

## Comparative Evaluation of Phytoconstituents By GC-MS Of Four Selected Herbal Drugs Used In Abakaliki Ebonyi State, Nigeria.

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**Abstract:** The present study deals with the investigation of bioactive components in four selected medicinal plants identified in Ebonyi State Nigeria using GC/MS. The fresh leaves of *Dodder esculenta*, *Vernonia cinerea*, *Erythrina senegalensis* and *Blighia unijugata* were collected and dried in Biotechnology laboratory, Ebonyi State University, Abakaliki at room temperature, for two weeks and the dried leaves were grinded into fine powder using mechanical blender and further sieved with 2mm size sieve. This was further extracted with methanol and used for phytochemical identification by GC-MS analysis. The result revealed the presence of the following phytochemicals: fifteen in *Erythrina senegalensis*, eleven in *Dodder esculenta*, eight in *Blighia unijugata*, and seven in *Vernonia cinerea* respectively. Thus, our results revealed that the selected plants possess important phytochemicals such as nonadecanoic acid hexadecanoic acid, n-pentylacetate, and methyl caprate. Medicinal use of these plants may be attributed to these bioactive components.

**Keywords:** *Blighia unijugata*, *Dodder esculenta*, *Erythrina senegalensis*, *Vernonia cinerea*, GC-MS analysis, medicinal plants, phytochemicals

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### I. Introduction

Plants are used medicinally in different countries and they are the source of many potent and powerful drugs. Plants have been an important source of medicine with qualities for thousands years. Normally on traditional remedies such as herbs for their history, they have been used as popular folk medicines [1]. Plants are capable of synthesizing low molecular weight organic compounds known as secondary metabolites, usually with a unique complex structure [1]. Many metabolites have been found to possess interesting biological activities such as bactericidal, fungicide, hepato-protective and muscle relaxant [2].

The plant *Dodder esculenta* is a perennial parasite herbs, commonly known as akashbella or amarbel, 'ogbaramborogwu yeleli' in Ezzamgbo. It is an unusual parasitic vine belongs to the convolvulaceae family. The parasitism of dodder is by rapping itself around the host plants after attaching to it [3]. If the host contains food beneficial to Dodder, it will produce the haustoria inserting themselves into vascular system of the host [4]. *Dodder esculenta* is a valuable medicinal herb. Stem of the plant is antibacterial and used externally to treat itch and internally in fever [5]. It is useful in the treatment of androgen induced alopecia [6]. It also gives anti-inflammatory and anti cancer activity [7]. The aqueous and alcoholic extract of *Dodder esculenta* has diuretic activity [8].

On the other hand, the plant *Vernonia cinerea* belonging to the family asteraceae is an annual plant widely distributed in most tropical and subtropical countries, and have long been used in traditional medicine to treat various types of diseases [9]. It is called 'ogwuoka' in Ezzamgbo in Ebonyi State. In recent years, the interest in the plant-based medicine had increased noticeably worldwide. *V. cinerea* have many therapeutic uses in the practice of traditional medicine. Every part of the plant can be used medicinally [10]. This herbs has been used to treat a number of disorder including inflammation, malaria, fever, worms, pain, diuresis, cancer, abortion and various gastro intestinal disorder [11]. The roots of the plant are used traditionally for the treatment of all types of eruptive boils and the juice is used for quicker healing of accidental wound and toxic viral fever [12]. The young leaves of this plant are used for the treatment of tonsillitis. The leaf juice extract is used to treat skin diseases and also for treating dysentery in children [12]. Beside these, the plant is used in smoking cessation, cough, Arthritis, Urinary calculi, and leprosy [13]. The plant possesses antimicrobial, antibacterial, antioxidant, antihelminthic, anti-inflammatory, analgesic, antipyretic, antispasmodic and antidiuretic properties [14]. Some of the phytochemicals compounds present are sterols, flavonoids, sesquiterpane lactones and a terpenoid, leupeol acetate which shows antihypoglycaemic and antulcer properties [15]. Leaves from this plant serve as vegetable and culinary herb in soup in traditional Nigerian homes, extracts of the plant are used as tonic, in the control of tick and treatment of constipation and hypertension [16].

Drugs derived from natural sources plays a significant role in the prevention and treatment of human diseases. In many developing countries, traditional medicine is one of the primary health care systems [17].

*Erythrina senegalensis* also known as “coral tree” is a thorny shrub or small tree with bright red flowers arranged in panicles at the ends of the branches [19]. *Erythrina senegalensis* are also widely used in the tropics and subtropics as street and parks trees, especially in drier areas [20]. The fruit pods are thin-walled, strongly curved or coiled, 8 - 15 cm long and its seeds are bright red and ovoid [21]. It is commonly called ‘ukwaka’ in Ezzangbo in Ohaukwu Local Government Area Ebonyi State Nigeria, grown in West Africa as an ornamental plant and one of the oldest known African medicinal plants [22].

*Blighia unijugata* is widespread in tropical Africa, extending from Guinea Bissau eastwards to Ethiopia and Kenya, and through DR Congo southwards to Angola, Zimbabwe and Mozambique and South Africa [23]. *Blighia unijugata* is a tree planted as shade tree in Nigeria [24]. It is commonly called ‘ukpocha’ in Ezzangbo, Ngbo, Izzi, Ezza in Abakaliki in Ebonyi State Nigeria. It is a tree, 3-18 metres high, flowers whitish; very fragrant; calyx-lobes about 1 mm long; petals 1-1.5 mm long; ripe fruits red or pinkish red with three (3) shining black seed, each with a yellow aril [25].

## II. Materials And Methods

### Biological materials

Fresh leaves of *Dodder esculenta* , *Vernonia cinerea* , *Erythrina senegalensis* and *Blighia unijugata* were collected from their natural habitat in Mgboagbaja Izzi L.G.A Ebonyi State of Nigeria and Amovu-Amike Ezzangbo in Ohaukwu L.G.A, Ebonyi State South east Nigeria respectively. They were authenticated and identified by Prof. S.E Okafor, a plant taxonomist of the Department of Applied Biology, Ebonyi State University, Abakaliki.



**Plate 1:** *Erythrina senegalensis*



**Plate 2:** *Blighia unijugata*



Plate 3: Vernonia cinera



Plate 4: Dodder esculanta

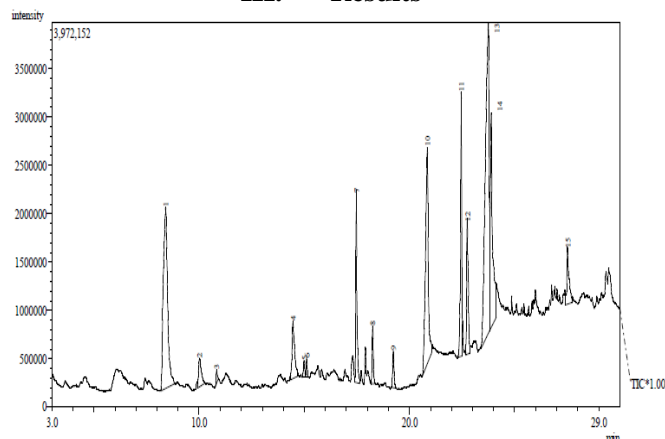
### Sample Preparation

The sample were washed, air-dried under room temperature in the laboratory and pulverized to powder in a mechanical grinder. This was sieved with 2 mm size sieve and packaged in an air tight glass and stored at 4°c until it was used. One hundred gram portion of the ground leave of *each* of the ground powdered plants were weighed, dissolved in 300 ml of methanol respectively. The system was allowed to stand for 48 hours and filtered with the white filter cloth. The filtrate was allowed to evaporate to dryness. The resulting extracts were scrept into sterile container covered, stored in the refrigerator and used for the analysis.

### GC-MS Analysis Method

Various phytoconstituents were tested for in the methanol extract of the plant. Methods employed were standard analytical GC/MS as outlined by [26].

## III. Results

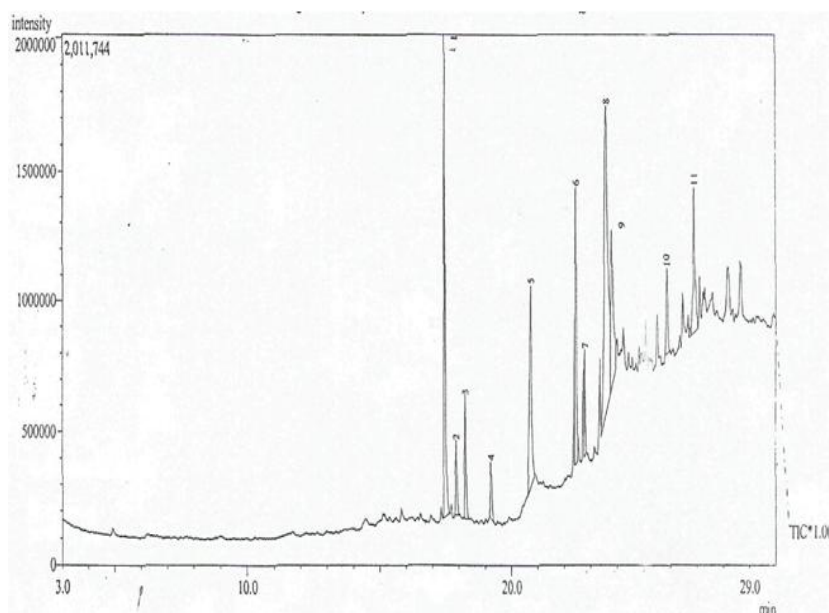


Legend: 1-15 are peaks of phytoconstituents

Plate 5: The Photo Microgram Of *Erythrina Senegenlensis*

**Table 1:** Result of Gas Chromatography Mass Spectrometry Contents of *Erythrina senegalensis*

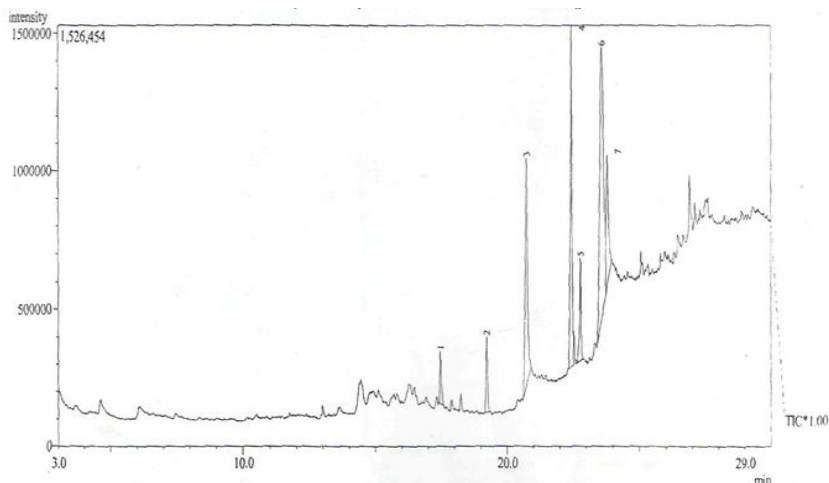
Peak	Compound	Molecular formular	Molecular weight	Retention time	% yield
1	n-pentylacetate	C <sub>7</sub> H <sub>14</sub> O <sub>2</sub>	130	8.405	15.19
2	Isopropyl butane	C <sub>9</sub> H <sub>14</sub> NO <sub>2</sub>	140	10.02	1.33
3	2- nitrobenzyl alcohol	C <sub>7</sub> H <sub>7</sub> NO <sub>3</sub>	153	10.82	0.47
4	1-(2- nitroa	C <sub>11</sub> H <sub>14</sub> N <sub>2</sub> O <sub>6</sub>	270	14.46	3.99
5	2-methylbutyl ester	C <sub>9</sub> H <sub>18</sub> O <sub>2</sub>	158	14.98	4.99
6	4-isopropenyl-1-methylcyclohexanol	C <sub>10</sub> H <sub>18</sub> O	154	15.11	0.66
7	Palmitaldehyde	C <sub>16</sub> H <sub>32</sub> O	240	17.47	10.64
8	1-nonyne	C <sub>9</sub> H <sub>16</sub>	124	18.25	1.33
9	pentadecanoic acid	C <sub>17</sub> H <sub>34</sub> O <sub>2</sub>	270	19.23	1.71
10	octadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	20.85	11.97
11	methyl 11-octadecenoate	C <sub>9</sub> H <sub>36</sub> O <sub>2</sub>	296	22.46	9.49
12	3,7,11,15-tetramethyl-2-hexadecen-1-ol	C <sub>20</sub> H <sub>40</sub> O	296	22.74	5.70
13	E-9-tetradecenal	C <sub>14</sub> H <sub>26</sub> O	210	23.75	17.09
14	n-nonadecanoic acid	C <sub>19</sub> H <sub>38</sub> O <sub>2</sub>	298	23.89	12.92
15	2,3-dihydroxypropyl ester	C <sub>19</sub> H <sub>38</sub> O <sub>4</sub>	330	27.51	6.84



**Legend:** 1-11 are peaks of phytoconstituents  
**Plate 6:** Photo microgram of *Dodder esculanta*

**Table 2:** Phyto Components Identified in *Dodder Esculenta*

Peak	Compounds	Molecular formular	Molecular weight	Retention time	% yield	Base peak
1	1-octadecyne	C <sub>18</sub> H <sub>34</sub>	250	17.47	18.6	43.00
2	2,6,10-trimethylundeca-1,3-diene	C <sub>14</sub> H <sub>26</sub>	194	17.9	2.76	66.05
3	2-tetradecyne	C <sub>14</sub> H <sub>26</sub>	194	18.25	4.1	57.05
4	Tridecanoic acid	C <sub>14</sub> H <sub>28</sub> O <sub>2</sub>	228	19.22	2.4	74.05
5	Octadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	20.74	9.76	43.05
6	11-octadecanoic acid	C <sub>19</sub> H <sub>36</sub> O <sub>2</sub>	296	22.45	8.7	55.05
7	2-hexadecen-1-ol	C <sub>20</sub> H <sub>40</sub> O	296	22.78	4.97	71.05
8	Oleic acid	C <sub>18</sub> H <sub>34</sub> O <sub>2</sub>	282	23.58	18.8	55.05
9	Nonadecanoic acid	C <sub>19</sub> H <sub>38</sub> O <sub>2</sub>	298	23.80	7.92	43.05
10	9-decen-2-one	C <sub>11</sub> H <sub>18</sub> O	166	25.88	8.83	43.00
11	10-undecenal	C <sub>11</sub> H <sub>20</sub> O	168	26.91	13.3	55.05

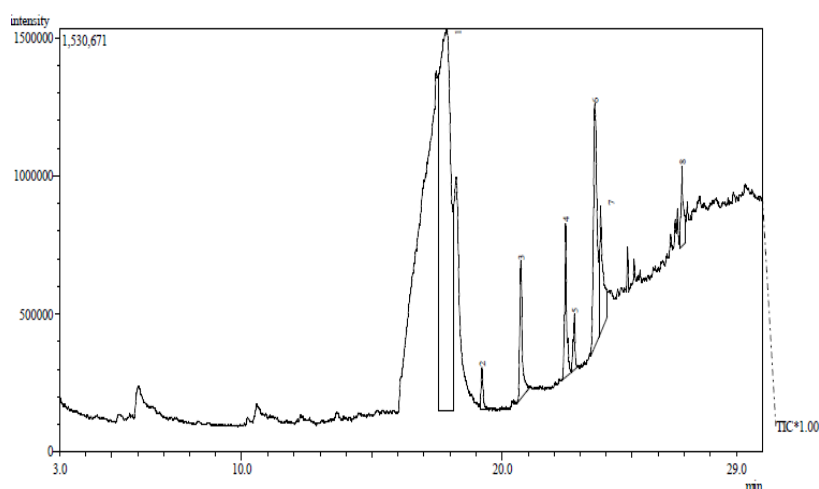


Legend: 1-7 The peaks of various phytoconstituents

Plate 7: Photo microgram of *Vernonia cinerea* indicating the various peaks representing bioactive components

Table 3: Phyto Components Identified In *Vernonia Cinerea*

Peak	Compounds	Molecular formular	Molecular weight	Retention time	% yield	Base peak
1	1-Octadecyne	C <sub>18</sub> H <sub>34</sub>	250	17.46	0.7	43.05
2	Pentanoic acid	C <sub>7</sub> H <sub>14</sub> O <sub>2</sub>	130	19.23	8.1	74.75
3	11-Octadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	20.75	20.0	43.05
4	6-octadecanoic acid	C <sub>19</sub> H <sub>36</sub> O <sub>2</sub>	296	22.45	20.2	55.05
5	Hexadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	22.79	8.7	74.05
6	4-Tridecene	C <sub>13</sub> H <sub>26</sub>	182	23.58	28.8	41.05
7	Octadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	23.81	13.8	43.05



Legend: 1-8 The peaks of phytoconstituents

Plate 8: The Photo Microgram of *Blighia unijugata*

Table 4: The results of Gas Chromatography Mass Spectrometry Contents of *Blighia unijugata*

Peak	Compound	Molecular formular	Molecular weight	Retention time	% yield
1	3-methylmannoside	C <sub>7</sub> H <sub>14</sub> O <sub>6</sub>	194	17.85	44.09
2	Methyl 6-methyl heptanoate	C <sub>9</sub> H <sub>18</sub> O <sub>2</sub>	158	19.23	20.47
3	n-octadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	20.73	4.25
4	10-octadecenoic acid	C <sub>19</sub> H <sub>36</sub> O <sub>2</sub>	296	22.45	6.93
5	Methyl caprate	C <sub>11</sub> H <sub>22</sub> O <sub>2</sub>	186	22.79	3.31
6	11-hexadecenoic acid	C <sub>16</sub> H <sub>30</sub> O <sub>2</sub>	254	23.58	8.03
7	n-hexadecanoic acid	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	256	23.80	3.67
8	Pentanoic acid	C <sub>16</sub> H <sub>30</sub> O <sub>2</sub>	254	26.92	9.24

#### IV. Discussion

Medicinal plants have been used by human beings since ages in traditional medicine due to their therapeutic potential and the search on medicinal plants has led to the discovery of novel drug candidates used against diverse diseases [27]. According to [28], more than 80% of the world population relies on traditional medicine for their primary healthcare needs.

*Dodder esculenta*, *Vernonia cinerea*, *Blighia unijugata* and *Erithrina senegalensis* as sources of bioactive compounds continue to play a dominant role in the maintenance of human health. Reports available on green plants represent a reservoir of effective chemo-therapeutics, these are non-phytotoxic, more systemic and easily biodegradable [29]. Hence, a thorough validation of the herbal drugs has emerged as a new branch of science emphasizing and prioritizing the standardization of the natural drugs and products [30]. This is because several of the phytochemicals present in these plants have complementary and overlapping mechanisms of action [31].

The compounds identified were fifteen (15) in *Erythrina senegalensis* among the major compounds include: n-pentylacetate (15.19%), palmitaldehyde (10.64%), 2-methylbutyl ester (4.99%), 1-(2-nitromethyl (3.99%) are presented in (Plate 5 and Table 1). Literature has shown that these compounds have medicinal merits [32].

The GC-MS analysis of *Dodder esculenta* showed the presence of 11 compounds. The result revealed that 1-octadecyne (18.6%), oleic acid (18.8%), 10-undecenal (13.3%), and octadecanoic acid (9.76%) were found as the major components of *Dodder esculenta* (Plate 6 and Table 2). Among the identified phytochemicals, 1-octadecyne possesses antimicrobial and antifungal activity [33]. While, 10-undecenal also possesses antimicrobial, antifungal, anti-inflammatory and anticancer activity [34]. Oleic acid possesses anticancer activity, anti-tumoral effect as reported in clinical studies. Numerous studies have reported an inhibition in cell proliferation induced by oleic acid in different tumor cell lines [35]. Oleic acid could suppress the overexpression of cancerous genes (oncogenes), which play a key role in the etiology, invasion, progression and metastasis in several human cancers [36]. Furthermore, Octadecanoic acid which is regarded as linoleic acid in nature, possesses anti-inflammatory, nematocidal, insecticidal, hypocholesterolemic, cancer preventive, hepatoprotective, antiacne, antihistaminic and antiarthritic [37].

However, the GC-MS analysis of *Vernonia cinerea* leaf methanol extract identified the existence of seven different compounds; 4-tridecane (28.8%), 6-octadecanoic acid (20.2%), 11-octadecanoic acid (20.0%) and octadecanoic acid (13.8%) were found as the major components in methanol extract (Plate 7 and Table 3). However, 4-tridecane (28.8%) possesses analgesic activity, antibacterial and antiviral activity according to [38]. Whereas, 6-octadecanoic acid and 11-octadecanoic acid which are stearic acid has been reported to possess antibacterial, anti-oxidant, antitumor, cancer preventive, immunostimulant and chemo preventive [39].

Other compounds identified in *Vernonia cinerea* have been reported to possess anti-inflammatory, antioxidant, nematocidal, pesticide, lubricant, antiandrogenic and haemolytic activity [40]. Pentanoic acid (8.1%) according to [41], possesses antioxidant activity and anti-inflammatory.

Among the major constituents identified in *Blighia unijugata* includes: 3-methylmannoside (44.09%), methyl 6-methyl heptanoate (20.47%), pentanoic acid (9.24%), 10-octadecenoic acid (6.93%) (Plate 8 and Table 4). Reports have shown that 3-methylmannoside enhances innate immunity [42], by aiding the molecular mechanisms by which organisms defend themselves against parasites. In vertebrates, such defense mechanisms against microbes and parasites include anti-glycan antibodies as well as a vast array of innate immune receptors, many of these receptor systems are required to eliminate infectious organisms [43], and they do so by either promoting their direct killing or through uptake and signaling to bring additional cell-mediated effector mechanisms to eliminate the threat [44]. Methyl 6-methylheptanoate functions as a potent antifungal active against yeasts, mold and more active against gram positive bacteria, than gram-negative bacteria [45]. Whereas [46], reported that Pentanoic acid serves as antifungal and antimicrobial agent [47]. Furthermore, 10-Octadecenoic acid methyl ester enhances the immunity of hydroxy unsaturated fatty acid [48]. Other compounds identified in *Blighia unijugata* such as n-Hexadecanoic acid had properties which are essential in forming cosmetic products and even act as soap releasing agents [49].

It is also a saturated fatty acid and just like other fatty acids, it has antibacterial and antifungal properties [50]. Fatty acids can modulate immune responses by acting directly on T cells [51]. The dietary, conjugated linoleic acid exerts anti-inflammatory effect by decreasing production of the inflammatory mediators such as prostaglandin E<sub>2</sub>, IL-6, IL-1 $\beta$ , TNF $\alpha$ , and nitric oxide [52]. It also serves as antioxidant, hypocholesterolemic, nematocidal, anti-androgenic flavour, antimalarial, haemolytic, 5- $\alpha$  reductase inhibitor, potent antimicrobial agent, [53].

The study confirms the presence of bioactive components which are known to exhibit medicinal value coupled with physiological activities. Hence, it may be inferred that the results obtained suggest that the identified phytochemical compounds may be the bioactive constituents and these plants are proving to be an increasingly valuable reservoir of phytoconstituents of substantial medicinal merit. Therefore, the data generated

from these experimental has provided the chemical basis and confirm the folkloric wide use of this plant as therapeutic agent for treating various ailments. Hence, it offer a platform of using *Erithrina senegalensis*, *Dodder esculenta*, *Vernonia cinerea* and *Bliaghia unijugata* leaves as herbal alternative for various diseases. The study was set out to investigate and compare the medicinal value of four selected medicinal plants commonly used in Ebonyi State. Our results have shown that all the four plants have phytochemical components of medicinal value, ranging from *Blighia unijugata*, *Vernonia cinera*, *Erithrina senegalis* and *Dodder esculanta*. Hence, the use of these plants in this locality for treatment of diseases may be attributed to these bioactive components and should be encouraged.

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