# Comparison of the Chemical Compositions of Two Different Non-polar Extracts of *Heliotropium sudanicum* Aerial Parts from Sudan


## Abstract

**Aim:** The aim of this work is to provide answers to impending research questions on compounds present in petroleum ether and n-hexane extracts of the aerial parts of *H.sudanicum.*

**Methodology:** Phytochemical analysis of plant extracts was performed by using GC/MS . Various constituents were identified after matching their mass fragmentation pattern with data available in GC/MS library of National Institute of Standards Technology (NIST).

**Results:** Petroleum ether extract of *H. sudanicum* was characterized by abundance of 1,6,10,14,18,22- Tetracosahexaen-3-ol , 2,6,10,15,19,23-hexamethyl-, (all-E)- (33.99%) , Squalene (20.94%) , Phytol (7.62%) and Hexatriacontane (7.49%). The constituents of the extract were fatty acid derivatives (15.24%) ,hydrocarbons (8.41%) and terpenoids (28.56%). *H.sudanicum* n-hexane extract was characterized by abundance of 9,12-Octadecadienoic acid (Z,Z)-, methyl ester (15.14%) , 9,12,15-Octadecatrienoic acid, methyl ester (14.67%) , Cholesterol (11.12%) , Fucosterol (10.30% ) , . gamma.-Sitosterol (9.59%) and1,6,10,14,18,22- Tetracosahexaen-3-ol, 2,6,10,15,19,23-hexamethyl-, (all-E)- (7.73% ).

**Conclusion:** The resulted compounds in this study provide evidence for the utilization of the plant as alternative natural resource of medicinal and industrial interest. To best of our knowledge this is the first report that directly compares the two extraction solvents for active compounds from *Heliotropium sudanicum* aerial parts shows that petroleum ether should be the solvent of choice .



**Keywords:** *Heliotropium sudanicum*; Boraginaceae; aerial parts; chemical composition; non-polar solvents; Sudan


## 1.INTRODUCTION

The genus *Heliotropium* L. (Boraginaceae) with about 300 species all over the world. Is a genus of herbs rarely shrubs. It is used in folk medicine for the treatment of gout, rheumatism and as antiseptic and inflammatory agent. In Sudan, the whole herb is used for animal wound healing [1,2,3]. It is one of the main sources of pyrrolizidine alkaloids (PAs). PAs – containing plants have been recognized responsible for poisoning of

livestock and other ruminants [1,4-6] . *H. sudanicum* aerial parts was found by AL-Hassan et al.,[2] contains alkaloids, coumarins, flavonoids, tannins and steroids/triterpenes.

The *H. sudanicum* F.W.Andr. plant is a branched annual herb . Stems sulcate ,hispid . Leaves alternate , petiolate .sinuate , often of unequal base ,5 cm. long. Spikees often in pairs . Corolla white , twice as long as calyx, pubescent outside .Stigma conical , thickened at base .Nutlets glabrous [7]. In the stem transverse section . the anatomical structure of stem[6] were shows that the upper epidermis is a single layer of barrel-shaped dense cytoplasm with conspicuous nuclei, it is found as one layer. The presence of hairs and other epidermal outgrowths have been noted. The hypodermis consist of 2-3 layers of collenchymatous cells. The cells of general cortex consist of parenchymatous cells in 4 layers. While in the leaf transverse section, the anatomical structure of leaf[7] shows that the epidermal cells are rectangular and show the presence of an external cuticular layer. The upper epidermal cells are somewhat larger than the lower. Unicellular epiderma; trichomes have been noted on both surfaces. Stomata is of anomocytic type and are distributed on the upper and lower epidermis of the leaves. Palisade layers range from 2 to 4 . The main vascular bundle is 1 The lateral vascular bundle are 4 [8].

The aim of the research is to compare chemical analysis of petroleum ether and n-hexane extracts from *H.sudanicum* aerial parts through gas chromatography mass spectrometer (GC/MS).

## 2.MATERIALS AND METHODS

### 2.1 Plant Materials:

Aerial parts from Heliotropium sudanicum were collected in April 2017 from Shambat area (Khartoum State, Central Sudan). The studied species was taxonomically identified by Prof. M. Kordofani (Department of Botany, Khartoum University) and Ustaz. H.Tag EL-Sir (Department of Agricultural Botany, Khartoum University).

### 2.2 Sample preparation :

Each plant sample was carefully dried in the shade at room temperature. The aerial parts were then ground to fine powder with a mechanical crusher and the powder was kept in glass container at room temperature.

### 2.3 Extraction:

Petroleum ether and n-hexane were used as solvents of extraction. The powder was extracted separately with 250 ml petroleum ether and n-hexane at room temperature for

24 hours. After extraction it was filtered and then the removal of solvent was done by distillation device. Then the (70ml) extract was kept in glass container.

### 2.4 Gas Chromatography Mass Spectrometer (GC/MS) :

GC was equipped with MS ( Shimatzo QP 2010 GC/MS instrument equipped with reference libraries ).packed material for column were 50% phenyl and 50% methyl polysiloscane , column length 30 meter , diameter 0.025 mm, the flow rate of helium as carrying gas was 1 ml/min .the temperature of program consisted of 60-270 C ,at rate of 4 C/min .MS were taken at ionization voltage 70 EV. Library search was carried out using Wiley GC/MS library.

#### 2.4.1 Identification of components :

Interpretation on mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (NIST). The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, formula, molecular weight and area % of the components of the test materials were ascertained.

## 3. RESULTS AND DISCUSSION

### 3.1 GC/MS Analysis of *H. sudanicum* Petroleum Ether extract:

A total of 15 compounds representing 100% of the Petroleum Ether extract were identified and is given in the Table 1. Quantitatively, the extract was characterized by abundance of 1,6,10,14,18,22- Tetracosahexaen-3-ol , 2,6,10,15,19,23-hexamethyl-, (all-E)- (33.99%) , Squalene (20.94%) , Phytol (7.62%) and Hexatriacontane (7.49%).

The identified compounds comprised many secondary metabolites as hydrocarbons , terpenes, phytosterol, Phenolic compounds and the majority is the fatty acids derivatives. The major component was identified as a 1,6,10,14,18,22- Tetracosahexaen-3-ol, 2,6,10,15,19,23-hexamethyl-, (all-E)-( 33.99%). Many methyl esters of fatty acids are identified including Hexadecanoic acid,methyl ester, 9,12,15-Octadecatrienoic acid, methyl ester, 9-Octadecenoic acid (Z)-,methyl ester , Methyl stearate.

The fatty acid methyl ester 9-Octadecenoic acid (Z)-,methyl ester (5.42%) known also as Methyl oleate is used in Industry as Intermediates , lubricants and lubricant additives, Solvents (which become part of product formulation or mixture). and the consumer use it in ink, toner, colorant products lubricants , greases and water treatment products [9].

Many hydrocarbons are identified such as Heneicosane , Hexatriacontane and they are acyclic alkanes (aliphatic hydrocarbon). The detected Phenolic compound was Phenol, 2,4-bis(1,1-dimethylethyl)- ( 0.16%) it is very toxic to aquatic organisms, and uses as a

chemical intermediate for the synthesis of UV stabilizers or antioxidants and a chemical intermediate for the synthesis of other chemical intermediates [10]. And also the triterpenoid, Squalene was detected .

The constituents of the extract were fatty acid derivatives (15.24%) ,hydrocarbons (8.41%) and terpenoids (28.56%).

Table 1. chemical compounds from *H. sudanicum* petroleum ether extract

| **Sr. N0.** | **R.time** | **Name of the Compound** | **0ther Name** | **Molecular Formula** | **Molecular weight** | **Area %** |
|---|---|---|---|---|---|---|
| 1 | 3.184 | Hexane,2-nitro | - | $C_6H_{13}NO_2$ | 131.1729 | 3.81 |
| 2 | 11.194 | Phenol, 2,4-bis(1,1-dimethylethyl)- | 2,4-di-tert-butylphenol | $C_{14}H_{22}O$ | 206.33 | 0.16 |
| 3 | 14.865 | 2-pentadecanone, 6,10,14-trimethyl- | Perhydrofarnesyl acetone | $C_{18}H_{36}O$ | 268.4778 | 2.08 |
| 4 | 14.902 | 1-Hexadecanol,3,7,11,15-tetramethyl- | Dihydrophytol | $C_{20}H42O$ | 298.5469 | 1.72 |
| 5 | 15.672 | Hexadecanoic acid,methyl ester | Methyl palmitate | $C_{17}H_{34}O_2$ | 270.4507 | 2.69 |
| 6 | 17.324 | 9-Octadecenoic acid (Z)-,methyl ester | oleic acid ester | $C_{19}H_{36}O_2$ | 296.4879 | 5.42 |
| 7 | 17.380 | 9-Octadecenoic acid (Z)-,methyl ester | oleic acid ester | $C_{19}H_{36}O_2$ | 296.4879 | 1.87 |
| 8 | 17.394 | 9,12,15-Octadecatrienoic acid, methyl ester | methyl octadeca-9,12,15-trienoate | $C_{19}H_{32}O_2$ | 292.4562 | 4.45 |
| 9 | 17.493 | Phytol | 3,7,11,15-Tetramethyl-2-hexadecen-1-ol | $C_{20}H_{40}O$ | 296.5310 | 7.62 |
| 10 | 19.072 | Methyl stearate | methyl octadecanoate | $C_{19}H_{38}O_2$ | 298.511 | 0.81 |
| 11 | 19.072 | Heneicosane | - | $C_{21}H_{44}$ | 296.583 | 0.92 |
| 12 | 20.684 | Tetratriacontyl heptafluorobutyrate | - | $C_{38}H_{69}F_7O_2$ | 690.9421 | 5.95 |
| 13 | 23.207 | Squalene | - | $C_{30}H_{50}$ | 410.73 | 20.94 |
| 14 | 23.612 | Hexatriacontane | - | $C_{36}H_{74}$ | 506.988 | 7.49 |
| 15 | 23.953 | 1,6,10,14,18,22-Tetracosahexaen-3-ol, | (6E,10E,14E,18E)-2,6,10,15,19,23- | $C_{30}H_{50}O$ | 426.729 | 33.99 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | | 2,6,10,15,19,23-hexamethyl-, (all-E)- | hexamethyltetracosa -1,6,10,14,18,22- hexaen-3-ol | | | |
| | | | | | | 100.00 |

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110 **3.2 GC/MS Analysis of *H. sudanicum* n-hexane extract:**

111 A total of 20 compounds representing 100% of the Petroleum Ether extract were
112 identified and is given in the Table 2. Quantitatively, the extract was characterized by
113 abundance of 9,12-Octadecadienoic acid (Z,Z)-, methyl ester (15.14%) , 9,12,15-
114 Octadecatrienoic acid, methyl ester (14.67%) , Cholesterol (11.12%) , Fucosterol
115 (10.30% ) , . gamma.-Sitosterol (9.59%) , 1,6,10,14,18,22- Tetracosahexaen-3-ol,
116 2,6,10,15,19,23-hexamethyl-, (all-E)- (7.73% ) , .beta.- Sitosterol (6.66%) and
117 Hexadecanoic acid,methyl ester (5.05%).

118 The constituents of the extract were fatty acid derivatives (42.04%), phytosterols
119 (37.67%) and terpenoids (5.52%).The identified compounds comprised many secondary
120 metabolites as hydrocarbons , terpenes, phytosterol, vitamin and the majority is the fatty
121 acids and their derivatives. The major component was identified as 9,12-Octadecadienoic
122 acid (Z,Z)-, methyl ester (15.14%) the second major component is 9,12,15-
123 Octadecatrienoic acid, methyl ester and they are fatty acid methyl ester.

124 The identified methyl esters of fatty acids are the same as that detected in the petroleum
125 ether extract beside three different methyl esters , they are Methyl tetradecanoate,
126 Eicosanoic acid, methyl ester, and Docosanoic acid, methyl ester.

127 Many phytosterols (plant sterols) were detected and they are .gamma.-Sitosterol, .beta.-
128 Sitosterol, Cholesterol, Fucosterol. .beta.-Sitosterol and these may have the potential for
129 prevention and therapy for human cancer, used to make medicine, used for heart disease
130 and high cholesterol and the common cold and flu (influenza). Cholesterol it is the major
131 precursor for the synthesis of vitamin D of the various steroid hormones. One vitamin
132 was detected it is Vitamin E (alpha tocopherol), and it's has antioxidant activity, It may
133 also have anti-atherogenic, antithrombotic, anticoagulant, neuroprotective, antiviral,
134 immunomodulatory, cell membrane-stabilizing and antiproliferative actions.

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137 Table 2. chemical compounds from *H.sudanicum* n-hexane extract :

| Sr. N0. | R.time | Name of the Compound | 0ther Name | Molecular Formula | MW | Area % |
|---|---|---|---|---|---|---|
| 1 | 13.571 | Methyl tetradecanoate | Methyl myristate | $C_{15}H_{30}O_2$ | 242.403 | 0.22 |
| 2 | 14.864 | 2-pentadecanone, 6,10,14-trimethyl- | Perhydrofarnesyl acetone | $C_{18}H_{36}O$ | 268.4778 | 0.47 |
| 3 | 14.900 | 6,10,14- Trimethyl-pentadecan-2-ol | - | $C_{18}H_{38}O$ | 270.501 | 0.33 |
| 4 | 15.671 | Hexadecanoic acid,methyl ester | Methyl palmitate | $C_{17}H_{34}O_2$ | 270.4507 | 5.05 |
| 5 | 17.325 | 9,12-Octadecadienoic acid (Z,Z)-, methyl ester | Methyl linolate | $C_{19}H_{34}O_2$ | 294.4721 | 15.14 |
| 6 | 17.380 | 9-Octadecenoic acid (Z)-,methyl ester | oleic acid ester | $C_{19}H_{36}O_2$ | 296.4879 | 4.04 |
| 7 | 17.397 | 9,12,15-Octadecatrienoic acid, methyl ester | methyl octadeca-9,12,15-trienoate | $C_{19}H_{32}O_2$ | 292.4562 | 14.67 |
| 8 | 17.491 | Phytol | 3,7,11,15-Tetramethyl-2-hexadecen-1-ol | $C_{20}H_{40}O$ | 296.5310 | 1.20 |
| 9 | 17.584 | Methyl stearate | methyl octadecanoate | $C_{19}H_{38}O_2$ | 298.511 | 2.04 |
| 10 | 18.731 | .gamma.-Sitosterol | Clionasterol | $C_{29}H_{50}O$ | 414.7067 | 9.59 |
| 11 | 18.886 | .beta.- Sitosterol | - | $C_{29}H_{50}O$ | 414.7067 | 6.66 |
| 12 | 19.003 | Cholesterol | - | $C_{27}H_{46}O$ | 386.664 | 11.12 |
| 13 | 19.341 | Eicosanoic acid,methyl ester | Methyl eicosanoate | $C_{21}H_{42}O_2$ | 326.5570 | 0,45 |
| 14 | 19.939 | Fucosterol | - | $C_{29}H_{48}O$ | 412.702 | 10.30 |
| 15 | 20.963 | Docosanoic acid,methyl ester | Methyl Behenate | $C_{23}H_{46}O_2$ | 354.6101 | 0.88 |
| 16 | 21.087 | Stigmast-5-en-3-ol,oleate | | $C_{47}H_{82}O_2$ | 679.171 | 1.98 |
| 17 | 21.764 | Vitamin E | alpha tocopherol | $C_{29}H_{50}O_2$ | 430.717 | 2.51 |
| 18 | 23.202 | Squalene | - | $C_{30}H_{50}$ | 410.73 | 4.36 |
| 19 | 23.607 | Hexatriacontane | - | $C_{36}H_{74}$ | 506.988 | 1.27 |
| 20 | 23.949 | 1,6,10,14,18,22-Tetracosahexaen-3-ol, | (6E,10E,14E,18E)-2,6,10,15,19,23- | $C_{30}H_{50}O$ | 426.729 | 7.73 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | | 2,6,10,15,19,23-hexamethyl-, (all-E)- | hexamethyltetracosa -1,6,10,14,18,22- hexaen-3-ol | | | |
| | | | | | | 100.00 |

## CONCLUSION

To best of our knowledge this is the first report that directly compares the two extraction solvents for active compounds from *Heliotropium sudanicum* aerial parts shows that petroleum ether should be the solvent of choice . GC-MS analysis result of large Quantities of 1,6,10,14,18,22-tetracosahexaen-3-ol,2,6,15,19,23-hexamethyl-,(all-E), squalene, phytol and hexatriacontane were extracted by petroleum ether and weren't extracted by n-hexane. 9,12,15-octacatrienoic acid,methyl ester and hexadecanoic acid, methyl ester were found in n-hexane extract in a large quantity compared by the extraction quantity in petroleum ether extract.

**CONFLICTS OF INTEREST:** Authors have declared that no competing interests exist.

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