WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.084
Volume 12, Issue 4, 81-94. Research Article ISSN 2277– 7105

GC-MS ANALYSIS AND THE ESTABLISHMENT OF MASS SPECTROMETRIC FINGERPRINTS OF CHEMICAL CONSTITUENTS PRESENT IN ETHANOL EXTRACT OF *SECURIDACA LONGIPEDUNCULATA* LEAVES

Samuel E. Egga[1*], Mbanefo M. Ekwenchi[1], Adams Theophilus[1], Chukuka Achuenu[1,2], Solomon D. Lokta[1]

[1]Department of Chemistry, Faculty of Natural Sciences, University of Jos, Jos, Plateau State, P.M.B. 2084, Nigeria.

[2]Département de Chimie Moléculaire (SERCO), Université Grenoble Alpes, 301 Rue de la Chimie, BP 53, 38058 Grenoble Cedex 9, France.

Article Received on
03 Jan. 2023,

Revised on 23 Jan. 2023,
Accepted on 13 Feb. 2023

DOI: 10.20959/wjpr20234-27325

***Corresponding Author**
Samuel E. Egga
Department of Chemistry, Faculty of Natural Sciences, University of Jos, Jos, Plateau State, P.M.B. 2084, Nigeria.

ABSTRACT

The aim of the present work is to evaluate the ethanolic extract of *Securidaca longipedunculata* leaves for its phytoconstituents using GC-MS. The extraction was carried out by maceration, and then followed by subsequent characterization of the extract by gas chromatography-mass spectrometry (GC/MS). Results for the identification of various phytochemical components in the plant were based on their NIST scores (above 70%). The phytochemical constituents identified from the leaves extract are phenolics and phthalates, fatty acids derivatives, esters and vitamin E. Both qualitative and quantitative profiles of the phytoconstituents are presented with structures and their corresponding percentage compositions. The obtained data suggest that the plant could be considered a good source of valuable bioactive agents. The observed phytoconstituents among other secondary metabolites may be responsible for its antioxidant property and hence it may be useful in medicinal applications. The presence of these compounds is consistent with previous works. Further isolation and evaluation of the biological activity are required in order to utilize the medicinal potentials of the plant for applications in agrochemical, cosmetic, food and pharmaceutical industries.

KEYWORDS: GC-MS analysis, *Securidaca longipedunculata*, Constituents of ethanol extract of *Securidaca longipedunculata* leaves, Fatty acids.

INTRODUCTION

Human beings have been using different parts of plants extract as source of drugs since the prehistoric times and today they are still the major sources of new drugs.[1][2] Medicinal plants are of interest to man worldwide due to the presence of various bioactive compounds which possess biological activities such as anticancer, antibacterial, antifungal, anti-inflammatory and antioxidant.[3][4] Recently, there are several medicinal based industries due to the increase in the search and use of medicinal plants throughout the world.[5][6] This could be as a result of their availability, renewability, sustainability and biodegradability. Secondary metabolites are produced within plants, which are of great pharmacological importance having differences in their molecular structure and property. Crude extracts isolated from plant species can produce enough sources for new drugs.[7]

Securidaca longipedunculata belongs to the family of Polygalaceae, the plant species is called violet tree, fibre tree (South Africa) or Rhodesian violet (English), Sanya (Hausa), Ipeta (Yoruba).[8] It is a semi-deciduous shrub or medium size tree that grows up to 12m tall. It is resistant to bush fire and is sensitive to frost. It is mostly found in rocky wooded bush land in various tropical African countries, including Nigeria.[9][10] *S. longipedunculata* is a multi-purpose plant with a long history of use in African traditional medicine to treat various sexual infections, hernias, coughs, fever, constipation, headaches, rheumatism, stomach ache, malaria, tuberculosis, pain, epilepsy, pneumonia, skin infections, and it is also used as an aphrodisiac for men.[11] Extracts from root bark is used as pesticides against grain pest, for chest complaints, tuberculosis and toothache. The ethanol root extracts contain alkaloids, flavonoids, saponins, tannins, terpenoids and some steroids.[12][13] In spite of the promising therapeutic and medicinal applications of this plant, its phytoconstituents have not been well documented. And it has surprisingly received limited attention from scientific researchers in Nigeria. Therefore**,** in this study we will present the phytoconstituents profile found in the ethanol extract of *Securidaca longipedunculata* leaves by GC-MS technique. This could further reveal the potentials of this locally available plant for further industrial applications, as well as providing scientific information for researchers and traditional medicine practitioners.

Figure 1: ***Securidaca longipedunculata.***

MATERIALS AND METHODS

Collection and Identification of Plant Materials

Leaves of *Securidaca longipedunculata* plant were collected from Shere hills, Jos, Plateau state, Nigeria and were identified and authenticated by Mr. Joseph J. Azila of the Department of Forestry Technology, Federal College of Forestry, Jos, Plateau State, Nigeria. A voucher specimen (No. 172) was deposited in the Herbarium unit of the College. The leaves were thoroughly washed with water and air dried under shade. After complete drying, the leaves were ground and sieved to powdery form and stored in plastic container for further use.

Preparation of Extracts

The dried leaf powder (10 g) was macerated in 100 ml absolute ethanol for 92 hours at room temperature in a beaker covered with aluminium foil. The extracts were filtered and evaporated on a water bath, concentrated on a hot plate to constant weight and then stored in refrigerator for further use.

Gas Chromatographic-Mass Spectrophotometric (GC-MS) Analysis

The GC-MS analysis of the ethanol extract was carried out using an AGILENT 7890B GC fitted with a 5977A MSD system at the Institute de Chimie Moléculaire de Grenoble, Université Grenoble Alpes, France. The system is equipped with NIST 20 standard reference database which was used in the documentation of the phytoconstituents.

RESULTS

The gas chromatogram (GC) of the ethanol extract of *S. longipedunculata* leaves (fig. 2) clearly showed fourty seven peaks indicating the presence of fourty seven phytochemical compounds. Table 1 shows some of the phytochemical components revealed by the GC-MS analysis. The identified compounds were ascertained by the NIST reference library data and were selected based on their high score and finger prints matching with corresponding mass spectrum (MS). The individual fragmentation patterns were illustrated in figures 2.1 to 2.17.

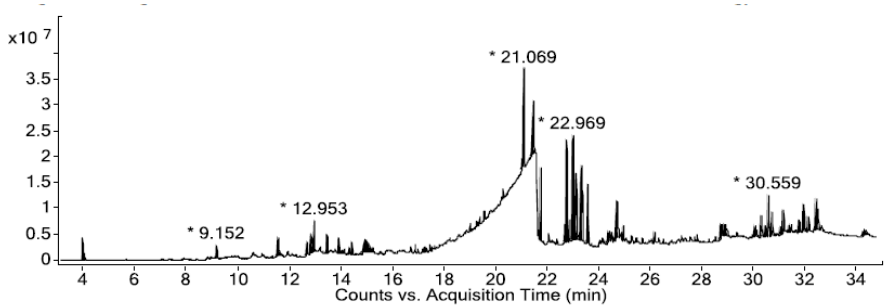

Figure 2: Gas chromatogram (GC) of the ethanol extract of *S. longipedunculata* leaves.

Table 1: Phytochemical compounds present in ethanol extract of *S. longipedunculata* leaves.

S/No.	RT (Min)	Name of Compound	Molecular Formula	Molecular Weight	Composition (%)	NIST score (%)
1	9.152	2-Ethyl-2-hexenal	$C_8H_{14}O$	126	7.26	95.0
2	11.546	2,3-Dihydro-3,5-dihydroxy-6-methyl-4H-Pyran-4-one	$C_7H_{10}O_3$	144	36.08	89.2
3	12.824	5-Hydroxymethylfurfural	$C_6H_6O_3$	126	36.33	87.8
4	14.413	3-Thiophenecarboxylic acid tetrahydro-4-oxo-methyl	$C_6H_8O_3S$	160	7.76	71.3
5	21.069	Methyl palmitate	$C_{16}H_{34}O_2$	270	72.45	88.9
6	21.455	Ethyl palmitate	$C_{18}H_{36}O_2$	284	74.98	86.9
7	21.724	Palmitic acid	$C_{16}H_{32}O_2$	256	48.46	93.1
8	22.733	Methyl (E)-octadec-9-enoate	$C_{19}H_{36}O_2$	296	71.73	93.6
9	22.851	Phytol	$C_{20}H_{40}O$	278	14.17	85.1
10	22.969	Methyl stearate	$C_{19}H_{38}O_2$	298	72.33	95.3
11	23.098	Oleic acid	$C_{18}H_{34}O_2$	282	100.00	92.8
12	23.302	Stearic acid	$C_{18}H_{36}O_2$	284	72.78	95.1
13	23.334	Ethyl oleate	$C_{20}H_{38}O_2$	310	33.60	91.3
14	23.559	Ethyl stearate	$C_{20}H_{40}O_2$	312	33.60	94.6
15	30.033	ɤ-Tocopherol	$C_{28}H_{48}O_2$	416	8.55	78.0
16	30.441	5,7-Dihydroxy-2-(3,4,5-trimethoxyphenyl)-4H-1-benzopyran-4-one	$C_{18}H_{16}O_7$	344	10.16	80.7
17	30.559	α-Tocopherol	$C_{29}H_{50}O_2$	430	31.21	90.7

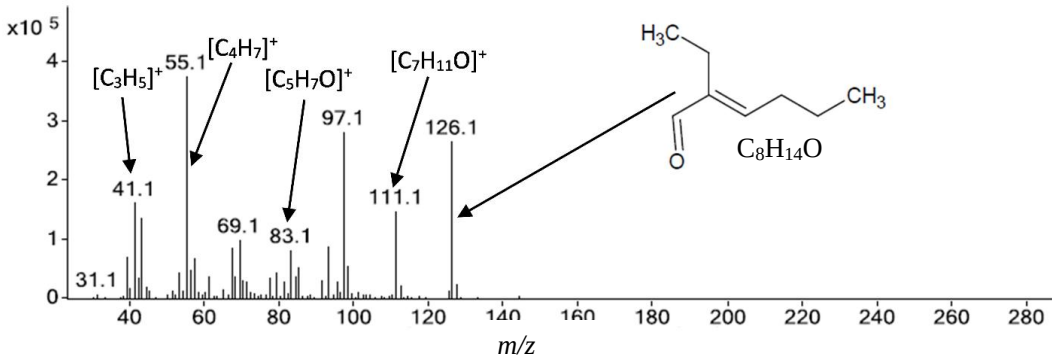

Figure 2.1: MS spectrum of 2-Ethyl-2-hexenal.

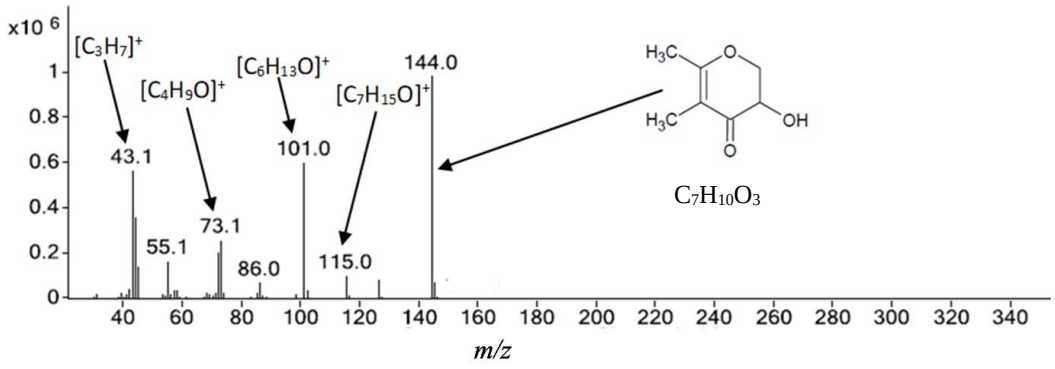

Figure 2.2: MS spectrum of 2,3-Dihydro-3,5-dihydroxy-6-methyl-4H-Pyran-4-one.

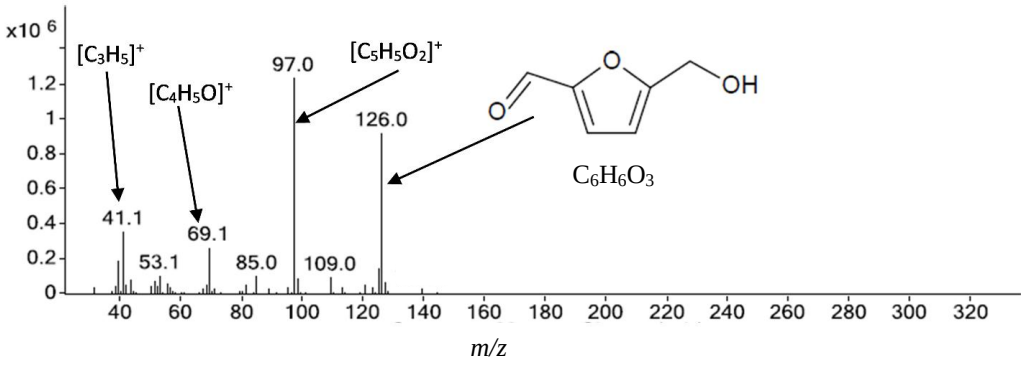

Figure 2.3: MS spectrum of 5-Hydroxymethylfurfural.

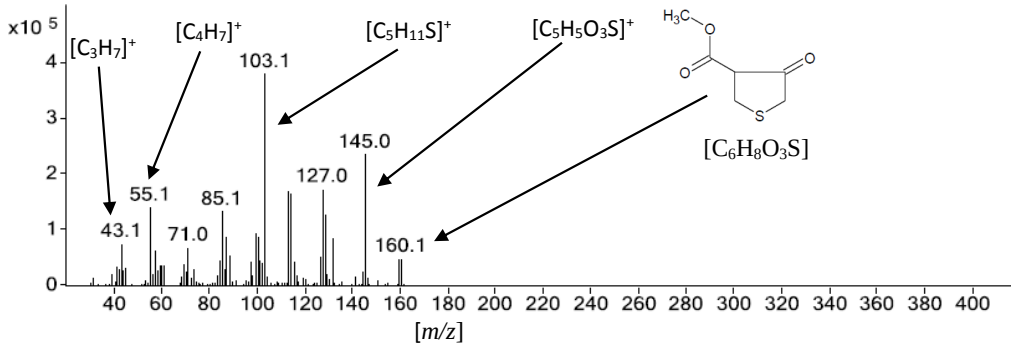

Figure 2.4: MS spectrum of Tetrahydro-4-oxo-methyl-3-thiophenecarboxylic acid.

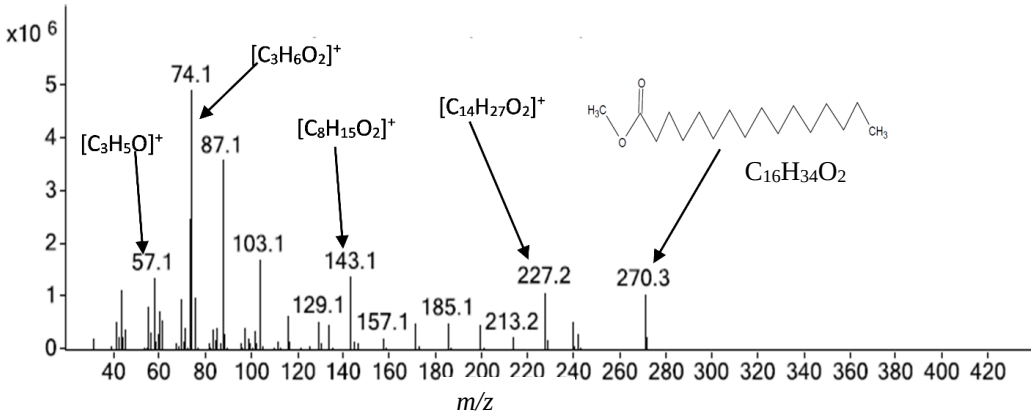

Figure 2.5: MS spectrum of Methyl palmitate.

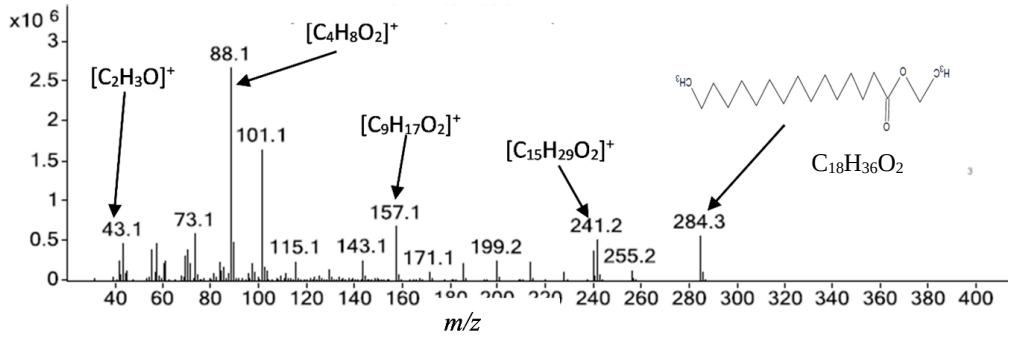

Figure 2.6: MS spectrum of Ethyl palmitate.

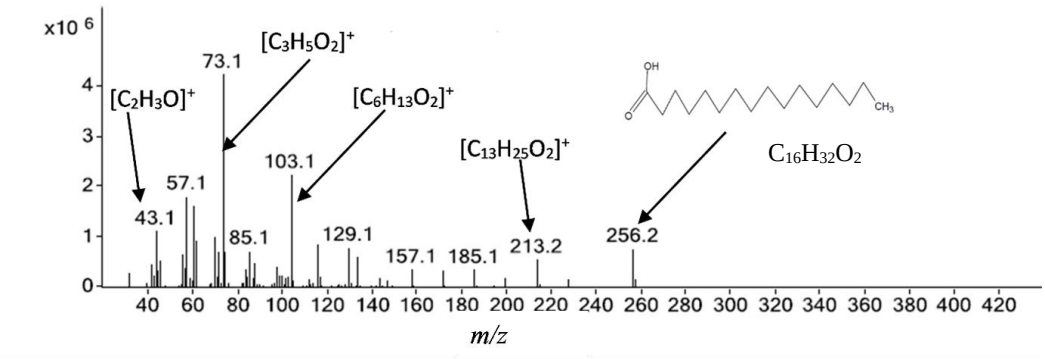

Figure 2.7: MS spectrum of Palmitic acid.

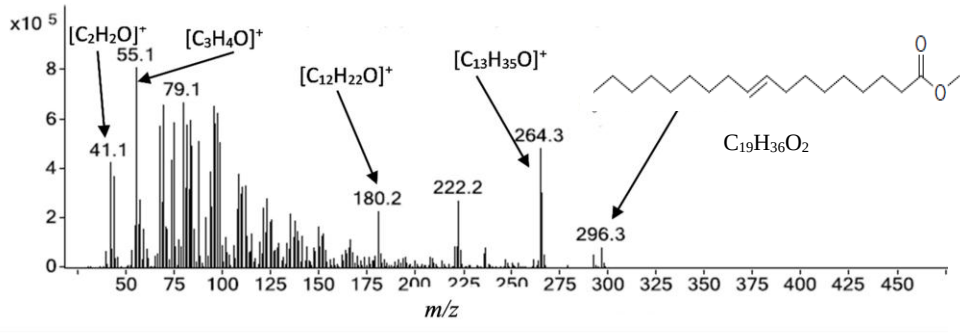

Figure 2.8: MS spectrum of Methyl (E)-octadec-9-enoate.

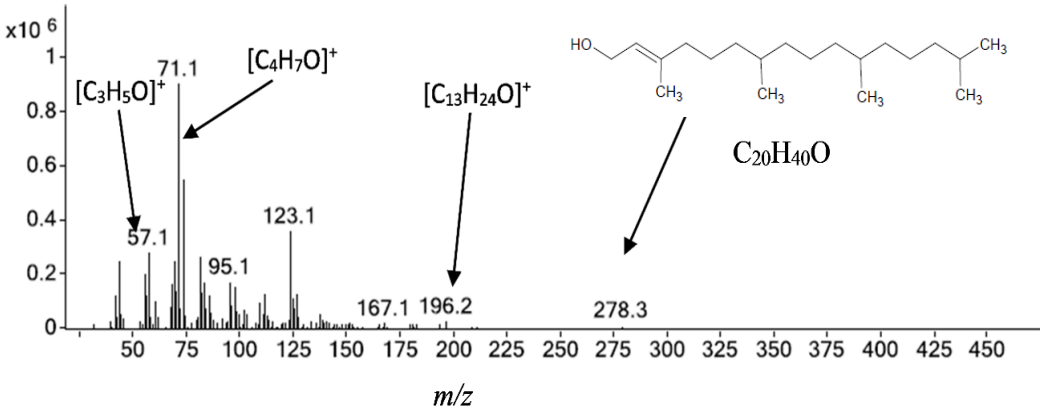

Figure 2.9: MS spectrum of Phytol.

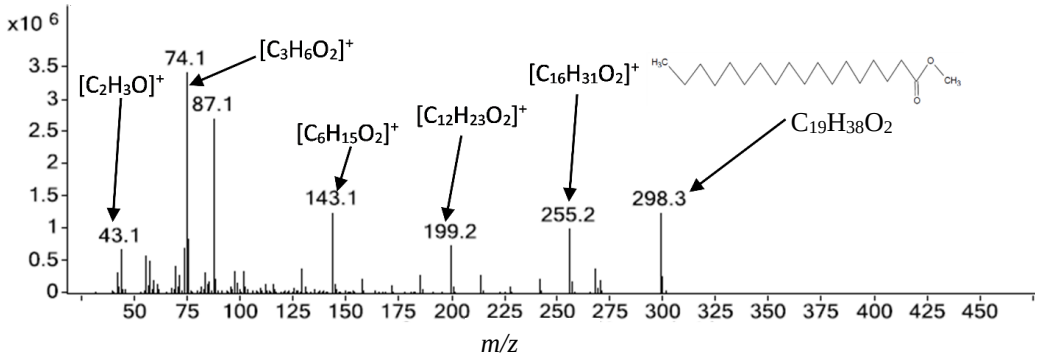

Figure 2.10: MS spectrum of Methyl stearate.

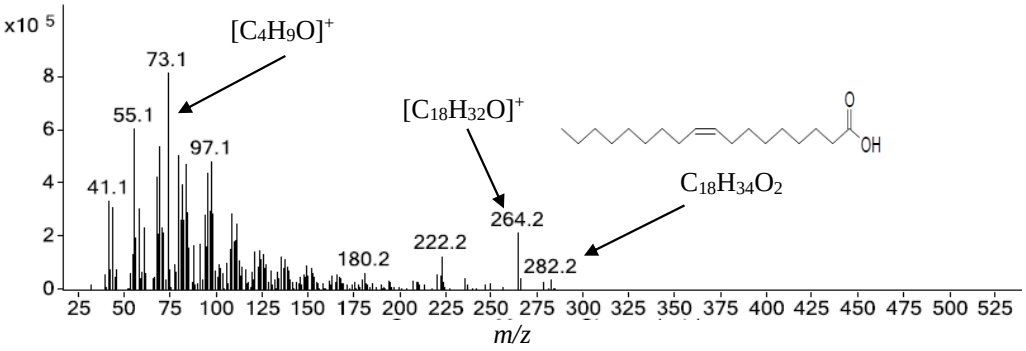

Figure 2.11: MS spectrum of Oleic acid.

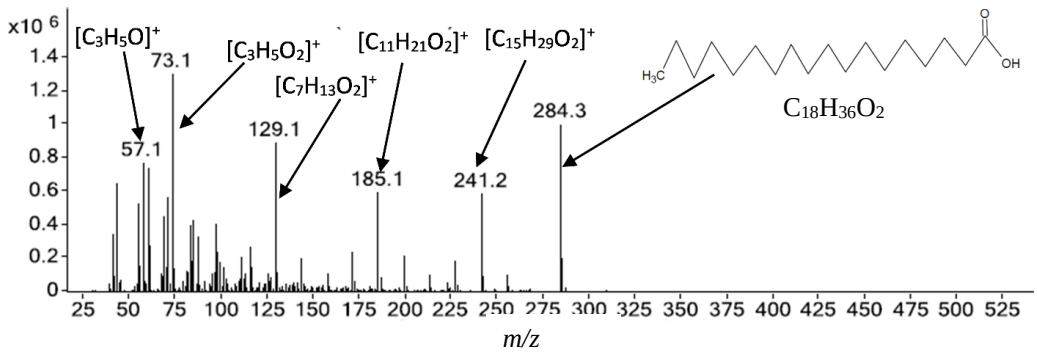

Figure 2.12: MS spectrum of Stearic acid.

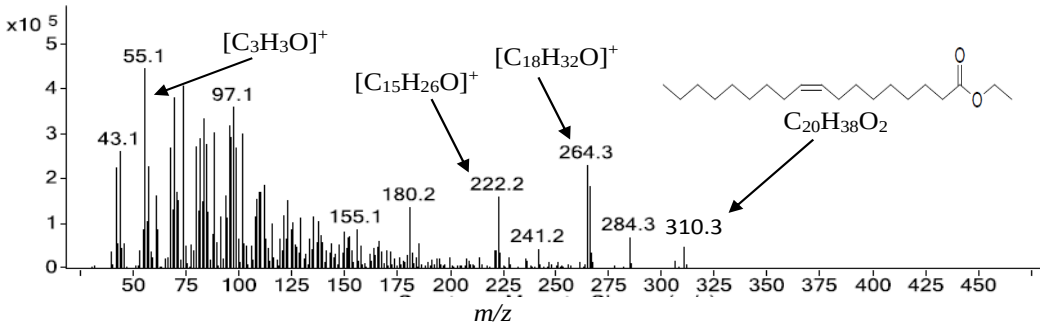

Figure 2.13: MS spectrum of Ethyl oleate.

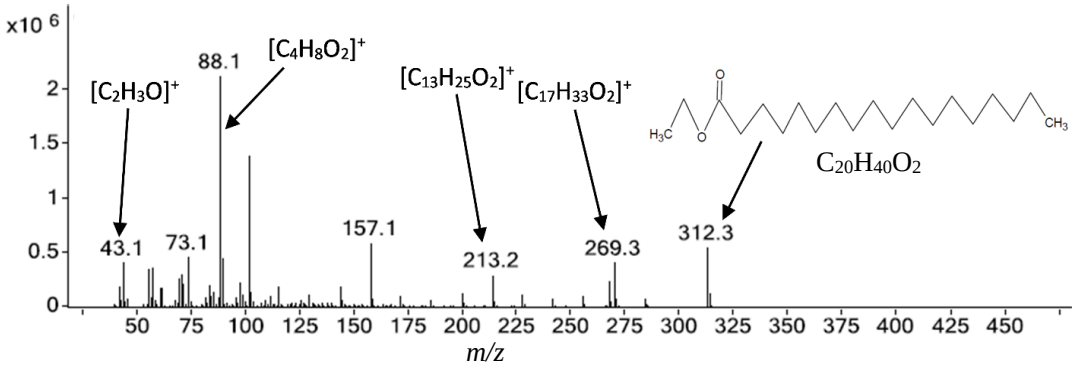

Figure 2.14: MS spectrum of Ethyl stearate.

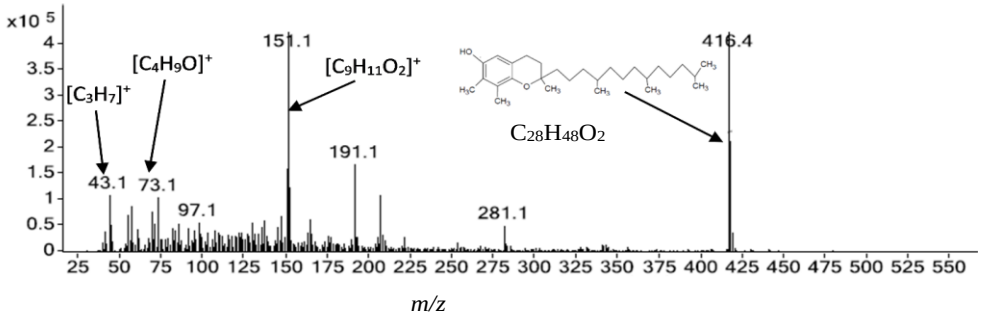

Figure 2.15: MS spectrum of ɤ-Tocopherol.

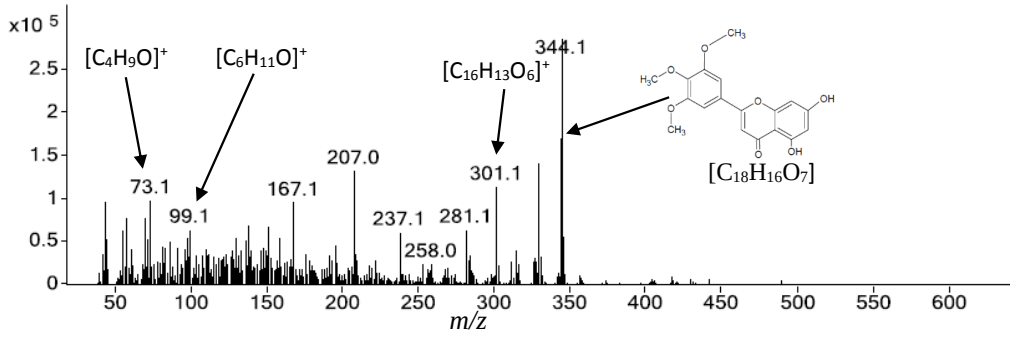

Figure 2.16: MS spectrum of 5,7-Dihydroxy-2-(3,4,5-trimethoxyphenyl)-4H-1-benzopyran-4-one.

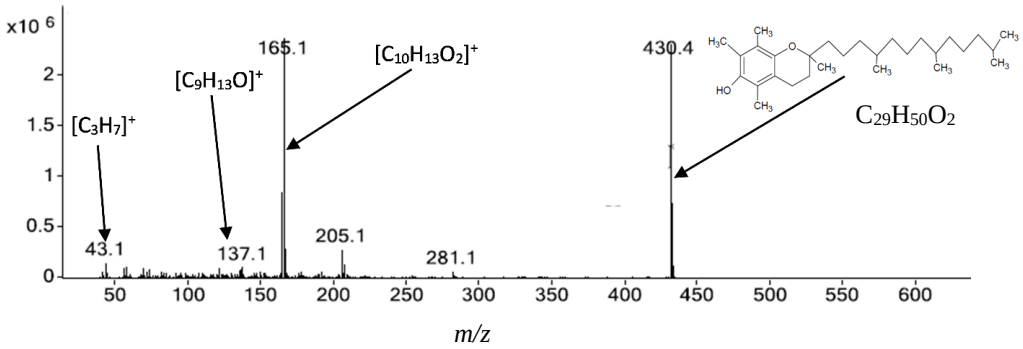

Figure 2.17: MS spectrum of α-Tocopherol.

DISCUSSION

The presence of these compounds in medicinal plants with the exception of 3-Thiophenecarboxylic acid tetrahydro-4-oxo-methyl have been well documented in previous literatures in the last few years. In 2012, Yeh *et al.* reported 2-Ethy-2-hexenal in the leaves of *Peperomia dindygulensis.*[14] Remarkably, the presence of 2-Ethyl-2-hexenal, 2,3-Dihydro-3,5-dihydroxy-6-methyl-4H-Pyran-4-one were observed in foods[15] and treatment of colon cancer cells with different concentrations of the compound (0.5–1.5 mg/mL) for various periods (0–48 h) inhibited the growth of colon cancer cells followed by the induction of apoptosis in a dose dependent manner.[16] 5-Hydroxymethylfurfural was identified in seed extracts of *Buchholzia coriacae*[17] and is the active ingredient in Aes-103, which is being clinically evaluated (Phase 2) for the long-term management of Sickle Cell Disease (SCD).[18] Other very valuable phytoconstituents documented include Methyl palmitate in *Jugans regia* L. seeds[19] and in biodiesel produced from *Cladophorra vagabunda.*[20]

Notably, fatty acids such as ethyl palmitate in extracts of *Isodon rugosus,*[21] Palmitic acid from leaves extract of *Nitraria retusa*[22] and in soybean,[23] methyl stearate from methanol extract of *Melastomastrum capitatum* leaves,[24] oleic acid and ethyl oleate in *Apium graveolens* L. plant,[25] stearic acid in volatile oils from plants and ethyl stearate in volatile oils from plants have been documented.[26] These are vital components present in cooking oils.[25] Very recently, Methyl (*E*)-octadec-9-enoate was reported in fungus *Pestalotiopsis sp.*[27] In 1969, phytol was described in plant materials[28] and has also been reported as the precursor for the synthesis of Vitamin E and Vitamin K1.[29][30] Another constituent present is ɤ-tocopherol which has been reported in seed oil of *Herbiscus sabdariffa.*[31] 5,7-Dihydroxy-2-(3,4,5trimethoxyphenyl)-4H-1-benzopyran-4-one was found in the extracts of *Tetrapleura tetraptera*[32] and α-tocopherol in edible tropical plants.[33]

The findings in this study suggest that *securidaca longipedunculata* leaves are potential candidates for similar applications with other known medicinal plants in pharmaceutical and drug industries. Further studies on this plant are required to provide detailed insight about its phytochemical and pharmacological properties.

CONCLUSION

In this study, seventeen phytoconstituents have been reported from the ethanolic extract of *Securidaca longipedunculata* leaves. The presence of these phytochemical constituents such as esters, fatty acids, phenolics and vatimin E justifies the use of this plant by traditional medicine practitioners for the treatment of variety of ailments. Besides, antimicrobial screening, antioxidant and toxicity investigations are currently being undertaken.

REFERENCES

1. Cragg GM, Kingston DGI, Newman DJ. Anticancer Agents from Natural Products, G. Brunner-Routledge Psychology Press, Taylor & Francis Group, Boca Raton, FL, USA, 2nd edition, 2012.
2. Ngulde SI, Sandabe UK, Abounader R, Dawson TK, Zhang Y, Iliya I, Hussaini IM. Ethanol extract of Securidaca longipedunculata induces apoptosis in brain tumor (U87) cells. BioMed Research International, 2019.
3. Ammal RM, Bai GV. GC-MS Determination of Bioactive Constituents of Heliotropium indicum leaf. Journal of Medicinal Plants, 2013; 1(6): 30-33.
4. Bai S, Seasotiya L, Malik A, Bharti P, Dalal S. GC-MS analysis of chloroform extract of Acacia nilotica L. leaves. Journal of Pharmacognosy and Phytochemistry, 2014; 2(6).

5. Ullah S, Ibrar M, Muhammad N. Pharmacognostic, larvicidal and phytotoxic profile of Coleus forskohlii and Rosmarinus officinalis. Journal of Pharmacognosy and Phytotherapy, 2013; 5(4): 59-63.
6. Arora S, Kumar G. Gas Chromatography-Mass Spectrometry (GC-MS) determination of bioactive constituents from the methanolic and ethyl acetate extract of *Cenchrus setigerus* Vahl (Poaceae). The Pharma Innovation Journal, 2017; 6(11): 635-640.
7. Mulula A, Ntumba K, Mifundu MM, Taba KM. Phytochemical Screening, Antibacterial and Antioxidant Activities of Aqueous and Organics Stem Extracts of Strophanthus hispidus DC. International Journal of Pharmaceutical Sciences and Research, 2017; 8(1): 95-100.
8. Orwa C, Mutua A, Kindt R, Jamnadass R, Simons A. Agroforestree Database: A Tree Reference and Selection Guide, version 4.0, 2009; (accessed 26.01.15).
9. Baloyi O, Tshisikhawe MP. The Population Ecology of Securidaca longepedunculata Fresen in Nylsvlei Nature Reserve, Limpopo Province, RSA. South African Journal of Botany, 2009; 75: 430.
10. Tshisikhawe MP, vanRooyen MW, Bhat RB. An Evaluation of the Extent and Threat of Bark Harvesting of Medicinal Plant Species in the Venda Region, Limpopo Province, South Africa. Phyton, 2012; 81: 89-100.
11. Mongalo NI, McGaw LJ, Finnie JF, Van Staden J. Securidaca longipedunculata Fresen (Polygaceae): A Review of its Ethnomedicinal Uses, Phytochemistry, Pharmacological Properties and Toxicology. Journal of Ethnopharmacology, 2015; 165: 215-226.
12. Junaid SA, Abubakar A, Ofodile AC, Echeonwu GON, Okwori AEJ, Ajetunji JA. Evaluation of Securidaca longipedunculata Leaf and Root Extracts for Antimicrobial Activities. African Journal of Microbiology Research, 2008; 2: 322–325.
13. Haruna Y, Elinge CM, Peni IJ, Dauda D, AikiF. In Vivo Trypanocidal Effect of Aqueous Root Extracts of Securidaca longepedunculata and its Phytochemical Analysis. Journal African of Pharmacy and Pharmacology, 2013; 7: 2838–2842.
14. Yeh CC, Wang CJ, Chen YJ, Tsai SH, Chung WH. Potential of a volatile-producing endophytic fungus Nodulisporium sp. PDL-005 for the control of Penicillium digitatum. Biological Control, 2021; 152: 104459.
15. Ledl F. Proof of 2, 3-dihydro-3, 5-dihydroxy-6-methyl-4H-pyran-4-one in foods. Z. Levensm. Unters. Forsch., 1976; 160: 367-370.
16. Ban JO, Hwang IG, Kim TM, Hwang BY, Lee US, Jeong HS, Yoon YW, Kim DJ, Hong JT. Anti-proliferate and Pro-apoptotic Effects of 2,3-dihydro-3,5-dihydroxy-6-methyl-

4H-pyranone through Inactivation of NF-κB in Human Colon Cancer Cells. Archives of Pharmacal Research, 2007; 30(11): 1455 – 1463.

17. Ojinnaka CM, Nwachukwu KI, Ezediokpu MN. The chemical constituents and bioactivity of the seed (fruit) extracts of Buchholzia coriacea Engler (Capparaceae). Journal of Applied Sciences and Environmental Management, 2015; 19(4): 795-801.
18. U. S. National Library of Medicine. Evaluation of Different Dose Regimens of Aes-103 Given for 28 Days to Subjects With Stable Sickle Cell Disease. Clinical Trials. gov., 2021; https://clinicaltrials.gov/ct2/show/NCT01987908. Retrieved: 22nd July, 2022.
19. Wang YN, Wang HX, Shen ZJ, Zhao LL, Clarke SR, Sun JH, Shi GL. Methyl palmitate, an acaricidal compound occurring in green walnut husks. Journal of Economic Entomology, 2009; 102(1): 196-202.
20. Sharmila S, Rebecca LJ. GC-MS Analysis of esters of fatty acid present in biodiesel produced from Cladophora vagabunda. Journal of Chemical and Pharmaceutical Research, 2012; 4(11): 4883-4887.
21. Lu Q, Liu T, Wang N, Dou Z, Wang K, Zuo Y. Nematicidal effect of methyl palmitate and methyl stearate against Meloidogyne incognita in bananas. Journal of Agricultural and Food Chemistry, 2020; 68(24): 6502-6510.
22. Boubaker J, Toumia I, Sassi A, Bzouich-Mokded I, Ghoul-Mazgar S, Sioud F, Chekir-Ghedira L. Antitumoral potency by immunomodulation of chloroform extract from leaves of Nitraria retusa, Tunisian medicinal plant, via its major compounds β-sitosterol and palmitic acid in BALB/c mice bearing induced tumor. Nutrition and cancer, 2018; 70(4): 650 – 662.
23. Carrero-Colón M, Hudson K. Reduced palmitic acid content in soybean as a result of mutation in FATB1a. Plos one, 2022; 17(3): e0262327.
24. Ukwubile CA, Ahmed A, Katsayal UA, Ya'u J, Mejida S. GC–MS analysis of bioactive compounds from Melastomastrum capitatum (Vahl) Fern. leaf methanol extract: An anticancer plant. Scientific African, 2019; 3: e00059.
25. Achuenu C, Adelakun EA, Ngurukwem EC, Chuwang DJ. Phytoconstituents Profile of The Essential Oils from Celery (Apium Graveolens L) Plant Cultivated in Nigeria by Gas Chromatography-Mass Spectrometry Analysis. World Journal of Pharmacy and Pharmaceutical Sciences, 2022; 11(4): 95 – 106.
26. Wang, H. Q., Liang, F., Qiao, N., Dong, J. X., Zhang, L. Y., Guo, Y. F., Dong, J. X. (2014). Chemical composition of volatile oil from two emergent plants and their algae inhibition activity. Pol. J. Environ. Stud, 23(6): 2371.

27. Liu CM, Wang YL, Wang X, Li GH. Active metabolites from the fungus Pestalotiopsis sp. ymf1. 0474. Chemistry of Natural Compounds, 2021; 57(2): 257-259.
28. Liljenberg C, Odham G. Gas chromatographic determination of phytol in plant material. Physiologia Plantarum, 1969; 22(4): 686-693.
29. Daines AM, Payne RJ, Humphries ME, Abell AD. The Synthesis of Naturally Occurring Vitamin K and Vitamin K Analogues. Current Organic Chemistry, 2003; 7(4): 1 – 15.
30. Thomas N. Synthesis of Vitamin E. Vitamins and Hormones, 2007; 76(7): 155 – 202.
31. Mohamed R, Fernandez J, Pineda M, Aguilar M. Roselle (Hibiscus sabdariffa) seed oil is a rich source of γ-tocopherol. Journal of food science, 2007; 72(3): 207-211.
32. Obeng AW, Boakye YD, Agana TA, Djameh GI, Boamah D, Adu F. Anti-trypanosomal and anthelminthic properties of ethanol and aqueous extracts of Tetrapleura tetraptera Taub. Veterinary Parasitology, 2021; 294: 109449.
33. Ching LS, Mohamed S. Alpha-tocopherol content in 62 edible tropical plants. Journal of Agricultural and food Chemistry, 2001; 49(6): 3101-3105.