



## Research Article

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## Chromatographic and Spectrometric Characterization of Bioactive Compounds from the Leaves of *Hyptis lanceolata* Poir

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### Abstract

The ethanolic extract of the leaves of *Hyptis lanceolata* was subjected to GC-MS analysis. Eight phytochemicals were identified which include imidodicarbonimidic diamide, N,N-dimethyl (5.37%), tetradecanoic acid (3.99%), 3-eicosyne (4.48%), hexadecanoic acid methyl ester (3.44%), hexadecanoic acid ethyl ester (21.87%), 2-hexadecen-1-ol, 3,7,11,15-tetramethyl (12.66%), 9,12,15-octadecatrienoic acid, ethyl ester (32.94%) and 9-octadecenoic acid, 12-hydroxy (15.24%). The use of *Hyptis lanceolata* in the treatment of skin diseases and other infections in herbal medicine in Nigeria could be as a result of the synergistic action of these phytochemicals. Also, the underutilization of the plant in phytomedicine was showcased as it could be used in the treatment of diabetes and tuberculosis as a result of its metformin and phytol contents respectively.

**Keywords:** *Hyptis lanceolata*, GC-MS, Skin infections, Herbal medicine, Underutilization.

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### 1. Introduction

Phytochemicals are plant naturally occurring chemicals. The use of plant chemicals in the treatment of infections dates back to several millennia ago. It is in fact the early use of these chemicals in phytomedicine that gave rise to the modern drugs that abound today. However, there is an increasing effort to harness the phytotherapeutic potentials of the rain forest vegetation of South Eastern Nigeria where *Hyptis lanceolata* Plant grows luxuriantly. *Hyptis lanceolata* Poir was selected for study because of its therapeutic properties in herbal medicine in Nigeria. The plant belongs to the family *Lamiaceae*. It is an erect, branching aromatic herb up to about 60 cm high, which reproduces from seeds [1]. The plant grows along roadsides and damp waste places throughout the region from Senegal to Fernando Po, and widely dispersed over the rest of Tropical Africa [2]. It also grows in America. The stem is 4-angled in cross-section, often many-branched and is sparsely covered with soft, short hairs while the leaves are opposite, *lanceolata*, about 7.5 cm long and 2.5 cm wide, are toothed at the margins, wedged-shaped at the base

and have very short petioles usually only about 0.5 cm long [1]. The leaves of *Hyptis lanceolata* Poir are used in the treatment of cutaneous and subcutaneous parasitic infections such as eczema, ringworm, rashes, athlete foot, etc [2]. They are also used as painkillers in herbal medicine in Nigeria. The leaf is used by the Ijo people of South Eastern Nigeria as a headache cure. The plant root is used in the treatment and or management of pulmonary troubles [2]. The paucity of documented information on the medicinal potentials and use of *Hyptis lanceolata* plant in herbal medicine prompted this research.

## 2. Materials and Methods

### Experimental

GC analyses were carried out in SHIMADZU JAPAN gas chromatography 5890-11 with a fused GC column (OV-101) coated with polymethyl silicon (0.25 mm × 50 m) and the conditions were as follows: temperature programming from 80-200 °C held at 80 °C for 1 minute, rate 5 °C/min and at 200 °C for 20 minutes, FID temperature 300 °C, injection temperature 250 °C, carrier gas nitrogen at a flow rate of 1 mL/min, split ratio 1: 75. GC-MS (Gas chromatography mass spectrometry) analysis was conducted using GCMS-QP 2010 Plus Shimadzu Japan with injector temperature of 230 °C and carrier gas pressure of 100 Kpa. 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 a mass spectrometer with a detector voltage set at 1.5 KV and sampling rate of 0.2 seconds. The mass spectrometer was also equipped with a computer fed mass spectra data bank. Hermle Z 233 M-Z centrifuge Germany was used. Solvents were all of analytical grade and were procured from Merck, Germany.

### Plant Materials

*Hyptis lanceolata* leaves were harvested from an abandoned fallowed farm land located at Ubakala, Umuahia South Local Government Area of Abia State, Nigeria. The leaves were then dried on the laboratory bench for 30 days and thereafter milled into a uniform and fine powder by a mechanically driven attrition mill.

### Extraction of Plant Materials

The powdered plant sample (300 g) was successfully extracted with 2 L of ethanol (8hrs/3 times/60 °C). The extract was concentrated under reduced pressure and the supernatant extract was decanted (5.39 g) after complete removal of the solvent. The extract was centrifuged at 10,000 rpm for 20 minutes and the clear supernatant extract was subjected to systematic GC-MS analysis.

### Components Identification

The components of the extracts were identified by matching the peaks with computer Wiley MS libraries and confirmed by comparing mass spectra of the peaks and those from literature [3-4].

## 3. Results and Discussion

The ethanol extract of *Hyptis lanceolata* leaves showed eight peaks from the chromatogram of the extract (Fig. 1). These peaks indicated the presence of eight compounds (1-8) in the extract (Figs. 2 and 3). The molecular formulae, percentage constituents and molecular masses of the compounds are shown in Table 1. These compounds comprise mainly alkaloids (5.37%), fatty acids (19.23%), hydrocarbon (4.48%), fatty acid esters (58.25%) and alcohols (12.66%). Compound **1** was an alkaloid identified as imidodicarbonimidic diamide, N,N-dimethyl and has molecular formula of  $C_4H_{11}N_5$  (m/z 129) with base peak at m/z 86 which resulted because of the cleavage of  $C_4H_9N_4$  group from the compound. The compound comprised 5.37% of the extract. Compound **2** was a fatty acid named tetradecanoic acid. It has molecular formula of  $C_{14}H_{28}O_2$  (m/z 228) and base peak at m/z 73 which was due to the detachment of  $C_3H_5O_2$  group from the compound.

The compound comprised 3.99% of the extract. Compound **3** was a hydrocarbon molecule identified as 3-eicosyne with a molecular formula of  $C_{20}H_{38}$  (m/z 278) and a base peak at m/z 67 due to the loss of  $C_5H_7$  group from the compound. The compound comprised 4.48% of the extract. Compound **4** was identified as hexadecanoic acid, methyl ester with a molecular formula of  $C_{17}H_{34}O_2$  (m/z 270) with a base peak at m/z 74 which was as a result of the cleavage of  $C_3H_6O_2$  group from the compound.

The compound comprised 3.44% of the extract. Compound **5** was named hexadecanoic acid, ethyl ester with molecular formula of  $C_{18}H_{36}O_2$  (m/z 284) and base peak at m/z 88 due to the detachment of  $C_4H_8O_2$  group from the compound. The composition of the compound in the extract was 21.87%. Compound **6** was identified as 2-hexadecen-1-ol, 3, 7, 11, 15-tetramethyl with molecular formula of  $C_{20}H_{40}O$  (m/z 278) and a base peak at m/z 71 which was as a result of the loss  $C_4H_7O$  group from the compound. The compound comprised 12.66% of the extract. Compound **7** was identified as 9,12,15-octadecatrienoic acid ethyl ester with molecular formula of  $C_{20}H_{34}O_2$  (m/z 306) and a base peak at m/z 79 due to loss of  $C_6H_8$  group from the compound. The compound comprised 32.94% of the extract. Compound **8** was identified as 12-hydroxy, 9-octadecenoic acid with molecular formula of  $C_{18}H_{34}O_3$  (m/z 298) and a base peak at m/z 55 due to  $C_3H_3O$  loss from the compound. The compound comprised 15.24% of the extract.

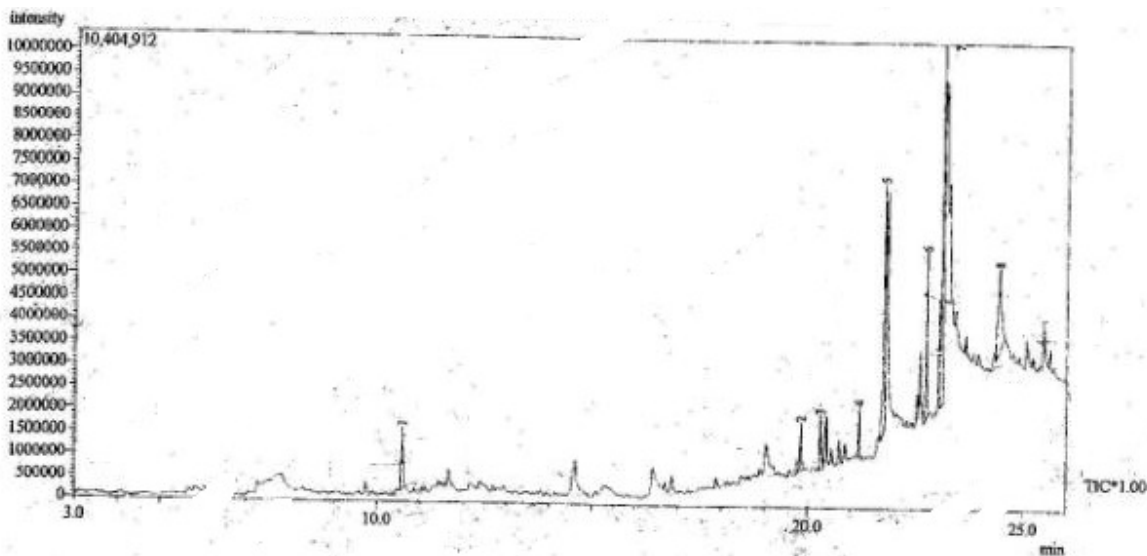


Fig. 1: GC-MS chromatogram of ethanolic extract of *Hyptis lanceolata*

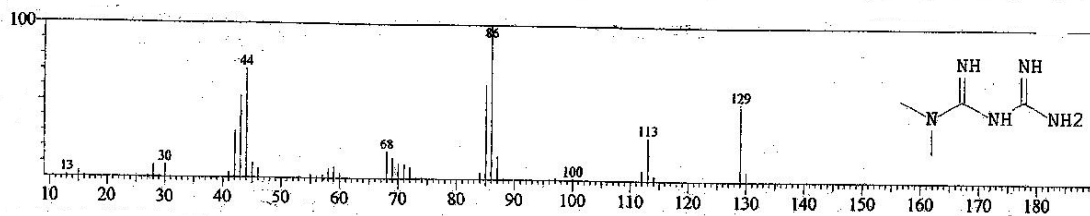


Fig. 2a: Imidodicarbonimidic diamide, N,N-dimethyl

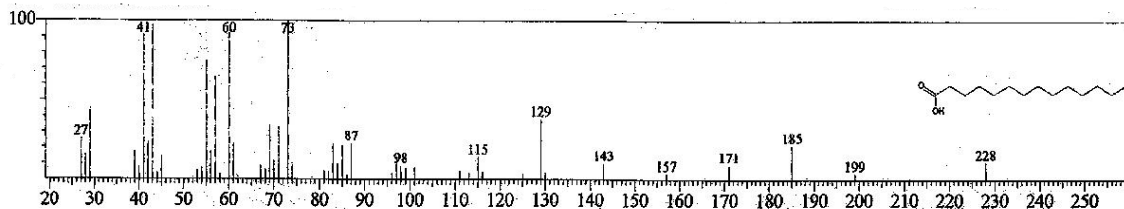


Fig. 2b: Tetradecanoic acid

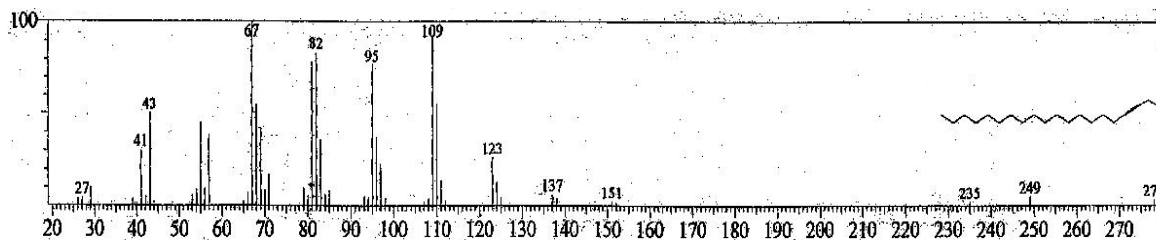


Fig. 2c: 3-Eicosyne

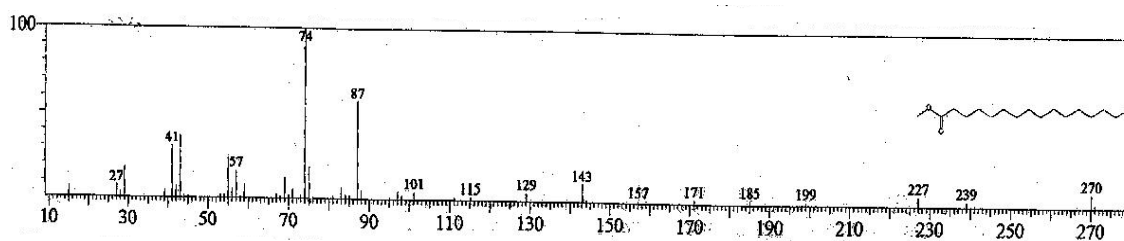


Fig. 2d: Hexadecanoic acid, methyl ester

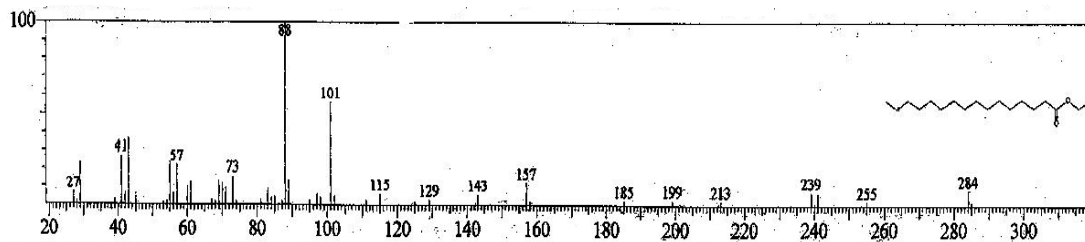


Fig. 2e: Hexadecanoic acid ethyl ester

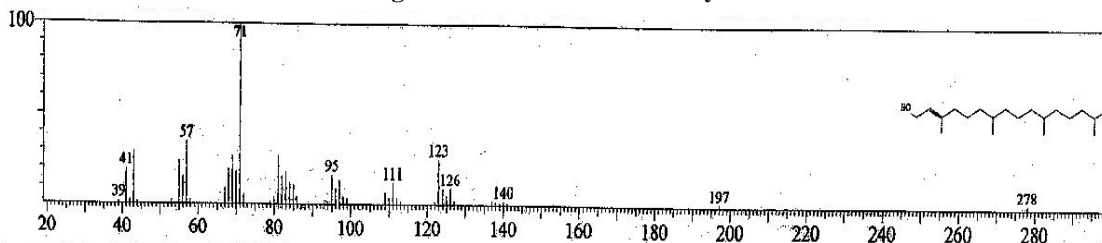


Fig. 2f: 2-Hexadecen-1-ol,3,7,11,15-tetramethyl

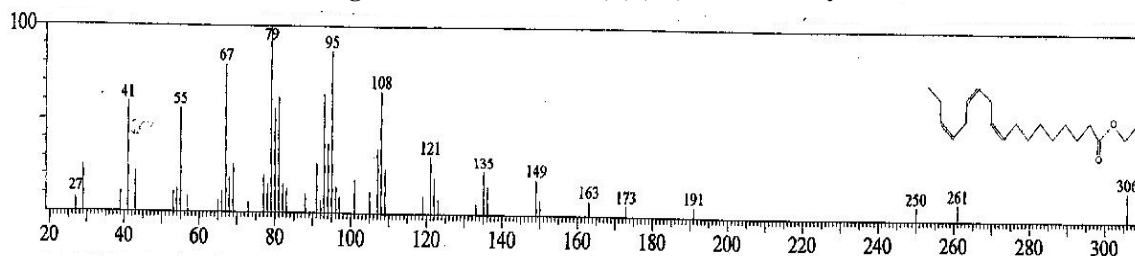


Fig. 2g: 9,12,15-Octadecatrienoic acid, ethyl ester

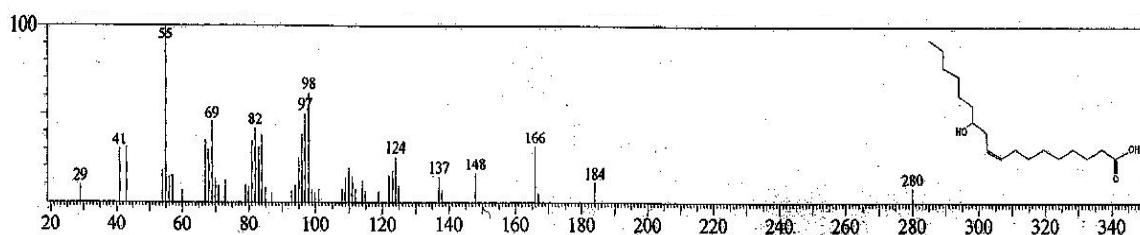


Fig. 2h: 9-Octadecenoic acid, 12-hydroxy

Table 1: Phytochemicals identified in the ethanolic leaf extract of *Hyptis lanceolata* by GC-MS

Chromatogram peak	Compound name	Molecular formula	Molecular weight	Retention time(min)	Peak area(%)	Nature of compound
1	Imidodicarbonimidic diamide, N,N-dimethyl	C <sub>4</sub> H <sub>11</sub> N <sub>5</sub>	129	10.575	5.37	Alkaloid
2	Tetradecanoic acid	C <sub>14</sub> H <sub>28</sub> O <sub>2</sub>	228	19.850	3.99	Fatty acid
3	3-Eicosyne	C <sub>20</sub> H <sub>38</sub>	278	20.292	4.48	Hydrocarbon
4	Hexadecanoic acid, methyl ester	C <sub>17</sub> H <sub>34</sub> O <sub>2</sub>	270	21.183	3.44	Fatty acid ester
5	Hexadecanoic acid ethyl ester	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	21.750	21.87	Fatty acid ester
6	2-Hexadecen-1-ol,3,7,11,15-tetramethyl	C <sub>20</sub> H <sub>40</sub> O	278	22.733	12.66	Alcohol
7	9,12,15-Octadecatrienoic acid, ethyl ester	C <sub>20</sub> H <sub>34</sub> O <sub>2</sub>	306	23.092	32.94	Fatty acid ester
8	9-Octadecenoic acid, 12-hydroxy	C <sub>18</sub> H <sub>34</sub> O <sub>3</sub>	298	24.425	15.24	Fatty acid

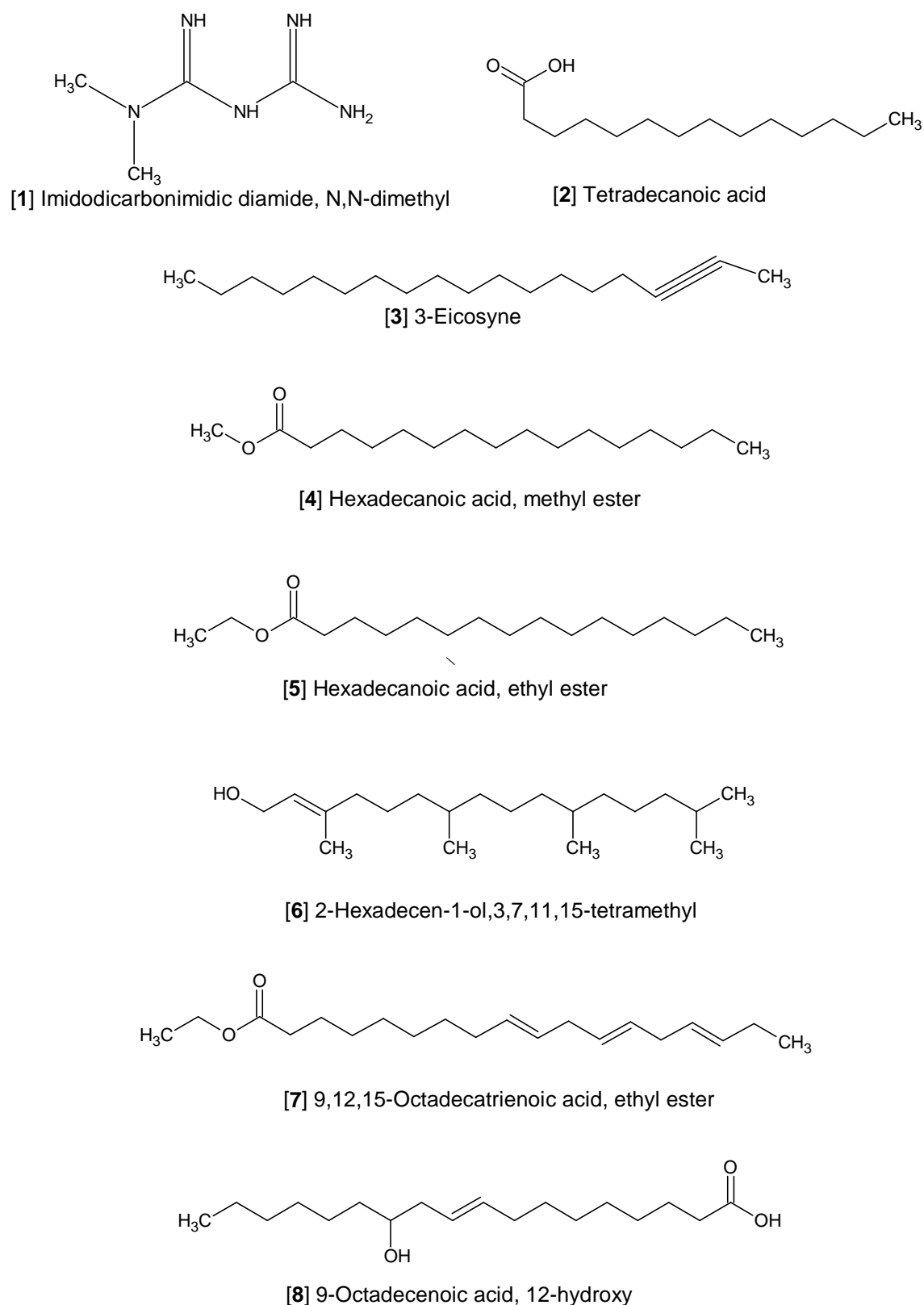


Figure 3: Structures of the phytochemicals from the ethanol leaf extract of *Hyptis lanceolata* Poir

Compound 1 which is also called metformin originally sold as glucophage is an oral antidiabetic drug that belongs to the biguanide class. It is the first-line drug of choice for the treatment of type 2 diabetes, in particular, in

overweight and obese people and those with abnormal kidney function [5]. It is also used in the treatment of polycystic ovary syndrome (PCOS) [6] and has been investigated for other diseases where insulin resistance may be an important factor such as in non-alcoholic fatty liver disease (NAFLD) and premature puberty [7-8]. Metformin works by suppressing glucose production by the liver [8]. The detection of 5.37% of metformin in the leaves of *Hyptis lanceolata* strongly suggests the use of the leaves in the treatment of diabetes in herbal medicine. The underutilization of this plant in herbal medicine is hereby revealed.

Fatty acids and alcohols in plant undergoes esterification reaction to form esters which frequently exude out of plants as resins and may be used in treating wounds and skin infections [3,9]. Compound **2** is also known as myristic acid which is commonly added co-translationally to the penultimate, nitrogen-terminus, glycine in receptor-associated kinases to confer the membrane localization of the enzyme [10]. The acid has a sufficiently high hydrophobicity to become incorporated into the fatty acyl core of the phospholipid bilayer of the plasma membrane of the eukaryotic cell. In this way, myristic acid acts as a lipid anchor in bio-membranes and may be used in cosmetics [10].

Compound **6** also known as phytol is an acyclic diterpene alcohol that can be used as a precursor for the manufacture of synthetic forms of vitamin E and vitamin K1 [11]. Phytol is used in the fragrance industry and in cosmetics, shampoos, toilet soaps, household cleaners and detergents [12]. Phytol uses may also include increasing energy and fighting infection and are natural alternatives to use for hypertension and cancer [12]. Phytol has been reported to have anti-mycobacterial activity against mycobacterium tuberculosis [13]. This suggests that the plant could be used in the treatment of tuberculosis. Again, the underutilization of *Hyptis lanceolata* plant in herbal medicine is stressed. The composition of phytol in the extract was 12.66% which was relatively high.

Compound **8** also called ricinoleic acid appears to be the most important constituent of the plant. Ricinoleic acid is categorized as omega 9 fatty acid that is found to be lethal weapon against viruses and bacteria. It is extremely effective when it comes to restricting growth of harmful microbes [14]. Ricinoleic acid acts as a natural antiviral drug. It is anti-inflammatory in nature and has germicidal and antiviral effects, and could be used on burns and wounds to protect them from infections [15]. The germicidal, insecticidal and fungicidal properties of ricinoleic acid protect the scalp and hair from microbial and fungal infections, the two prime causes for hair loss. The composition of ricinoleic acid in the extract was 15.24% which was relatively high. The use of the extract from *Hyptis lanceolata* leaves in herbal medicine in Nigeria for the treatment of skin infections like ringworm, acne, skin moles, eczema, skin dryness, cracked ankles, skin inflammation, itching and skin irritation must be as a result of the presence of ricinoleic acid. Ricinoleic acid possesses antimicrobial properties which when applied to open wounds, can act as a disinfectant [15].

#### 4. Conclusion

As ricinoleic acid provides antibacterial, antiviral, antifungal and germicidal properties against skin infections, phytol adds fragrance and is also antimicrobial while myristic acid ensures ricinoleic acid anchors in bio-membranes of the skin. The fatty acids present in the extract help the skin to restore the natural moisture balance, thus preventing unnecessary dryness. The synergistic action of these bioactive molecules no doubt was responsible for the use of *Hyptis lanceolata* plant in the treatment of microbial skin infections in herbal medicine in Nigeria. The detection of metformin and phytol in the leaf extract of *Hyptis lanceolata* suggests that the plant could be used in the treatment of diabetes and tuberculosis. By this, the underutilization of the plant in herbal medicine is revealed. Topical application of the extract can work wonders to get rid of acne, eczema, ringworm, rashes, dryness, moles, warts, hair loss, itching, irritation, redness and skin inflammation. The extract is hereby recommended to be added in cosmetics and for further pharmaceutical studies.

#### 5. References

1. IO Akobundu; CW Agyakwa. A handbook of West Africa weeds, 2<sup>nd</sup> ed., International Institute of Tropical Agriculture, Ibadan, Nigeria, 1998; pp.276-277.
2. HM Burkill. The Useful Plants of West Tropical Africa, Royal Botanic Gardens, Kew, 1985; p 3.
3. OU Igwe; DE Okwu. International Journal of Chemical Sciences, **2013**,11, 357.
4. AJD Britto; PAS Selvakymari. *Journal of Medicinal and Aromatic Plant Science*, **2006**, 28, 578.
5. American Diabetes Association. Diabetes Care, **2009**, 32 suppl. 1, 513-61
6. JM Lord; IHK Flight; RJ Norman. *BMJ*, **2003**, 327, 951.
7. G Marchesini; M Brizi; G Bianchi; S Tomassetti; M Zoli; N Melchionda. *Lancet*, **2001**, 358, 893.
8. L Ibáñez; K Ong; C Valls; MV Marcos; DB Dunger; F Zegher. *Journal Clinical Endocrinol Metabolism*, **2006**, 9, 2888.
9. DE Okwu; BU Ighodara. (2009). *Der Pharma Chemica*, **2009**, 2, 261.
10. Lexicon of Lipid Nutrition (IUPAC Technical Report). Pure and Applied Chemistry, **2001**, 73,685.

11. AM Daines. (2003). *Current Organic Chemistry*, **2003**,7, 1625.
12. D McGinty. *Food and Chemical Toxicology*, **2010**, 48, 559.
13. MS Rajab. *Planta Med.* , **1998**, 64,2.
14. J Nicks. Castor Oil for Skin Care, <http://www.buzz.com/article>. **2012**.
15. Organic Information Services Pvt. Ltd., Organic Facts, <http://www.organicfacts.net>. **2013**.