8',8'-Heptamethyltetrasiloxan-2'-y |
| 7 | 41.288 | 13000722 | 16.35 | 3676453 | 26.82 | 3.54 | 9,12-Octadecadienoic acid (Z,Z)-, methyl ester |
| 8 | 41.469 | 15235911 | 19.17 | 4232968 | 30.89 | 3.60 | 9-Octadecenoic acid, methyl ester, (E)- |
| 9 | 41.592 | 406786 | 0.51 | 99316 | 0.72 | 4.10 | 6-Octadecenoic acid, methyl ester, (Z)- |
| 10 | 42.130 | 1000514 | 1.26 | 302490 | 2.21 | 3.31 | Methyl stearate |
| 11 | 42.584 | 460671 | 0.58 | 108772 | 0.79 | 4.24 | Methoprene |
| 12 | 43.624 | 67666 | 0.09 | 17571 | 0.13 | 3.85 | 1-Octadecyne |
| 13 | 44.150 | 8084 | 0.01 | 3380 | 0.02 | 2.39 | 1-Tridecyn-4-ol |
| 14 | 46.217 | 799068 | 1.01 | 150764 | 1.10 | 5.30 | Cyclopentadecanone, oxime |
| 15 | 46.428 | 159768 | 0.20 | 33670 | 0.25 | 4.75 | 14-Heptadecenal |
| 16 | 47.054 | 99756 | 0.13 | 31285 | 0.23 | 3.19 | Methyl 2-hydroxydodecanoate |
| 17 | 49.164 | 164729 | 0.21 | 44614 | 0.33 | 3.69 | 1-Octadecyne |
| 18 | 49.292 | 207023 | 0.26 | 58195 | 0.42 | 3.56 | 1-Octadecyne |
| 19 | 49.572 | 614100 | 0.77 | 169557 | 1.24 | 3.62 | Cyclononasiloxane, octadecamethyl- |
| 20 | 50.247 | 1448690 | 1.82 | 334895 | 2.44 | 4.33 | Z,Z-8,10-Hexadecadien-1-ol |
| 21 | 50.367 | 4659702 | 5.86 | 569101 | 4.15 | 8.11 | 9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E |
| 22 | 51.299 | 50300 | 0.06 | 38106 | 0.28 | 1.32 | Dichloroacetic acid, 4-tridecyl ester |
| 23 | 51.611 | 345252 | 0.43 | 64002 | 0.47 | 5.39 | Lauroyl peroxide |

**Table 2:** Bioactive compounds in *T. pachysiphon* methanol leaf extract (continue)

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 24 | 52.724 | 777478 | 0.98 | 217614 | 1.59 | 3.57 | Cyclononasiloxane, octadecamethyl- |
| 25 | 54.885 | 647612 | 0.81 | 160491 | 1.17 | 4.04 | Z,Z-8,10-Hexadecadien-1-ol |
| 26 | 54.972 | 1422263 | 1.79 | 285774 | 2.09 | 4.98 | 9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E |
| 27 | 55.143 | 47764 | 0.06 | 23573 | 0.17 | 2.03 | 1-Octadecyne |
| 28 | 55.475 | 179672 | 0.23 | 22197 | 0.16 | 8.09 | Undecanal |
| 29 | 55.687 | 1128589 | 1.42 | 266811 | 1.95 | 4.23 | Cyclononasiloxane, octadecamethyl- |
| 30 | 57.389 | 23599845 | 29.69 | 1230741 | 8.98 | 19.14 | Dodecanoic acid, 1,2,3-propanetriyl ester |
| 31 | 58.462 | 4766783 | 6.00 | 392591 | 2.86 | 12.14 | d-Mannitol, 1-O-(22-hydroxydocosyl)- |
| 32 | 61.579 | 5352187 | 6.73 | 264508 | 1.93 | 20.23 | Dodecanoic acid, 1,2,3-propanetriyl ester |
| | | 79492414 | 100.00 | 13705565 | 100.00 | | |

*Peak area = % composition of compound in the leaf extract; Num. = Number; A/H = Area/Height

### 3.2 Discussion

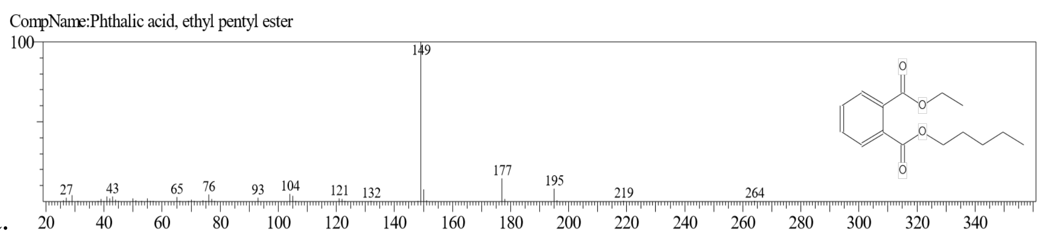
Results presented in Fig. 2 show the demographic characteristics of the population surveyed in this study. The findings indicate that more women than men use *T. pachysiphon*-based herbal medicines. This is in strong agreement with observations of Onyipat et al. [34] in Enugu, southeast Nigeria, but inconsistent with the views of Aina et al. [35] where men dominated the usage of herbal medicine in Ibadan, western Nigeria. Data from this study indicated that middle-aged people were largely involved with preparations and usage of *T. pachysiphon* as medicine in the study area. This is, however, not consistent with the reports of Aina et al. [35] where usage of herbal medicines was common amongst the elderly who are above 70 years old. On the other hand, findings from this study where respondents with religious background in orthodox Christian and traditional faith systems used more of herbal medicine is also in accord with the views expressed in previous studies [35].

This study also showed that the usage of traditional medicine is higher amongst natives with little or no formal education, a view sustained by submissions of earlier findings [35,36]. Ethnicity, education, employment and family size and financial status have been reported as some of the principal predictors of usage of traditional medicines amongst aborigines [37]. Findings in this survey indicated that Yoruba migrants who dwell in the Kingdom were less likely to use *T. pachysiphon* recipes compared to their Igbo and Ibibio counterparts who make up the majority of inhabitants of the Kingdom. This observation is in strong agreement with previous reports [35].

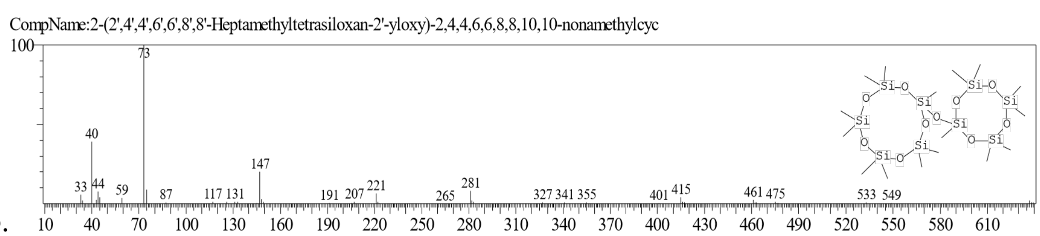
Moreover, findings from this study indicated that respondents with less than secondary education were more likely to use herbal medicines to assuage disease conditions, this view aligns with observations made by previous workers [35-37]. Some workers were of the view that usage of traditional medicine was common amongst traders and artisans [36]. This aligns with findings from the current study, where **farmers** and **traders** were the dominant groups engaged in the preparation and use of *T. pachysiphon*-based recipes for treating medical conditions.

Additionally, earlier studies have reported that **affordability, accessibility,** and **efficacy** are the primary drivers behind the widespread use of herbal medicines in local environments. Given the absence of modern private or public healthcare centers and clinics in the Kingdom, these factors likely explain the high utilization of traditional medicines, including *T. pachysiphon* recipes, among the surveyed population.

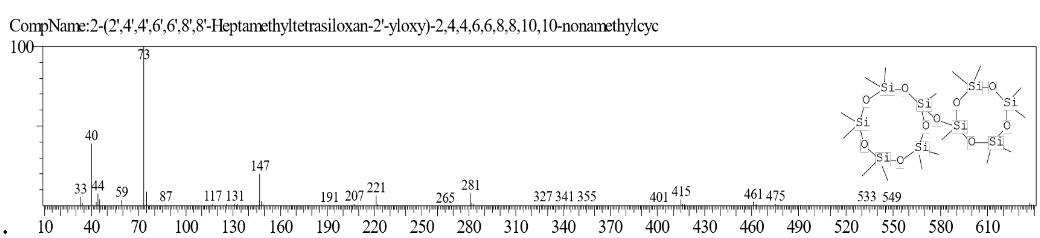
The results presented in Table 2 indicate that the traditional medicine men of the study area use all parts (leaves, stem and bark and seeds) of *T. pachysiphon* as medicine against wounds, metabolic and pathological diseases. Amongst the many diseases for which the plant finds medicinal use include dementia, tumors, malaria, dysmenorrhea, fevers, diabetes, stomach troubles, syphilis and gonorrhea, and other venereal diseases as well as scabies amongst other skin diseases.



a.

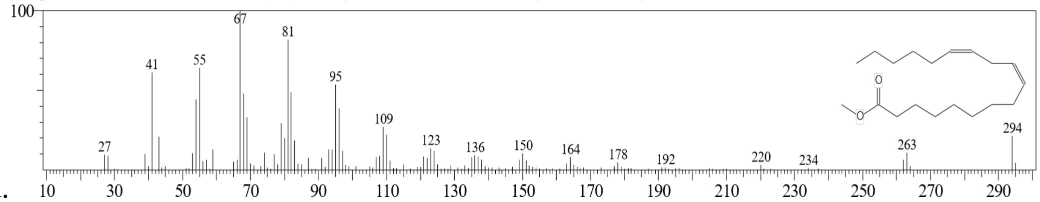


b.



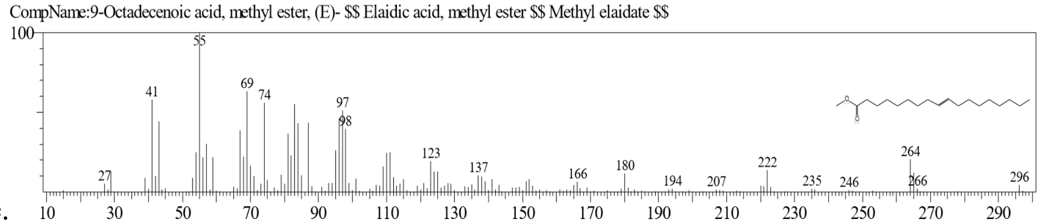
c.

CompName:9,12-Octadecadienoic acid (Z,Z)-, methyl ester $$ Linoleic acid, methyl ester $$ Methyl cis



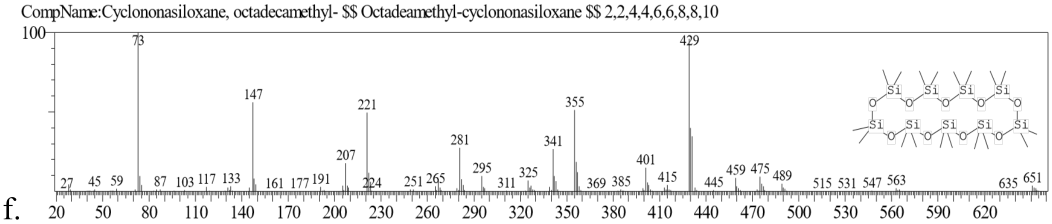

d.

CompName:9-Octadecenoic acid, methyl ester, (E)- $$ Elaidic acid, methyl ester $$ Methyl elaidate $$



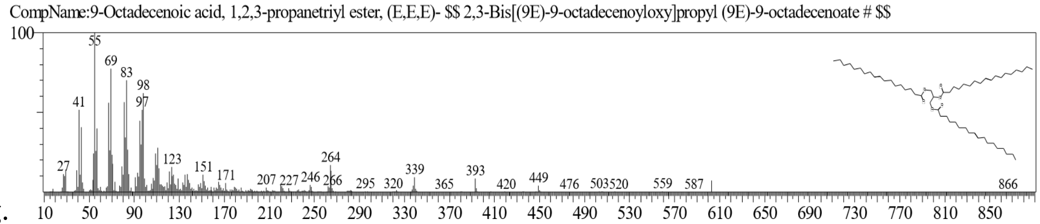

e.

CompName:Cyclononasiloxane, octadecamethyl- $$ Octadeamethyl-cyclononasiloxane $$ 2,2,4,4,6,6,8,8,10



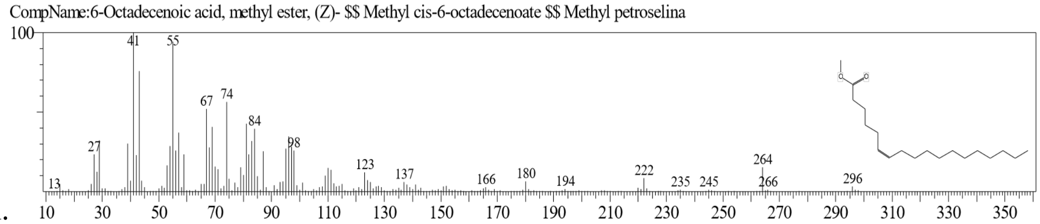

f.

CompName:9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E,E,E)- $$ 2,3-Bis[(9E)-9-octadecenoyloxy]propyl (9E)-9-octadecenoate # $$



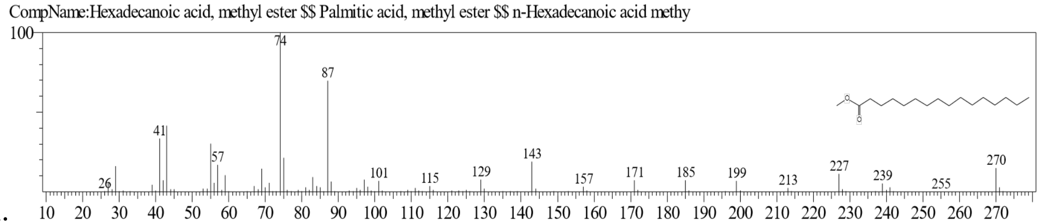

g.

CompName:6-Octadecenoic acid, methyl ester, (Z)- $$ Methyl cis-6-octadecenoate $$ Methyl petroselina



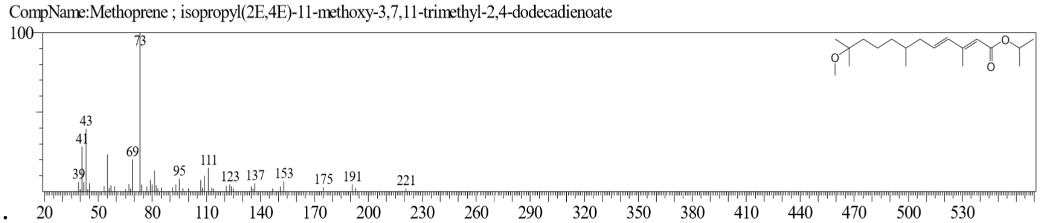

h.

CompName:Hexadecanoic acid, methyl ester $$ Palmitic acid, methyl ester $$ n-Hexadecanoic acid methy




i.

CompName:Methoprene ; isopropyl(2E,4E)-11-methoxy-3,7,11-trimethyl-2,4-dodecadienoate




j.

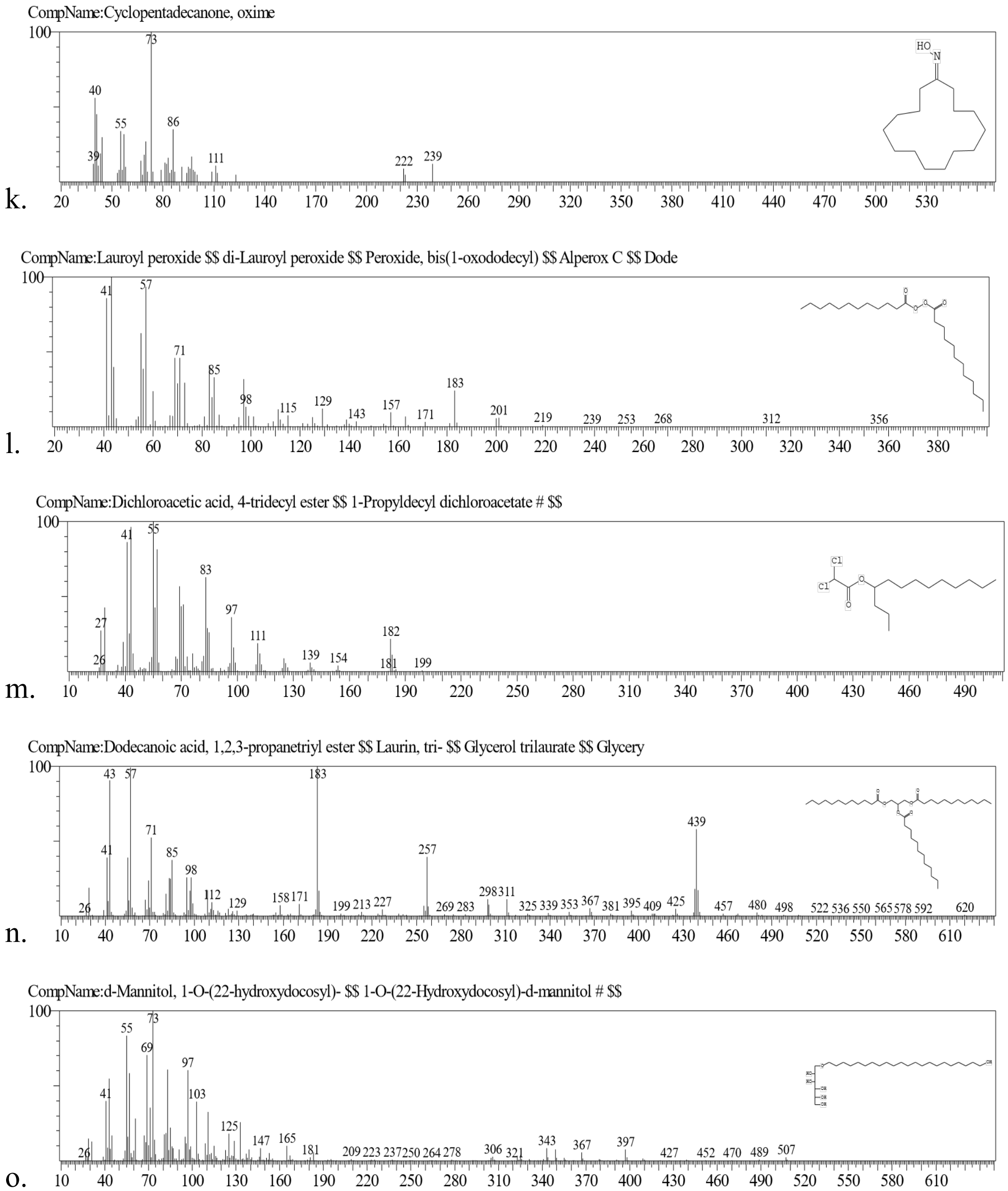



**Figure 5.** Structures of some bioactive chemical compounds from *Tabernaemontana pachysiphon* leaf. a. Phthalic acid, b. 2-(2',4',4',6',6',8',8'-Heptamethyltetrasiloxan-2'-y; c. 2-(2',4',4',6',6',8',8'-Heptamethyltetrasiloxan-2'-y, d. 9, 12-Octadecedienoic acid ME, e. 9-Octadecanoic acid ME, (E)-, f. Cyclononasiloxane, g. 9-Octadecenoic acid, 1,2,3-propyltryl, h. 6-Octadecenoic acid, i. Methaprene, j. Hexadecanoic acid ME, k. Cyclopentadecanone, l. lauryl peroxide, m. Dichloroacetic acid, n. Dodecanoic acid, o. d-mannitol.

In the case of fevers, the plant leaves alone or in conjunction with other parts of the plant are decocted and used in the early hours of the morning to steam the body of the patient, followed by a bath, and a cup (100 ml) of the decoction taken twice daily. Latex from the leaves and stem bark is used as a styptic, and in some cases, a poultice prepared from the leaves, twigs, and stem bark is applied to palliate wounds, lacerations, sores, and to promote the healing of ulcers. The plant is also prepared as tea or hot infusions to alleviate dementia, insanity, and other mental health conditions.

Other respondents agreed to prescribing the plant as a gin-based recipe (elixirs, macerations) of the stem or root bark for matters suspected to be venereal diseases such as herpes, gonorrhea and syphilis, enteric fever,

stomach aches, and dysmenorrhea. This agrees with the reports of Omino and Kowaro [3] and Elia [1] that tropical natives use preparations of members of this genus to treat venereal, abdominal and skin diseases amongst several other ailments.
In conditions relating to diabetes for example, data obtained indicate that the seeds are ground and mixed with coconut (*Cocos nucifera*) water and consumed about 50 ml thrice daily. While leaf juice is applied to itching skin, decoction or infusion of the leaves is drunk to palliate ringworm, scabies and other irritation of the skin. These findings are also consistent with the submissions of Elia [1], who reported the use of *T. pachysiphon* in the treatment of scabies.
For issues suspected to be toxicity from ingesting contaminated substance or poison as a result of snake bites, centipede or scorpion stings on the other hand are treated by mixing leaf juice of the plant with honey and given to the patient to neutralize and induce vomiting of the poisonous substances or their mucilaginous derivatives. This is congruent with the views of Vineetha et al. [33] who noted that in Brazil, Singapore and elsewhere, roots, leaves and flowers *T. alternifolia*, *T. divaricarta* (L.) R.Br. ex Roem. Schult. and *T. catharinensis* (A. DC) are used by native Latin Americans and Asians for the treatment of snake bites such as the Indian cobra, *Echis carinatus* and scorpion stings.
Members of *Apocynaceae* remain one of the primary sources of medicines used in traditional healthcare systems of the tropics to treat numerous diseases including infections, tumors and cancers [24]. Data from this survey showed that all parts of the plant are decocted or macerated in local gin (*kai kai*) and taken to ameliorate hyperglycemia, and to deflate tumors or for arresting development and spread of carcinomas; a view which is in strong accord with the reports of several other previous researchers [4,38-39].
Additionally, alcoholic extracts of leaves and/or root and stem bark of the plant is also used by the native as analgesic against joint aches, and body pains. Edinoff [10] found alkaloidal isolates from *Tabernaemontana* sp. as a chronic pain killing compound. This lends support to the use of *T. pachysiphon* preparations by natives of the study Kingdom as analgesic as seen in this study.
The findings from this study are also consistent with Boligon [40] who reported that *Tabernaemontana* species play important role in complementary medicine of tropical countries, by minimizing oxidative stress-induced diseases, inflammations and tissues damages. Toxicity evaluations revealed that aqueous root bark extract of *T. pachysiphon* (5g/kg) induced neither death nor observable toxicity in mammals [41]. Similarly, fluorescent organoleptic and elemental analysis of the leaves, stem and latex of its close relative, *T. ventricosa* Hochst. ex. A.DC., vindicated the genus of noxious compounds [6].

Data from this survey indicate also that traditional medicine men and natives of the Kingdom use water, local gins and honey to prepare their *T. pachysiphon* medicines. This is consistent in part with the view expressed by some workers about solvents for making traditional medicines as reported by Abduralman and Kolawale [42] and Mgbeahuruike [27]. However, the use of honey as a medium to prepare herbal recipes as noted in this study is divergent from their submissions.

Several bioactive fatty acids and volatile compounds have been identified and characterized from aqueous and organic extracts of many genii of higher plants including *Tabernaemontana* species [43-46]. In this study, different bioactive chemical compounds were identified as shown in the chromatogram of *T. pachysiphon* (Table 1). The predominant compounds were dodecanoic acid 1,2,3-propanetriyl (lauric acid) (29.68%), 9-octadecenoic acid methyl ester (oleic acid) (19.17%), and 9,12-octadecadienoic acid (Z,Z)-methyl ester (linoleic acid) (16.35%). Oleic, α-linoleic and pentadecanoic acids were also found to be major bioactive fatty acids in GC-MS analysis of hexane and methanol leaf, stem and latex extract of *T. ventricosa and T. pachisiphon* [20]. In parallel studies, different solvent extracts of the leaves and seeds oil of *Cnidoscolus aconitifolius*, *Entandrophragma angolense*, Ajuga *relicta*, and bitter apple afforded 9,12-octadecadienoic acid (Z,Z)- methyl (43.20-50.3%), oleic and palmitic acids (23.51-29.50%) amongst others as the most abundant constituents of the plant materials [46-49]. These results are in tandem with findings in this study where these volatile compounds were detected abundantly in *T. pachysiphon* leaf (Table 2).

Data from this study indicated that dodecanoic (lauric) acid was the highest occurring compound in methanol leaf extract of *T. pachysiphon* in this study (Table 2). This is however, not in accord with reports of previous workers where n-hexadecanoic acid an analogue of octadecanoic acid [44] had the highest peak areas ranging between 11.18-27.49% in methanol stem, leaf and flower extracts of *T. pachysiphon* and *T. divaricata* [19-21,43]. Differences in age, genetic constitution, parts of *Tabernaemontana* species, geographic origin, type of extracting solvent, or particle size of the plant powder used in this study may explain the disparities in type and amount of constituents afforded by the test plants.
Strong biological and antimicrobial activities have been attributed to 9, 12-Octadecadienoic acid (Z, Z) - methyl ester, 9-Octadecenoic acid methyl ester, and Dodecanoic acid 1, 2, 3-propanetriyl), the predominant ingredients afforded by *T. pachysiphon* in this study [30,50]. Linolenic acid (9, 12-octadecadienoic acid (Z, Z) - methyl ester, has been associated with hepato-protective, anti-acne, anti-inflammatory, anti-cell proliferation, and anti-cancer activities [19]. Hexadecanoic and n-hexadecanoic (palmitic) acids which are analogues of linolenic acid, exhibits antioxidant, anti-diabetic, anticancer, antimicrobial, anti-enzyme and anti-viral activities [19,44-45]. Hexadecanoic acid present in significant amounts in aqueous root and fruit extracts of *Carica papaya* L., was fingered for recovery of diabetic rat exposed to aqueous root treatment of *C. papaya* in a trial [44-45]. Natives in the present study area use *T. pachysiphon* for the treatment of diabetes, and these esters may be responsible for its medicinal efficacy against the disease.
High concentration (29.68%) of lauric acid (dodecanoic acid 1, 2, 3-propanetriyl) was recorded in methanol extract of *T. pachysiphon* in this study. Kernels of *Cocos nucifera* L. and *Elais guineense* Jacq. kernel oils are rich in dodecanoic acid 1, 2, 3-propanetriyl [51-52]. This ester compound was reported to demonstrate strong antimicrobial and antibacterial potentials against a wide array of bacteria, as well as antiviral activities against HIV-I and HIV-II, HSV-I and II, measles virus, and human stomatitis virus etc. [53]. It is also thought to inform the protective properties of breast milk in neonates. In addition, antibacterial activity against medically important bacterial pathogens including *S. aureus, S. epidermidis, Streptococcus agalactiae, E. coli, Chlamydia pneumonia, Propionibacterium acne* due to this ester have been documented [54-55]. Lauric acid (5%) exhibited potent anti-*Streptococcus* sp. and *S. aureus* activity comparable with ciproxacin. The growth and reproduction of *Candida albicans* was also retarded by this compound *in vitro* [56-57]. In crop protection, this ester effectively impeded mycelial growth of *Rhizoctonia solani* and *Pythium ultimum in vitro* and barred expression of powdery mildew (*Blumeria graminisi f. sp hordei*) of bailey plants in the field [58]. These compounds may be the major reasons underscoring the fungitoxic and antimicrobial effects of this plant against ringworms and diarrhea amongst others in ethno-medicine.

Earlier investigations revealed the presence of hexadecenoic acid, octadecane and oleate in *A. flourubunda* [59] and hexadecenoic acid methyl ester, 1,2,3-propanetriiyl ester (12.8%) from oil fraction of *Alstonia boodei* [60-61]. These compounds were also identified in varying concentrations in this study. Several of the volatile compounds identified in this study such as phthalate, hexadecenoic acid methyl esther, cyclononasiloxane octadecamethyl, dichloroacetic acid (DCA), 6-Otadecenoic acid and lauryl peroxide possess significant antifungal activities (Table 2). Antifungal activity of some of these compounds against *Aspergillus fumigatus* has been reported to be due to oxidation of carbon-hydogen bond, altering radical chemistry, nucleophilic substitution and electrophilic ring opening [62]. Similarly, a recent evaluation of the volatile constituents of the seed oil attributed its antibacterial activity against species of *Kiebsiella* and *Salmonella* to 11-octadecenoic acid methyl ester [47].

Cyclononasiloxane octadecemethyl was identified in this study on *T. pachysiphon* (Peaks 19, 24, 29) The compound was associated with anti-cancer, anitioxidant, antibacterial, and antifungal activities [31,63]. Cyclononasiloxane octadecemethyl was implicated for impeding the growth and development of *Penicillium digitatum, Fusarium solani, Aspergillus niger* and *Borytis cinerea* by oil of *Marribium vulgare* [64].

GC/MS analysis of n-hexane extracts of neem leaf, stem and root barks besides affording 11-octadecenoic and 13-octadecenoic acids, also yielded dibutyl phthalate, and oxime. These compounds amongst others demonstrated significant antifungal activity [65-66].

Similarly, lauryl (benzoyl) peroxide is an anti-acne, bleaching and oxidizing agent [67]. It showed superior antibacterial activity against *Proteus* species*, S. aureus* and *Pseudomonas aureginosa* over gentamicin and phenoxyethanol which are known antibacterial agents [68]. Phthalic acid (ethyl pentyl ester) recorded in this study (Table 2) is associated with strong anti-enzyme and antioxidant activities. The compound in previous crop protection studies has shown anti-enzyme activity against several enzymes including chitinases responsible for degrading the chitin exo-skeleton during pre-molt phase in insects [69]. These workers found that diethyl phthalate and dibutyl phthalate were fungitoxic to *Aspergillus* sp., *Fusarium* spp*., Candia* spp*., Drechslera haloides, Alternaria* sp. and *Rhizoctonia solani* [70-72]. This activity was thought to be due to infringement of the actions of the extracellular enzymes of the fungus that played important roles in its nutrition and growth [69]. Similarly, the anti-enzyme activity against pyruvate dehydrogenase kinase (PDK), anti-cancer and antifungal activities against *Rhizoctonia solani* and *F. oxysporum* of dichloroacetic acid (DCA) recorded for *T. pachysiphon* (Table 2) have been reported. DCA has been identified from hexane extract of marine macroalgae [73]. The mechanism of action of this compound is linked to pyruvate dehydrogenase kinase inactivation, and fostering oxidative phosphorylation in target organs [63]. Its anticancer activity is suggested to hinge on depolarizing abnormal cells in the mitochondria of glioblastoma cancer cells leading to induced apoptosis amongst others [74]. The compound also played roles reducing lactate levels in Leigh syndrome patients. Z, Z, 8, 10-hexadecenoic acid (Table 2) has been reported as an important steroid compound, having similar activity to pheromones against insect pests [75-76]. As a result, it could be useful as eco-friendly insect attractant in integrated pest management (IPM) programs. Unfortunately, most of these compounds were identified at slightly sizeable or trace quantities in *T. pachysipon*. Medicinal plants contain a wide spectrum of chemical compounds known to act jointly or synergistically to bring about resultant effects that usually surpass total activity of any one single constituent [6,43]. Presence of some these compounds [77] in the test plant used in this study may have contributed to medicinal actions of the test plant in the indigenous medical practices of people of the Kingdom and its environs.

## 4. CONCLUSION

The results of this study indicate that *T. pachysiphon* is used in the herbal medicine practices of people of Ekebedi ancient Kingdom and its environs. The plant is used in the treatment of various diseases including fevers, pains, malaria, inflammatory conditions, diabetes, tumors, dementia, syphilis, scabies, and pathogenic infections etc.

Crude medicinal recipes are prepared in various forms, such as infusions, decoctions, alcoholic macerations in local gin (*kai kai*) or leaf juice mixed with honey in cases involving snake and centipede bites or scorpion stings. The study found that more women than men, farmers/traders, people with no education or that has primary education, orthodox Christians or traditional worshippers and grossly of Igbo or related ethnicity utilized *T. Pachysiphon* as medicine more. GC-MS signature carried out on the methanol leaf extract of the plant identified a large spectrum of fingerprinting consisting of 32 bioactive compounds in the leaf extract. To the best of our knowledge this is the first time that the spectrum of fatty acids components of leaf of *T. pachysiphon* is being reported. These fatty acid ingredients likely contribute to and underscore, at least in part, the plant's efficacy and relevance in the folk medicine practices of the Kingdom's people.

## 5. ACKNOWLEDGMENTS

The authors would like to express profound gratitude to all the traditional medicine men of Ekebedi ancient Kingdom and its environs who volunteered indigenous knowledge for this work and, the Staff members of the Analytical Chemistry Unit, FIIRO, Oshodi Lagos for carrying out GC-MS analysis of the plant samples.

## REFERENCES

1. Elia, J., 2006. *Tabernaemontana pachysiphon* Stapf. In: Schmelzer, G.H. & Gurib-Fakim, A. (Editors). Prota 11(1): Medicinal plants/Plantes médicinales 1. [CD-Rom]. PROTA, Wageningen, Netherlands.
2. Pratchayasakul, W., Pongchaidecha, A., Chittipakorn N. and Chittipakorn, S. (2008). Ethnobotany and ethnopharmacology of Tabernaemontana divaricata. Indian Journal Medicinal Plant Research 127: 313-335.
3. Omino, E.A., Kokwaro, J.O. (1993). Ethnobotany of apocynaecea species in Kenya. J. ethnopharmacol. 40(3): 167-180. DOI: 10.106/0378-8741(93)90065-D.
4. Mansoor, T.A., Borrhalno, P.M., Dewanjee, S…Ferreira, M-J.U. (2013). Monoterpen bisindole alkaloids from the African medicinal plant *Tabernaemontana elegans*, induce apoptasis in HCT116 human colon cancer cells. J. Ethnopharmacol. 149(2); 463-470 DOI: 10.1016/j.jep.2013.06.051.
5. Abubakar. I. B., Loh, H-S. (2016). A review of the ethnobotany, Pharmacology and phytochemistry of *Tabernaemontana corymbosa*. J. Pharm. Pharmacol. 68(4): 423-432. DOI: 10.1111/jphp.12523.
6. Naidoo, C.M., Naidoo, Y., Dewir, Y.H., Singh, M., Lin, J. (2023). Phytochemical composition and antibacterial evaluation of *Tabernaemontana ventricosa* Hochst Ex A. DC leaf, stem and latex extracts. South Afr. J. Bot. 152: 147-164. DOI: 10.1016/j.sajb.2022.11.026.
7. Uwumarongie, O.H., Onwukaeme, D.N., Obasuiyi, O. (2007). Antimicrobial activity of methanolic leaf extract of *Tabernaemontana pachysiphon* Stapf. Nigerian Journal *Natural products Medicine* 11: 23-25. DOI: 10.4314/njnpm.v11i1.11872.
8. Edinoff AN, Patel AS, Baker MW, Lawson J…Kaye AD. (2021). Conolidine: A novel plant extract for chronic pain. Ansth. Pain Med. 11(6): e121438 DOI: 10.5812/aapm.121438.
9. Duru, C.M., Mbata, T.I. (2010). The antimicrobial activities and phytochemical screening of ethanolic leaf extracts of *Hedranthera bateri* Hook and *Tabernaemontana pachysiphon* Stapf. J. Develop Biol. Tissue Engineering 2(1): 001-004.
10. Ruttoh, E.K., Bli, C., Tarus, P.K., Karini, L.K., Okemo, P. (2009). Antifungal activity of *Tabernaemontana stapfiana* Britten organic extracts. Pharmacognosy Research 1: 387-391.
11. Marathe, M.P., Rasane, M.H., Kumar, H., Patwardhan, A.A., Shouche, Y.S., Diswanan, S.S. (2013). *In vitro* antibacterial activity of *Tabernaemontana alternifolia* stem bark aqueous extract against clinical isolates of methicillin resistant *Staphylococcus aureus*. Annals of Clinical Microbiology and Antimicrobials 12: 26-33.
12. Duru, C.M., Anyadoh-Nwadike, S.O., Okechukwu, R.I. (2015). Antimicrobial activity and phytochemical analysis of aqueous and ethanolic extracts of the bark of *Tabernaemontena pachysiphon* Stapf. Scientific Journal of Public Health 3(1-5): 008-013.
13. Hoft, M., Verporte, R., Beck, E. (1998). Leaf alkaloid contents of *Tabernaemontana pachysiphon* as influenced by endogenous and environmental factors in natural habitats. Planta Med. 64(2): 148-152. DOI: 10.1055/s-2006-957393.
14. Beek, T. A., Kaujlaers, F. L. C., Thomassen, P. H. A. M. Verporte, R., Svendsen, A. B. (1984). Phytochemistry 23(8): 1771-1778.
15. Pallant, C.A., Cromarty, A.D., Steemkamp, V. (2012). Effect of an alkaloidal fraction of

*Tabernaemontana elegans* (Stapf.) on selected micro-organisms Journal Ethnopharmacol 140(2):398-404. Doi: 10.1016/j.jep.2012.01.036.

16. Piana, M., Boligin, A.A., de Brum, T.F., Zadra, M,…Boligan, A.A., Athayde, M.L. (2014). Phytochemical analysis and antioxidant capacity of *Tabernaemontana catharinensis* A. DC fruits and branches Anais de Academia Brasiliera de Ciencias 86(2): 881-888.
17. Pallant. A. C. (2010). Bioactivity of alkaloidal fraction of *Tabernaemontana elegans* Stapf. Thesis (*MagisterScientiae*) submitted to the Department of Pharmacology, Faculty of Health Sciences, University of Pretoria, 2010.
18. Naidoo, C.M., Naidoo, Y., Dewir, Y.H., Murthy, H.M., El-Tendawy, S., Al-Suhaibani, N. (2021). Major bioactive alkaloids and biological activities of Tabernaemontana Species. MDPI Plants 10: 313 Doi: 10.3390/plants.10020313.
19. Kalaimagal, C., Umamaheswari, G. (2015). Bioactive compounds from leaves of *Tabernaemontana divaricata* (L.). Int. Journal Recent Res. 6(4): 3520-3522.
20. Uwumaringie, H.O., Onwukaeme, D.N., Ighodaro I. (2018). GC-MS metabolite profiling, antnorciceptive and antipyretic activities of methanol stem bark extract of *Tabernaemontana pachysiphon* Stapf (Apocynaceae). Journal Sci. Practice 5(1): 228-230.
21. Kalaimagal C. (2019). Identification of bioactive compounds in flower of *Tabernaemontana divaricata* (L.) using GC-MS analysis. Asian J. Pharm. Clin. Res. 12(9): 129-132. 34559. Doi: 10. 22139/ajpcr.2019.v12i9.34559.
22. Kim, B-R., Kim. H.M., Jin, C.H., Kang, S-Y., Kim, J.B., Jean, Y.G., Park, K.Y., Lee, I-S., Han. A-R. (2020). Composition and antioxidant activities of volatile organic compounds in radiation-bred Coreoppsis cultivars. MDPI Plants 9: 717 Doi: 10.3390/plants9060717.
23. Mgbeahuruike, E.E., Yrjonen, T., Vourela, H., Holm, Y. (2017). Bioactive compounds from medicinal plants: focus on *PIper* species. South African J. Botany112: 54-59.
24. Kadija, E.O, Adelfattah, E.M, Laila, B, Dalila B. (2021). Ethnobotanical study of medicinal plants used in the treatment of cancers in the city of Casablanca (West-Central Morocco). Tropical Journal Nat. Prod. Research 5(6): 1044-1054.
25. Antimicrobial Resistance Collaborators (2022): Global burden of antimicrobial resistance in 2019: A systematic analysis. Lancet 2022 399 (10325): 629-655 DOI: 10.1016/S0140-6736(21)027274-0. Erratumin: Lancet 2022 400(10358): 1102. DOI: 10.1016/S0140-6736(21)02653-2.
26. Santhyami, L., lina, A., Putri, A. (2024). Exploring urban ethnobotany: A case study of medicinal plants traded in Gede Hardjonagoro market, Surakata, Indonesia. Trop. J. Nat. Prod. Res. 8(4): 6839-6851.
27. Mgbeahuruike, E.E., Holm, Y., Vourela, H., Amandikwa, C., Fyrquist, P. (2018). An ethnobothanical survey and antifungal activity of *Piper guineense* used for the treatment of fungal infections in West African traditional medicine. J. Ethnopharmacology 229: 157-166.
28. Okwu, D.E., Ukanwa, N. (2010). Isolation and characterization of flavonoids chalcones and anthocyanidines from Bridelia ferruginea Benth. Der Chemica Sinica 1(2): 21 – 28.
29. Gopinath, S., Sakthidevi, G., Muthukumaraswamy, S., Mohan, V.R. (2013). GC-MS analysis of bioactive constituents of *Hypericum mysorense* (Hypericaceae). Journal of Current Chemistry Pharmaceutical Sciences: 3(1): 6-15.
30. Omotosho, A.E., Oluwafemi, E.O., Maikailu, S. (2014). Phytochemical analysis of *Cnidoscolus*

*aconitifolius* (Euphobiaceae) leaf with spectrometric techniques. Nigerian Journal of Pharmaceutical and Applied Research 3(1): 38-49.

31. USDA (United States Department of Agriculture) (1995). Names and structural partterns of phytochemicals. In: Dr Jim Dukes Phytochemical and Ethnobotanical Databases, UDSA, 1995. http://phytochemicals.nal.usda.gov, Accessed September 23, 2017.
32. NIST (National Institute for Standards and Technology). https://www.nist.gov/pml.
33. Vineetha, M. S., Bhavya, J., Veena, S. M., Mirajkar, K. K., Muddapur, U., Ananthraju, K. S., Zarmeer, F., More, S. S. (2020): In vitro and in vivo inhibitory effects of *Tabernaemontana alternifolia* against Naja naja venom. Saudi Pharm. J. 28(6): 692-697 DOI: 10.1016/j.jsps.2020.04.010.
34. Onyipat, J-L. E., Okoronkwo, I. L., Ogbonnaya, N. (2011). Complementary and alternative medicine use among adults in Enugu, Nigeria. BMC Complement. Alternative Med. 11: 19. DOI: 10.1186/1472-6882-11-19.
35. Aina, O., Guatam, L., Simkhada, P., Hall, S. (2020). Prevalence and determinants of knowledge about herbal medicine and non-hospital utilization in southwestern Nigeria: a cross sectional study. BMJ Open 10(9): DOI: 10.1136/bmjopen-2020-040769.
36. Li et al. (2020). Traditional medicine usage among adult women in Ibadan, Nigeria: A cross sectional study. BMC Complement. Med. Ther. 20: 93 DOI: 10.1186/s12906-020-02881-z.
37. Jackson, I. L., Akpan, M. R., Akwaowoh, A. E., Sampson, V. I. (2024). The attributes and determinants of herbal medicine use amongst pregnant women attendin antenatal clinics in Uyo, Nigeria. J. Herbal Med. 46: DOI: 10.1016/j.herbmed.2024.100891.
38. Poorima, K., Gopalakrishnan, V.K. (2014). Anticancer activity of *Tabernaemontana coronaria* against carcinogen induced clear cell renal cell carcinoma. Chinese J. Biol. 2014(1): 584074.
39. Puranik, S., Hiremath, M., Neril, R., Ghagane, S. (2018). Evaluation of in vitro antioxidant and anticancer activities of Tabernaemontana divaricata leaf extracts against T-24 human bladder cell lines. Int. J. Cancer Res. DOI; 10.3923/ijcr.2018.100.108.
40. Boligon, A.A., Piana, M., Schawnz, T.G., Periera, R.P., Rocha, J.B.T., Athayde, M.L. (2014). Chromatographic and antioxidant capacity of *Tabernaemontana cathar*inensis. Nat Prod Comm 9 (1): 61-64.
41. Uwumarongie, O.H., Onwukaeme, D.N. (2011). Pharmacognostic and toxicity evaluation of the stem bark of *Tabernaemoontana pachysiphon* Stapf. (Apocynaceae). Nigeria Journal of Pharmaceutical Research 9(1): 63-71
42. Adulrahaman, A. A., Kolawole, O. M (2006). Traditional preparations and uses of maize in Nigeria. Ethnobotanical Leaflets 10: 219-227.
43. Ambukkarasi, M., Thomas, P. A., Sundarajan, M., Pitchairaj G. (2016). GC-MS analysis and in vitro antioxidant activity of ethanolic extract of leaves of *Tabernaemontana divaricata*. Pharmacognosy J. 8(5): 451-458.
44. Ezekwe, S.A,. Chikezie, P.C. (2017b). GC-MS analysis of aqueous extract of unripe fruit of *Carica papaya*. Journal Nutrition Food Sci. 7(3): 602-606.
45. Ezekwe, S.A., Chikezie, P.C. (2017a). GC-MS, hypoglycemic activity of aqueous extract of *Carica papaya* and its effect on blood lipid profile and hepatorenal tissues biomarkers of diabetic rats. Journal Diabetic Metabolism 8: 740. Doi: 10.4172/2156-6156.1000740.S.
46. Barathi, V., Anand, A.V., Anitha, P., Santhi, S. (2016). Chemical characterization of GC-MS

analysis of Tabernaemontana divaricata. Euro. J. Biomed. Pharm. Sciences 5(4): 451-454.

47. Orishapide, A.T., Ibekwe, N.N., Adesomoju, A.A., Okogun, J.I. (2012). Chemical composition and antibacterial activity of the seed oil extract of *Entandrophragma angolense (*Welw) C.D.C. African Journal of Pure and Applied Chemistry 6(13): 184-187.
48. Nazan, C., Yusuf, Z.K., Aygan, A. (2019). Investigation of bioactive compounds in relict endemic Ajuga relicta P. H. Davis (lamiaceae) from Turkey. Int. Journal Sec. Metabolite 6(3): 223-232.
49. Barwal, M. K., Ram, C., Gurjar, P. S…Kumar, P. (2022). The bioactive compounds and fatty acid profile of bitter apple seed oil obtained in hot, arid environments. Horticulturea DOI: 10.3390/horticurea8030259.
50. Rajeswari, G. Murugan, M., and Moghan, V. R. (2012). GC-MS analysis of bioactive components of *Hugonia mystax* L. (Linaceae). Research Journal of Phamacy Biology and Chemica*l* Science 3(4): 301-308.
51. Dayrit, F.M. (2014). The properties of Lauric acid and their significance in coconut oil. Journal of the American Oil Chemists' Society 92: 001-015.
52. De Roos, N.M., Schouten, E.G., Katan, M.B. (2001). Consumption of solid fat rich in lauric acid results in a more favourable serum lipid profile in healthy men and women than consumption of a solid fat rich in trans-fatty acids. The Journal of Nutrition 131(2): 242-245. Doi: 10.1093/jn/131.2.242.
53. Dayrit, F.M., Newport, M.T. (2020). The potential of coconut oil and its derivatives as effective and safe antiviral agent against the novel coronavirus (n-CoV-2019). icp.org.ph/2020/01/the-… Accessed June 13, 2021.
54. Nakatsuji, T., Kao, M.C., Fang, J-Y., Zouboulis, C.C., Zhang, L., Galo, R.L. Huang, C-M. (2009). Antimicrobial property of lauric acid against propionibacterrium acnes: Its therapeutic potential for inflammatory acnes vulgaris. Journal Invest. Dermatol. 129(10): 2480-2488.Doi: 10.1038/jid.2009.93.
55. Matsue, M., Mori, Y., Nagase, S., Sugiyama, Y., Hirano, R., Ogai, K., Ogura, K., Kurihara, S., Okamoto, S. (2019). Measuring the antimicrobial activity of lauric acid against various bacteria and human gut microbiota using new method. Cell Transplant 28(12): 1528-1541. Doi: 10.1177/0963689719881366.
56. Ugbogu, O.C., Onyeagba, R.A., Chigbu, A.O. (2006). Lauric acid content and inhibitory effects of palm kernel oil on two bacterial and *Candida albicans.* African Journal of Biotechnology 5(11): 1045-1047.
57. Nitibani, F.E., Siswanta, J.D., Solikhah, E.N. (2016). Isolation and antibacterial activity test of lauric acid from crude coconut oil (*Cocos nucifer*a L). Procedia Chemistry 18: 132-140.
58. Walters, D.R., Walker, R.L., Walker, K.C. (2003). Lauric acid exhibits antifungal activity against plant pathogenic fungi. J. Phytophathol. 151: 228-230.
59. Lifongo, L.L., Simoben, C.V., Ntie-Nkang, F., Babiaka, S.B., Judson, P.N. (2014). A bioactivity versus ethnobotanical survey of medicinal from Niger, West Africa. Natural Products Bioprospecting 4(1): 001-019.
60. Okwu, D.E., Ighodaro, B.U. (2010). GC-MS analysis of bioactive compounds and antibacterial activity of the oil fraction from the leaf of *Alstonia boonei* De Wild. Der Pharma Chemica 2(10: 261-272.
61. Huang, C.Y., Hong, P., Zhang, Z.Y., Song, J. (2010). Evaluation of antioxidant and anti-tumor activities of lemon essential oil. Journal of Medicinal Plant Research 4(18): 910-915.
62. Salamus, J., Baird, M.S. (1995). Biologically active cyclopropanes and cyclopropenes. Journal of Medicinal Chemistry 2: 511-542.

63. Anand, C.S., Mehendale, L.M. (2014). Dichloroacetic acid (DCA). Sciencedirect.com/topics/medicine-and-dentistry/dichloroacetic-acid… Accessed June 15, 2021.
64. Zaral, Z., Kadri, A., Choba, I.B., Mansour, R.B., Bekir, A., Mejdoub, H., Gharsallah, N. (2011). *In vitro* evaluation of antibacterial, antifungal, and cytotoxic properties of *Marrubium vulgare* L. essential oil grown in Tunisia. Lipids and Health 10: 161-168.
65. Akpuaka, A., Ekwenchi, M. M., Dashak, D. A., Dilda, A. (2013). Biological activities of characterized isolates of n-Hexane extract of *Azadirachta Indica* A. Juss (Neem) leaves. Nature and Science 11(5): 141-147.
66. Oshiobugie, M.J., Olaniyi, A.M., Rapael, A.O. (2017). AAS and GC-MS analysis of phytocomponents in the leaf, stem and root of *Azadirachta indica* A. Juss (Dongoyaro). British Journal Pharm. Research 15(4): 1-12.
67. Cherney, K., Kramer, O. (2019). Benzyol peroxide for acne: How to treat acne with benzyol peroxide. www.healthline.com/health/benzyol-peroxide-for-acne… Accessed June 5, 2021.
68. Matsugo, S. Mizuno, M., Konishi, T. (1985). Free radical generating and scavenging compounds as a new type of drugs. Current Medicinal Chemistry 2(4): 763-780.
69. Premjanu, N., Jaynthy, C. (2014). Antimicrobial activity of diethyl phthalate: An *insilico* approach. Asian Journal of Pharmacy and Clinical Research 7(4): 141-145.
70. Mustafa, M.F.M., Alamri, S.A., Taha, T.H, Alrumman, S.A. (2013). In vitro antifungal activity of Argemone ochrolueca Sweet latex against some pathogenic fungi. Afr. J. Biotechnol. 12(1): 1132-1137.
71. Mangamuri, U., Muvva, V., Poda, S., Naragani, K., Munaganti, R.K., Chitturi, B., Yenamanndra, V. (2016). Bioactive metabolites produced by *Streptomyces cheonanensis* VUK-A from Coringa mangrove sediments: isolation, structure elucidation and bioactivity. Biotechnology 6: 63 Doi: 10.1007/s13205-016-0398-6.
72. Ahsan, T., Chen, J., Zhao, X., Irfan, M., Wu, Y. (2017). Extraction and identification of bioactive compounds (eicosane and dibutyl phthalate) produced by Streptomyces strains KX852460 for the biological control of *Rhizoctonia solani* AG-3f strain KX852461to control target spot disease in tobacco. AMB Express 7(1): 54 Doi: 10.1186/s13568-017-0351-z.
73. Shobier, A.H., Ghani, S.A.A., Khouloud, M., Barakat, K.B. (2016). GC/MS spectroscopic approach and antifungal potential of bioactive extracts produced by marine macroalgae. Egyptian Journal of Aquatic Research 42: 289-299. Doi.org/10.1016/j.ejar.2016.07.003 1687.
74. Tataranni, T., Piccoli, C. (2019). Dichloroacetic acid and cancers? An overview towards clinical applications. Homeostasis and Cancer Article ID.8201079. Doi: 1155/2019/8201079.
75. Wang, H-L., Brattstrom, O., Brakefield, P.M., Franke, W., Lofstedt, C. (2014). Identification and synthesis of novel male specific esters in the wings of the tropical butterfly, *Bycyclus martius sanaos.* J. Chem. Ecology 40: 549-559.
76. Wang, H-L., Geertsema, H., van Nieukerken, E.J., Lofstedt, C. (2015). Identification of the female-produced sex pheromone… of the leafminer *Holocacista capensis* infesting grapevine in South Africa. J. Chem. Ecology 41: 724-731.
77. Poorima, K., Perumal, P.C. (2014): Protective effects of ethanolic extract of *Tabernaemontana divaricata (L.)R. Br.* against DEN and Fe NTA induced liver necrosis in wistar rats. Biomed Res. Intern.2014, 1-9.