Journal of Coastal Life Medicine

journal homepage: www.jclmm.com

Original article doi: 10.12980/jclm.3.2015j5-135

Phytochemical screening and GC-MS determination of bioactive constituents from methanol leaf extract of *Senna occidentalis*

Aisha Mohammad Ibrahim[1*], Bashir Lawal[1], Ndababru Amos Tsado[2], Abubakar Awwal Yusuf[3], Adisa Mohammed Jimoh[4]

[1]*Department of Biochemistry, Tropical Disease Research Unit, Federal University of Technology, PMB 65, Minna, Nigeria*

[2]*Department of Basic and Applied Sciences, Niger State Polythechnic, Zungeru, Nigeria*

[3]*Department of Biochemistry, Ibrahim Badamasi Babangida University, PMB 11, Lapai, Niger State, Nigeria*

[4]*Department of Chemistry, Ibrahim Badamasi Babangida University, PMB 11, Lapai, Niger State, Nigeria*

ARTICLE INFO

Article history:
Received 3 Aug 2015
Received in revised form 14 Aug 2015
Accepted 25 Nov 2015
Available online 4 Dec 2015

Keywords:
Phytochemicals
GC-MS
Senna occidentalis
Bioactive constituent
Methanol extract

ABSTRACT

Objective: To identify the active ingredients presented in methanol extract of *Senna occidentalis* (*S. occidentalis*).
Methods: Dried powdered leaves of *S. occidentalis* were extracted with methanol by Soxhlet extraction and the extract was subjected to preliminary phytochemical screening by using standard procedure and methods. Gas chromatography-mass spectrometer (GC-MS) analysis was performed by comprising a GC-MS (model: QP2010 Plus Shimadzu, Japan) comprising an AOC-20i auto-sampler and gas-chromatograph interfaced to a mass spectrometer.
Results: The phytochemical study revealed the presence of tannins, alkaloids, glycoside, flavonoids, steroids, saponins, anthraquinones and phlobatannins while cardiac glycoside was not detected. GC-MS chromatogram showed nine peaks. A total of 31 compounds were identified when the mass spectra of the constituents was compared with the National Institute Standard and Technology library. The first compounds identified with less retention time (15.929 s) were n-hexadecanoic acid, octadecanoic acid and pentadecanoic acid while decanoic acid, decyl ester, ether, octadecyl vinyl, oleic acid, hexyl ester, stearic acid, octadecyl ester and decyl fluoride took the longest retention time (20.600 s) for identification.
Conclusions: The presence of these compounds in the plant extract may at least be responsible for one of the pharmacological properties of *S. occidentalis* and thus could be of considerable interest to the development of new drugs.

1. Introduction

Nature has presented to humanity the gift of biological and cultural diversity of natural products for healing practices. More than 80% of the world's population still rely primarily on their traditional natural products mainly from plants for their daily health care needs[1]. The use of plants as a source of medicine has been inherited and become an important component of the health care system. Despite the lack of adequate information on the components and the mode of action, herbal medicines of plants have been widely used to treat a vast array of clinical diseases. Studies have shown that commonly consumed medicinal plants are good sources of phytochemicals including polyphenols, saponins, flavonoids and phenylpropanoids[2]. These compounds display a vast variety of pharmacological activities such as anti-inflammatory, anticancer, anticarcinogenic, antibacterial, antioxidant, antifungal and antiviral activities[3].

Senna occidentalis (*S. occidentalis*) is highly reputed for its numerous medicinal uses and is known to be used ethnomedicinally as the remedy for several human and animal ailments. Recent studies show that *S. occidentalis* has anti-diabetic, anticancer and anti-ulcer effects and hepatoprotective

*Corresponding author: Aisha Mohammad Ibrahim, Department of Biochemistry, Tropical Disease Research Unit, Federal University of Technology, PMB 65, Minna, Nigeria.
Tel: +234-8039677503
E-mail: aishbm@gmail.com

and antitrypanosomal activity[4,5]. The stem bark and leaf extract of *S. occidentalis* have been found to contain important phytochemicals such as anthraquinones, carbohydrates, glycosides, cardiac glycosides, steroids, flavanoids, saponins, phytosterols, gum and mucilage[6]. Moreover, studies on this plant show that the nature and amount of the phytochemicals varies according to the season and geographical location[7].

A knowledge of the chemical constituents of this plant is desirable not only for the discovery of therapeutic agents, but also for the great value in disclosing new sources of bioactive principle for the synthesis of complex chemical substances and discovering the actual significance of folkloric remedies[8]. A literature survey revealed scanty information on the phytoconstituents, so the present study aims to identify the active ingredients presented in methanol extract of *S. occidentalis*.

2. Materials and methods

2.1. Plant sample

Fresh leaves of *S. ocidentalis* were obtained from Minna, Niger State, Nigeria. Taxonomic authentications of the plants were carried out in National Institute of Pharmaceutical Research and Development, Abuja, Nigeria.

2.2. Sample preparation and extraction procedure

The collected fresh leaves of *S. ocidentalis* were destalked, washed with clean water, dried at room temperature and finally ground by a grinder mill. The extraction of plant materials was performed by weighing 200 g of the powdered plants and extracted by Soxhlet extraction using 300 mL each of methanol. The marc was filtered with muslin cloth and solvents were removed under reduced pressure in a rotary evaporator. Green coloured pastes were obtained, weighed and stored in a refrigerator at 4 °C until they were required.

2.3. Gas chromatography-mass spectrometer (GC-MS) analysis

The GC-MS analysis of methanol leaves of *S. ocidentalis* was performed by using GC-MS Clarus 500 Perkin-Elmer system comprising an AOC-20i auto-sampler. The instrument is equipped with a VF-5ms fused silica capillary column with 30 m of length, 0.25 mm of diameter and 0.25 μm of film thickness. The temperatures employed were: column oven temperature 80 °C, injection temperature 250 °C at a pressure of 108.0 kPa with total flow and column flow of 6.20 mL/min and 1.58 mL/min respectively. The linear velocity was 46.3 cm/s and the purge flow was 3.0 mL/min. The GC program ion source and interface temperature were 200.00 °C and 250.00 °C respectively with solvent cut time of 2.50 min. The MS program starting time was 3.00 min which was ended at 30.00 min, with event time of 0.50 s, scan speed of 1 666 μL/s, scan range 40–800 μs and an injection volume of 1 μL of the extract (split ratio 10:1). The total running time of GC-MS was 30 min. The relative percentage of the extract was expressed as the percentage with peak area normalization as previously reported[8].

2.4. Identification of the components

Interpretation on the mass spectrum was conducted by using the database of National Institute Standard and Technology (NIST) which has more than 62 000 patterns. The fragmentation pattern spectra of the unknown components were compared with those of known components stored in the NIST library. The relative percentage amount of each biocomponent was calculated by comparing its average peak area to the total area. The name, molecular weight and structure of the components of the test materials were ascertained.

3. Results

Table 1 shows the phytochemical compositions of methanol leaves extract of *S. ocidentalis*. It revealed the presence of tannins, alkaloids, glycoside, flavonoids, steroids, saponins, anthraquinones and phlobatannins while cardiac glycoside was not detected. GC-MS chromatogram of methanol leaves of *S. ocidentalis* showed nine peaks (Figure 1). The active principle, area of peak concentration (%), retention time, molecular weight, and molecular formula in the methanol extract as identified through the NIST database were listed in Table 2.

Table 1
Phytochemical compositions of methanol leaves extract of *S. ocidentalis*.

Phytochemicals	Availability
Alkaloids	++
Flavonoids	++
Saponins	+
Steroids	+
Anthraquinone	+
Tannins	+++
Glycosides	++
Phlobatannins	+
Cardiac glycoside	-

-: Absent; +: Slightly present; ++: Significant; +++: Very significant. The classification was based on the extent of reaction during the quantitative study.

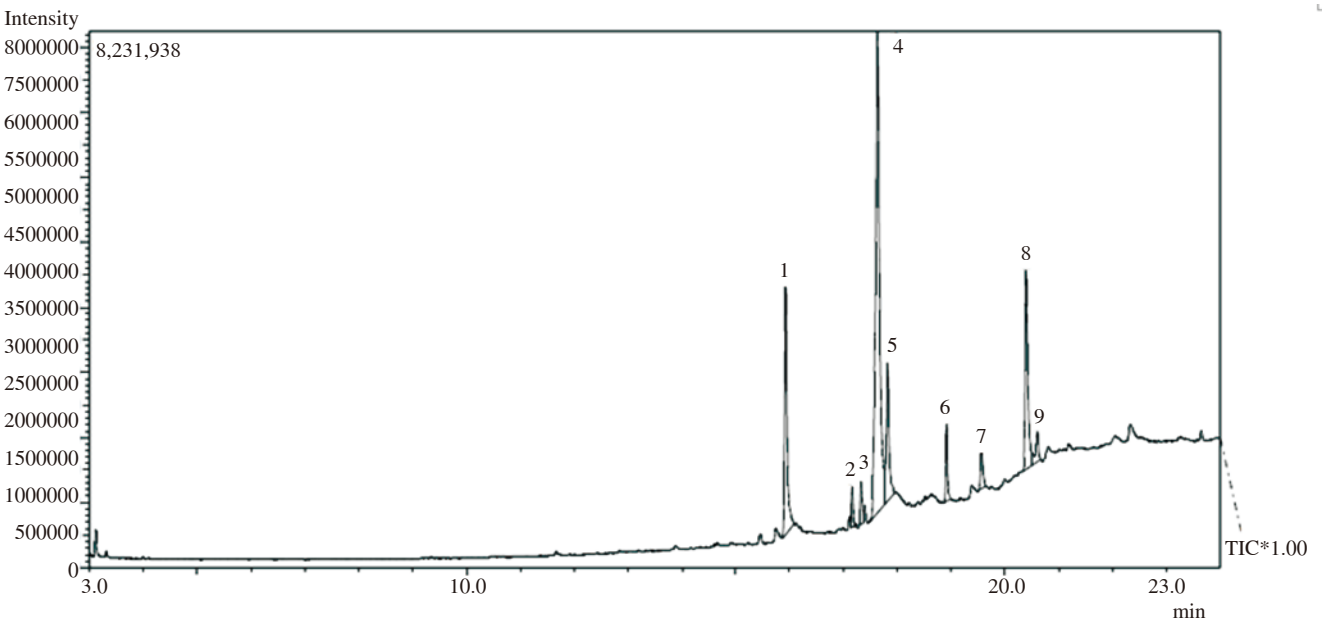

Figure 1. GC-MS chromatogram of methanol extract of *S. ocidentalis.*

Table 2
Bioactive components identified in methanol extract of *S. occidentalis* by GC-MS.

Peak No.	Retention time	Compound	Molecular formula	Molecular weight (g/mol)	Peak area (%)	Peak height (%)
1	15.929	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	15.94	19.00
1	15.929	Octadecanoic acid	$C_{18}H_{32}O_2$	284	15.94	19.00
1	15.929	Pentadecanoic acid	$C_{15}H_{30}O_2$	242	15.94	19.00
1	15.929	Heptadecanoic acid	$C_{17}H_{34}O_2$	270	15.94	19.00
2	17.165	Methyl cis-6-octadecenoate	$C_{19}H_{36}O_2$	296	1.69	3.14
2	17.165	Methyl 5-(2-undecylcyclopropyl)pentanoate	$C_{20}H_{38}O_2$	310	1.69	3.14
3	17.329	Phytol	$C_{20}H_{40}O_2$	296	1.80	3.28
3	17.329	Dihydrogeraniol	$C_{10}H_{20}O$	156	1.80	3.28
3	17.329	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	$C_{20}H_{40}O$	296	1.80	3.28
4	17.632	Oleic acid	$C_{18}H_{34}O_2$	282	48.76	37.03
4	17.632	9-Hexadecenoic acid	$C_{16}H_{30}O_2$	254	48.76	37.03
4	17.632	Erucic acid	$C_{22}H_{42}O_2$	338	48.76	37.03
4	17.632	Oxacyclotetradecan-2-one	$C_{13}H_{24}O_2$	212	48.76	37.03
4	17.632	Z-10-Pentadecen-1-ol	$C_{15}H_{30}O_2$	226	48.76	37.03
5	17.818	Octadecanoic acid, 2-(2-hydroxyethoxy)ethyl ester	$C_{22}H_{44}O_4$	372	9.58	10.63
6	18.916	2,3-Dihydroxypropyl palmitate	$C_{19}H_{38}O_4$	330	3.40	6.00
6	18.916	Palmitin, 1,2-di-, 2-aminoethyl hydrogen phosphate	$C_{37}H_{74}NO_8P$	691	3.40	6.00
6	18.916	Ether, octadecyl vinyl	$C_{20}H_{40}O$	296	3.40	6.00
6	18.916	Decyl fluoride	$C_{10}H_{21}F$	160	3.40	6.00
7	19.563	Eicosanoic acid	$C_{20}H_{40}O_2$	312	2.54	3.20
7	19.563	5,9-Dimethyl-1-decanol	$C_{12}H_{26}O_2$	186	2.54	3.20
8	20.386	1,6-Cyclodecadiene	$C_{10}H_{16}$	136	14.21	15.38
8	20.386	Cyclododecyne	$C_{12}H_{24}$	164	14.21	15.38
8	20.386	Tricyclo[4.2.1.1(2,5)]decane	$C_{10}H_{16}$	136	14.21	15.38
8	20.386	Cis-8-methyl-exo-tricyclo[5.2.1.0(2.6)]decane	$C_{11}H_{18}$	150	14.21	15.38
8	20.386	Cyclooctene, 3-vinyl-1-cyclooctene	$C_{10}H_{16}$	136	14.21	15.38
9	20.600	Decanoic acid, decyl ester	$C_{20}H_{40}O_2$	312	2.09	2.33
9	20.600	Ether, octadecyl vinyl	$C_{20}H_{40}O$	296	2.09	2.33
9	20.600	Oleic acid, hexyl ester	$C_{24}H_{46}O_2$	366	2.09	2.33
9	20.600	Stearic acid, octadecyl ester	$C_{26}H_{72}O_2$	536	2.09	2.33
9	20.600	Decyl fluoride	$C_{10}H_{21}F$	160	2.09	2.33

4. Discussion

Phytochemicals are secondary metabolites of plants. They are found in various parts of plants, where they elicit diverse roles like insect attraction for pollination, colour and vigour provision to plant while some act as feeding defence against predators. These phytochemicals, however, elicit varied medicinal and pharmacological actions when whey were ingested by animals[9].

The use of and search for plant derived drugs have accelerated in recent years. Biochemist, pharmacologists, botanists, microbiologists and natural products chemists in the whole world are continuously in search of medicinal plants for

bioactive phytoconstituents which could serve as a drug lead for treatment of various human ailment[10]. Hence, in the present study, preliminary phytochemical screening of methanol extract of *S. ocidentalis* was carried out. The results as seen in Table 1 suggested the phytochemical properties for curing various ailments and phytochemicals possess potential anti-inflammatory, antimicrobial and antioxidant, leading to the isolation of new and novel compounds. Since most alkaloids have a strong bitter taste and are very toxic, they are used by plants to protect themselves against herbivory and attack microbial pathogens and invertebrate pests[11]. Phenolics and flavonoids have been known for their anti-inflammatory, antioxidant, anticancer, antibacterial and antiviral properties[12]. Tannins are complex moieties produced by majority of plants as protective substances, and they have wide pharmacological activities. They have been used from the past as tanning agents and they possess astringent, anti-inflammatory, anti-diarrhoeal, antioxidant and antimicrobial activities[13]. Saponin and steroid also have relationships with sex hormones like oxytocin which regulates the onset of labour in pregnant women and subsequent release of milk[14]. The presence of important phytochemicals is an indication that if this plant is properly screened, it could yield a drug of pharmaceutical significance. However, the absence of cardiac glycoside was confirmed to early studies of Lawal *et al*. which also found that not all phytochemicals are present in all plants[9].

The GC-study led to the identification of number of compounds from the GC of methanol extract of *S. ocidentalis*. These compounds were identified through mass spectrometry attached with GC. The gas chromatogram shows the relative concentrations of various compounds getting eluted as a function of retention time. The large compound fragments into small compounds give rise to the appearance of peaks at different *m/z* ratios. These mass spectra are fingerprints of those compounds which can be identified from the data library[15]. When the mass spectra of the constituents from the extract were compared with the NIST library, a total of 31 different chemical compounds were characterized and identified. The first compounds identified with less retention time (15.929 s) were n-hexadecanoic acid, octadecanoic acid and pentadecanoic acid while decanoic acid, decyl ester, ether, octadecyl vinyl, oleic acid, hexyl ester, stearic acid, octadecyl ester and decyl fluoride took the longest retention time (20.600 s) to identify. Most of the compounds identified have been reported for anti-alopecic, anti-androgenic, antioxidant, haemolytic, hypercholesterolemic, lubricant nematicide, pesticide, propecia, flavour 5-α reductase inhibitor, anti-inflammatory and antiatherogenic[16].

The presence of these bioactive compounds in the plant extract may be responsible for one of the pharmacological properties of *S. occidentalis* and thus could be of considerable interest to the development of new drugs. A further research is, however, recommended to isolate, purify and characterize these chemical constituents.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgments

We acknowledge Adamu Mohammed of National Cereals Research Institute Zaria, Nigeria, for help during the GC-MS analysis.

References

[1] Bashir L, Shittu OK, Sani S, Busari MB, Adeniyi KA. African natural products with potential anti-trypanosoma properties: a review. *Int J Biochem Res Rev* 2015; **7**(2): 45-79.

[2] Loizzo MR, Saab AM, Statti GA, Menichini F. Composition and alpha-amylase inhibitory effect of essential oils from *Cedrus libani*. *Fitoteraptia* 2007; **78**(4): 323-6.

[3] World Health Organization. Traditional medicine growing needs and potential - WHO policy perspectives on medicines, No. 002, May 2002. Geneva: World Health Organization; 2002. [Online] Available from: http://apps.who.int/medicinedocs/en/d/Js2293e/ [Accessed on 26th July, 2015]

[4] Adamu M, Nwosu CO, Igbokwe IO. Toxicity and phytochemical constituents of aqueous extract of *Ocimum gratissimum* leaf. *Niger Vet J* 2008; **29**(3): 48-57.

[5] Basson W, Page ML, Myburgh DP. Human trypanosomiasis in southern Africa. *S Afr Med J* 1977; **51**(14): 453-7 .

[6] Colle F, Kaminsky R, Nkunya MH, Ogwal EN, Brun R. Trypanocidal activity of African plants. *J Ethnopharmacol* 2003; **5**: 1-11.

[7] Yadav JP, Arya V, Yadav S, Panghal M, Kumar S, Dhankhar S. *Cassia occidentalis* L.: a review on its ethnobotany, phytochemical and pharmacological process. *Fitoterapia* 2010; **81**(4): 223-30.

[8] Lawal B, Shittu OK, AbdulRasheed-Adeleke T, Ossai PC, Ibrahim AM. GC-MS determination of bioactive constituents of giant African snail (*Archachatina maginata*) haemolymph. *IOSR J Pharm Biol Sci* 2015; **10**(2): 59-64.

[9] Lawal B, Ossai PC, Shittu OK, Abubakar AN. Evaluation of phytochemicals, proximate, minerals and anti-nutritional compositions of yam peel, maize chaff and bean coat. *Int J Appl Biol Res* 2014; **6**(2): 21-37.

[10] Acharya D, Shrivastava A. *Indigenous herbal medicines: tribal formulations and traditional herbal practices.* Jaipur: Aavishkar Publishers and Distributors; 2008, p. 440.

[11] Harbone AJ. *Phytochemical methods: a guide to modern techniques of plant analysis.* London: Springer; 1998, p. 302.

[12] Manach C, Scalbert A, Morand C, Rémésy C, Jimenez L. Polyphenols: food sources and bioavailability. *Am J Clin Nutr* 2004; **79**(5): 727-47.

[13] Killedar SG, More HN. Estimation of tannins in different parts of *Memecylon umbellatum* Burm. *J Pharm Res* 2010; **3**(3): 554-6.

[14] Kalidass C, Mahapatra AK. Evaluation of the proximate and phytochemical compositions of an underexploited legume *Mucuna pruriens* var. *utilis* (Wall ex Wight) L. H. Bailey. *Int Food Res J* 2014; **21**(1): 303-8.

[15] Janakiraman N, Johnson M, Sahaya SS. GC-MS analysis of bioactive constituents of *Peristrophe bicalyculata* (Retz.) Nees. (Acanthaceae). *Asia Pac J Trop Biomed* 2012; **2**: S46-9.

[16] Duke J. Dr. Duke's phytochemical and ethnobotanical database; 2015. [Online] Available from: http://www.ars-grin.gov/duke/ [Accessed on 20th July, 2015]