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Original Research Article

Phytochemical Profiling of *Basella alba* Using Gas chromatography-Mass Spectrometry

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ABSTRACT

The therapeutic activities of medicinal plants have been associated with their constituent bioactive compounds called phytochemicals. The phytochemicals of methanol and aqueous leaf extracts of Basella alba were assessed using gas chromatography-mass spectrometry technique and pharmacological potential of the phytochemicals were elucidated. The results showed the presence of 8-Methyl-6-nonenoic acid, Valeric acid, but-3-yn-2-yl ester, 3-Decyne, 2-Butynedioic acid, Benzene, propyl-, 5-Ethyl-2-furaldehyde, 2-Pyrazoline, Furan, 3-methyl-, Caryophyllene oxide, Hexadecanoic acid methyl ester, 6-Octen-1-ol, 3,7-dimethyl- and trans-Geranylgeraniol in the methanol extract. Naphthalene, 1,2,3,4-tetrahydro-, Tetradecanoic acid, 1,5-Heptadiyne, 2-Pentadecanone, 6,10,14-trimethyl, 1,2Benzenedicarboxylic acid,1-Tetradecanol, Heptyl triacontyl ether, Pentadecanoic acid, n-Hexadecanoic acid, Cyclohexadecane, 9-Octadecenoic acid, Trichloroacetic acid, Cyclohexaneethanol, Heptadecanoic acid, 1-Octadecene, 1-Hexadecanol, and 1-Hexadecanol, 3,7,11,15-tetramethyl were identified in the aqueous extract. Some of the pharmacological properties of the identified phytochemicals include: antioxidant, anticancer, antimicrobial, antiproliferative, anti-inflammatory and neuroprotective effects. The findings derived from this study hereby elucidate the therapeutic value of the plant especially the aqueous extract which contained more phytochemicals with more prominent antioxidant effects. Hence, B. alba plant could serve as a veritable source of therapeutic agent for treatment and management of diseases.

Keywords: *Basella alba, Phytochemicals, Gas chromatography, Mass spectrometry*

Introduction

Medicinal plants are significant component of the natural plant biodiversity which are applied since antiquity for the purpose of management and treatment of different disease conditions.[1,2] The therapeutic activities of medicinal plants, which could be associated with some or the whole parts of the plants, are usually determined by the constituent bioactive compounds referred to as phytochemicals.[3,4] In other words, phytochemicals are secondary metabolites derived from the medicinal plants which can be applied for the treatment of pathologies based on their synergistic, additive or individual mechanisms of action.[4,5] Phytochemical screening of medicinal plants has identified constituents like flavonoids, tannins, saponins, alkaloids, terpenoids, steroids and phenols which are applied for diverse therapeutic activities.[4,6] Hence, phytochemical screening of medicinal plants is a significant phase of novel drug discovery and development from medicinal plants.[6]

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Basella alba is a member of the *Basellaceae* family which is commonly cultivated as vegetable in different parts of Asia and Africa. It is also called Malabar spinach, Indian spinach, Ceylon spinach, climber spinach and vine spinach.[7] The local names of the plant include: Lu luo kui (China), Pazu (Turkey), Mong toi (Vietnam), *Espinaca blanca de Malabar* (Spain), *Basella* (France), Spinacio della cina (Italy), indischer spinat (German), Alugbati (Philippines), and Amunututu (Yoruba, Nigeria).[8] It is a succulent, herbaceous, climbing, perennial vine which grows into several meters in length. The leaves are heart-shaped, about 5-12 cm long and tapers to a pointed tip; the stems are greenish or purplish while the fruits are purple in colour, spherical in shape, stalkless and fleshy.[8]

The various parts of the plant such as the leaves and stems have been reportedly applied for the treatment of diverse pathological conditions due to their potential pharmacological or biological activities which include: anti-ulcer, antioxidant, anti-inflammatory, anti-hypoglycemic, anti-proliferative, wound-healing, antimicrobial, and many more. Generally, these activities are dependent on the phytochemicals present in different parts of the plant which include: carotenoids, organic acids, flavonoids, betacyanin, and many more.[8] The identification of the phytochemical constituents of the study plant is required for the assessment of its pharmacological properties and application in treatment of diseases. Essentially, phytochemical analysis can be achieved through different methods including gas chromatography which provides qualitative as well as quantitative assessments especially when coupled with mass spectrometry.[9]

The aim of the current study was to characterize the phytochemical constituents of the methanol and aqueous extracts of *B. alba* using the gas chromatography-mass spectrometry (GC-MS) technique. The findings of this study will elaborate the pharmacological properties of the plant extracts on the basis of their constituent phytochemical compounds.

Materials and Methods

Collection, identification and preparation of plant material
Fresh sample of the *B. alba* plant was collected from the suburb of Ede town in Osun State, Nigeria (7^0 43′ 60.00″N and 4^0 25′ 60.00″E) during the month of April, year 2022 and authenticated by taxonomist at the Herbarium, Department of Botany, Obafemi Awolowo Univeristy, Ile-Ife, Nigeria (Voucher number: IFE-18294). Fresh stock of study plant was further collected and the leaves were detached, air-dried for fifteen (15) days at the ambient temperature (25^0C).

Preparation of methanol and aqueous extracts
The dried leaves were pulverized into powdered form with the aid of a mechanical grinder. Measured quantity (100g) of the pulverized plant material was each macerated in 1000 mL of methanol and distilled water respectively for seventy two (72) hours. The mixtures were filtered and evaporated to dryness with the aid of a rotary evaporator at 50^0C and 40 rpm. The residues derived were cooled, weighed to determine the percentage yield of each extract and preserved in a refrigerated chamber (4^0C) until further use.

Phytochemical profiling of the extracts using Gas Chromatography-Mass Spectrometer (GC-MS)
For each extract, 0.1g was dissolved in 1 ml of methanol and filtered. Furthermore, 1 µl of each filtrate was used for the phytochemical analysis using GC-MS (Agilent Technologies, United States) and following the procedure described by Omotoso and Eze.[10] The phytochemical constituents of each extract were detected via comparison of their mass spectra with the reference spectra in the National Institute of Standard and Technology (NIST) library database.

Results and Discussion

Plant extract yield
After the completion of the extraction process, the percentage yield for the methanol extract was 11.5% while that of the aqueous extract was 15%.

GC-MS phytochemical profiling of the methanol extract of B. alba
The gas chromatogram of the methanol extract of *B. alba* (Figure 1) showed the retention time (RT) and peak area (PA) for the twelve (12) phytochemicals identified in the extract during the GC-MS phytochemical analysis. In addition to the RT and PA, the molecular formula, molecular weight and class of the compounds are presented in Table 1 while the molecular structures of the phytochemicals are presented in Figure 3. The phytochemicals identified in the methanol extract of *B. alba* include: 8-Methyl-6-nonenoic acid, Valeric acid, but-3-yn-2-yl ester, 3-Decyne, 2-Butynedioic acid, Benzene, propyl-, 5-Ethyl-2-furaldehyde, 2-Pyrazoline, Furan, 3-methyl-, Caryophyllene oxide, Hexadecanoic acid methyl ester, 6-Octen-1-ol, 3,7-dimethyl- and trans-Geranylgeraniol.

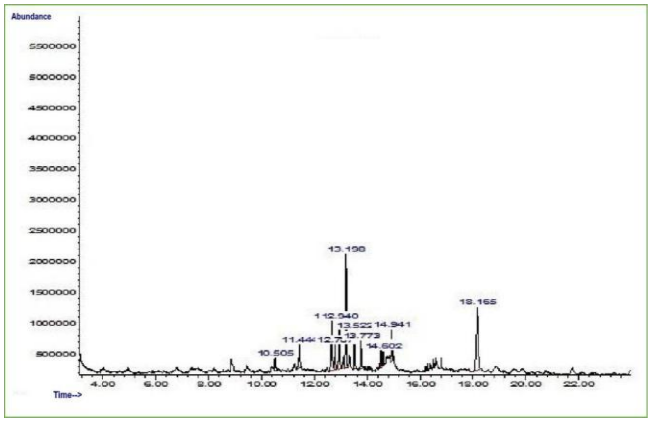

Figure 1. GC-MS chromatogram of methanol extract of *B. alba*

GC-MS phytochemical profiling of aqueous extract of B. alba
The gas chromatogram of the aqueous extract of *B. alba* (Figure 2) showed the retention time and the peak area for the seventeen (17) phytochemicals identified in the extract during the GC-MS phytochemical analysis. In addition to the RT and PA, the molecular formula, molecular weight and class of the compounds are presented in Table 2 while the molecular structures of the phytochemicals are presented in Figure 4. The compounds identified in the aqueous extract of *B. alba* include: Naphthalene, 1,2,3,4-tetrahydro-, Tetradecanoic acid, 1,5-Heptadiyne, 2-Pentadecanone, 6,10,14-trimethyl, 1,2Benzenedicarboxylic acid, 1-Tetradecanol, Heptyl triacontyl ether, Pentadecanoic acid, n-Hexadecanoic acid, Cyclohexadecane, 9-Octadecenoic acid, Trichloroacetic acid, Cyclohexane ethanol, Heptadecanoic acid, 1-Octadecene, 1-Hexadecanol, and 1-Hexadecanol, 3,7,11,15-tetramethyl.

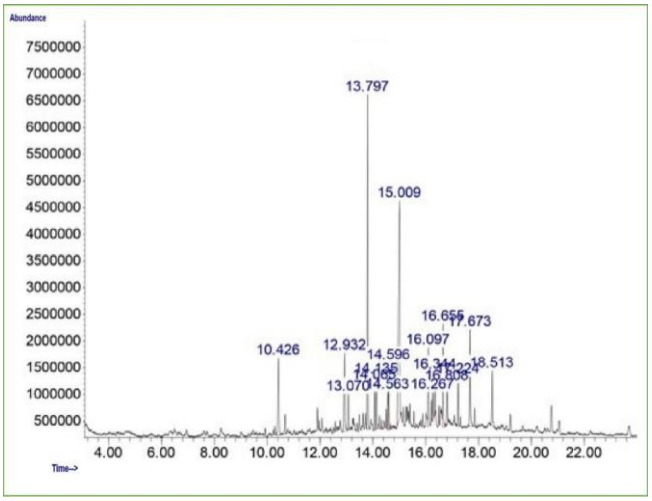

Figure 2. GC-MS chromatogram of aqueous extract of *B. alba*

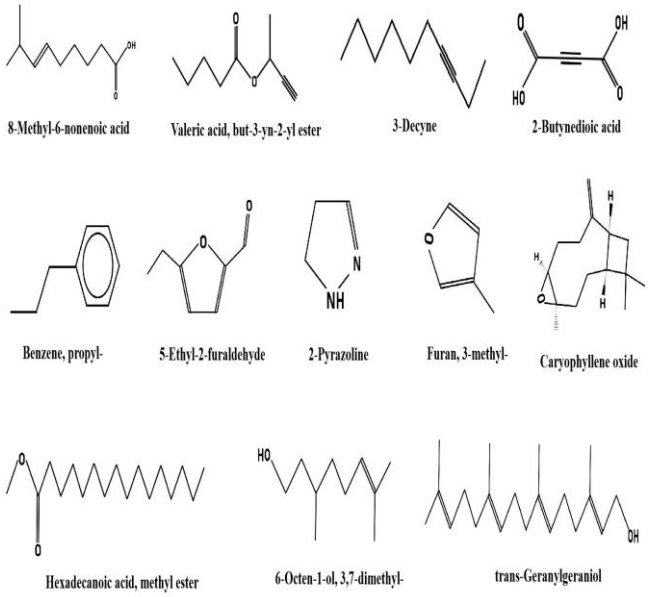

Figure 3. Molecular structures of GC-MS phytochemicals of methanol extract of *B. alba*

Phytochemicals have been regarded as the non-nutritive, bioactive compounds which are produced for protective and disease-preventive effects on the plants itself.[11] On the basis of their structures and properties, phytochemicals can be categorized into primary phytochemicals such as carbohydrates, lipids, proteins or amino acid and secondary phytochemicals like alkaloids, phenols, saponins,

glycosides, flavonoids, tannins, triterpenes, coumarins, carotenoids and other nitrogen-containing compounds.[12,13] Moreover, ethnopharmacological studies have revealed the therapeutic potential of phytochemicals derived from medicinal plants in biological systems which invariably confers on the plants their medicinal value.[14] As a result, the application of medicinal plants for therapeutic purposes has gained prominence in the past few decades especially due to their relatively easy accessibility and affordability as well as negligible side effects.[13]

One of the most important steps to determine the therapeutic efficacy of medicinal plants is the process of extraction of phytochemicals using selective solvent (either polar or non-polar) through various procedures such as: Decoction, infusion, maceration, percolation, sonication, Soxhlet extraction, microwave-assisted extraction, supercritical fluid extraction, and others.[12,15] The factors that determine method of extraction employed include: the type of plant material, experimental protocol, properties of solvent, and purposed used of extract. Furthermore, a wide range of phytochemicals have been identified from medicinal plants through different screening techniques with their biological or pharmacological activities (beneficial or adverse) elucidated.[10]

Table 1. GC-MS phytochemical profile of methanol extract of *B. alba*

Compounds	RT (Min)	PA (%)	Molecular formula	Molecular weight (g/mol)
8-Methyl-6-nonenoic acid	10.51	2.71	$C_{10}H_{18}O_2$	170.2
Valeric acid, but-3-yn-2-yl ester	11.45	5.35	$C_9H_{14}O_2$	154.2
3-Decyne	12.66	11.94	$C_{10}H_{18}$	138.2
2-Butynedioic acid	12.78	4.86	$C_4H_2O_4$	114.1
Benzene, propyl-	12.94	10.01	C_9H_{12}	120.2
5-Ethyl-2-furaldehyde	13.20	19.59	$C_7H_8O_2$	124.1
2-Pyrazoline	13.52	5.71	$C_3H_6N_2$	70.1
Furan, 3-methyl-	13.78	4.14	C_5H_6O	82.1
Caryophyllene oxide	14.51	3.00	$C_{15}H_{24}O$	220.4
Hexadecanoic acid methyl ester	14.60	1.96	$C_{17}H_{34}O_2$	270.5
6-Octen-1-ol, 3,7-dimethyl- or Citronellol	14.94	5.28	$C_{10}H_{20}O$	156.3
trans-Geranylgeraniol	18.17	25.45	$C_{20}H_{34}O$	290.5

Table 2. GC-MS phytochemical profile of aqueous extract of *B. alba*

Compounds	RT (Min)	PA (%)	Molecular formula	Molecular weight (g/mol)
Naphthalene, 1,2,3,4-tetrahydro-	10.42	7.04	$C_{10}H_{12}$	132.2
Tetradecanoic acid	12.93	6.96	$C_{14}H_{28}O_2$	228.4
1,5-Heptadiyne	13.07	2.22	C_7H_8	92.1
2-Pentadecanone, 6,10,14-trimethyl	13.80	17.07	$C_{18}H_{36}O$	268.5
1,2-Benzenedicarboxylic acid	14.07	2.34	$C_8H_6O_4$	166.1
1-Tetradecanol	14.13	2.76	$C_{14}H_{30}O$	214.4
Heptyl triacontyl ether	14.56	1.72	$C_{37}H_{76}O$	536.9
Pentadecanoic acid	14.59	3.46	$C_{15}H_{30}O_2$	242.4
n-Hexadecanoic acid	15.01	26.89	$C_6H_{32}O_2$	256.4
Cyclohexadecane	16.10	5.03	$C_{16}H_{32}$	224.4
9-Octadecenoic acid	16.27	1.50	$C_{18}H_{34}O_2$	282.5
Trichloroacetic acid	16.34	1.86	$C_2HCl_3O_2$	163.4
Cyclohexaneethanol	16.66	6.16	$C_8H_{16}O$	128.2
Heptadecanoic acid	16.81	2.46	$C_{17}H_{34}O_2$	270.5
1-Octadecene	17,2	2.76	$C_{18}H_{36}$	252.5
1-Hexadecanol	17.67	6.06	$C_{16}H_{34}O$	242.4
1-Hexadecanol, 3,7,11,15-tetramethyl	18.51	3.71	$C_{20}H_{42}O$	298.5

The GC-MS is a specialized technique that allows the identification and quantification of the phytochemicals of medicinal plant especially the non-polar compounds, fatty acids, essential oils and alkaloids.[16] Based on the GC-MS profiling of methanol and aqueous extracts of *B. alba* in the current study, the classes of phytochemicals identified in the extracts include: fatty acids, aromatic compounds, aliphatic compounds, monoterpene alcohol, isoprenoid, methyl esters, and heterocyclic

compounds. The phytochemicals in the methanol extracts include: 8-Methyl-6-nonenoic acid, Valeric acid, but-3-yn-2-yl ester, 3-Decyne, 2-Butynedioic acid, Benzene, propyl-, 5-Ethyl-2-furaldehyde, 2-Pyrazoline, Furan, 3-methyl-, Caryophyllene oxide, Hexadecanoic acid methyl ester, 6-Octen-1-ol, 3,7-dimethyl-, and trans-Geranylgeraniol. On the other hand, the phytochemicals in the aqueous extracts include: Naphthalene, 1,2,3,4-tetrahydro-, Tetradecanoic acid, 1,5-Heptadiyne, 2-Pentadecanone, 6,10,14-trimethyl, 1,2-Benzenedicarboxylic acid, 1-Tetradecanol, Heptyl triacontyl ether, Pentadecanoic acid, n-Hexadecanoic acid, Cyclohexadecane, 9-Octadecenoic acid, Trichloroacetic acid, Cyclohexaneethanol, Heptadecanoic acid, 1-Octadecene, 1-Hexadecanol, and 1-Hexadecanol, 3,7,11,15-tetramethyl.

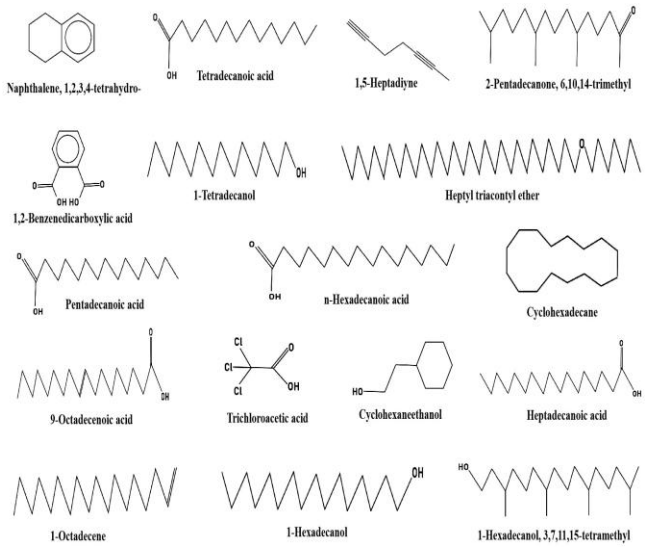

Figure 4. Molecular structures of GC-MS phytochemicals of aqueous extract of *B. alba*

Many of the phytochemicals identified in the extracts have been documented to exhibit diverse pharmacological or biological effects. Regarding the phytochemicals in the methanol extract, 8-Methyl-6-nonenoic acid exhibited antioxidant and anticancer effects; Valeric acid, but-3-yn-2-yl ester exhibited anticancer effect; 2-Butynedioic acid exhibited antimicrobial effect; 5-Ethyl-2-furaldehyde and Caryophyllene oxide exhibited anti-proliferative effect; 2-Pyrazoline exhibited anti-inflammatory and anticancer effects; Hexadecanoic acid methyl ester exhibited antimicrobial effect; and Geranylgeraniol exhibited anti-inflammatory, anti-proliferative, and neuroprotective effects.[17-24] Furthermore, regarding the phytochemicals in the aqueous extract, Naphthalene, 1,2,3,4-tetrahydro-, 1,2-Benzenedicarboxylic acid, and Pentadecanoic acid exhibited anticancer effect; Tetradecanoic acid exhibited antimicrobial effect; 1,5-Heptadiyne, 2-Pentadecanone 6,10,14-trimethyl, n-Hexadecanoic acid, 9-Octadecenoic acid, 1-Octadecene, 1-Hexadecanol, 1-Hexadecanol, 3,7,11,15-tetramethyl exhibited antioxidant effect; 1-Tetradecanol and Cyclohexaneethanol exhibited anti-inflammatory effect; Cyclohexadecane exhibited antimicrobial effect; Heptadecanoic acid exhibited anticancer effect.[16, 25-35] According to the findings of the current study, the phytochemical characterization of *B. alba* extracts revealed the therapeutic potential of the plant extracts especially due to the antioxidants property of some of the phytochemical constituents. The aqueous extract particularly revealed more phytochemical compounds with prominent antioxidant effect which would invariably attribute more therapeutic value on the extract. The findings of the current study thereby highlight the *B. alba* plant extract especially the aqueous extract as a veritable source of novel therapeutic drug for treatment and management of disease conditions.

Conclusion

The GC-MS phytochemical characterization of methanol and aqueous leaf extracts of *B. alba* revealed different phytochemical constituents that have prominent pharmacological activity especially due to the antioxidant effect. More phytochemical compounds were identified in the aqueous leaf extract with more prominent antioxidant effects, which would imply more therapeutic value. Therefore, the *B. alba* plant could be applied as a veritable source of therapeutic agent for the treatment and management of some diseases.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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