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A STUDY TO DETERMINE OF CHEMICAL COMPOSITION OF *CONVOLVULUS SCAMMONIA* L. IN SYRIA BY USING GC-MS

Sarhan Layka [1], Ahmad kara- Ali [2], Abeer Sultan 3, Mayada zahlout[4]

[1]Department of Botany, Faculty of Sciences, Tishreen University, Lattakia, Syria.
[2] Department of High Institute Marine Research, Tishreen University , Lattakia , Syria
[3] Department of Botany, Faculty of Science, Tishreen University, Lattakia, Syria.
[4]PhD Student, Department of Botany, Faculty of Sciences, Tishreen University, Lattakia, Syria.
*Corresponding author e-mail: zahloutmeada@gmail.com

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ABSTRACT

The current study aims to analyze the biochemical compounds likely to be found in the roots , leaves, and the flowers of c.scammonia L., which is one of the distinctive species of the genus convolvulus (Connolvolaceae), which is originally from the countries of the eastern part of the Mediterranean basin, known as mahmoda helablab. It has been used since Ancient eras in the treatment of infectious and chronic diseases such as: vitiligo diseases, arthritis, and digestive disorders, and scientific research recently showed its anti-cancer effectiveness. ,the plants were collected from Slenfeh,east of Lattkia –Syria..the active compounds were identified by using gas chromatography coupled with mass spectrometry (GC-MS) analysis.21 compounds were identified in the roots, 18 in the leaves, and 6 in the flowers. The prevailing compounds in the roots are n-Hexadecanoic (palmitic) acid, 1,2 Benzenedicarboxylic acid , mono (2-ethylhexyl) este, and Eicosane (27.09, 13.69, ,15.17, %), respectively. While in the leaves are Oxalic acid, isobutyl tetradecyl ester (5.73%), Stearic acid, 3-(octadecyloxy)prop yl ester Eicosane (9.60%) and phytol (10.63%) Esters are among the important compounds in the manufacture of men's perfumes and cosmetics , flavors, and pharmaceutical preparations.And we noticed the presence of two compounds of fatty acids in flowers chloropropionic acid (2.02%) and (palmitic) acid (7.16%), which shows vital activity as antioxidant, antibacterial, and antifungal.This study paves the way for the use of extracts in Pharmaceutical and industrial applications

INTRODUCTION

Medicinal plants have been used in the management of diseases for thousands of years and continue to provide an inexhaustible source of natural compounds to serve society.(Salamatullah,2022); as over 3.4 billion individuals in the developing world use traditional plant-based treatments. This equates to more than 88 percent of the world's population, with conventional medicine providing the majority of primary health care. The World Health Organization (WHO) supports traditional medicine if it can be proven effective and safe (WHO 1985). So that ,there is a need to verify herbal medicine's ethnomedicinal use and then isolate and characterize the chemicals that are likely to be added to the prospective. (Velmurugan and Anand;2017) The medicinal property of a plant depends upon the physiologically active biochemical compounds called secondary

metabolites. Plants have an almost limitless ability to synthesize secondary metabolites which present in the plant parts like leaves, fruits, buds, stem, flowers, bark, roots, *etc* (Hussein and El-Anssary; 2018) *Convolvulus* genus is a representative of the family of Convolvulaceae. *Convolvulus* plants are broadly distributed all over the world and has been used for many centuries as herbal medicine.(Salehi et al;2021) *Convolvulus scammonia* L. belong to the morning glory family (Convolvulaceae) . It is a family of flowering plants containing over 59 genera with close to 1950 species. (Staples; 2018).Flowering between April and July; fruiting between June and September. Distribution:East Aegean Islands, Palestine, Iraq, the Crimea, Lebanon, Syria and Turkey and ecology :Road- and field sides, shaly, sandy, calcareous, limestone and rocky slopes, macchie (0-1540 m).(Aykurt,.and Sümbül, 2014) Common names: Arabic: Sigmonia, mahmoda, helablab; English: scammony Syrian bindweed; French: scammonée; German: kleinasiatische Winde; India: sak munia, Spanish: escamonia; Swedish: hartsvinda .*Convolvulus scammonia* is bindweed native to the countries of the eastern part of the Mediterranean. (USDA,2015)

Researchers have paid More attention to Convolvulus plant extracts and essential oils due to their biological properties, bioavailability, clinical efficacy, and safety (Chen et al., 2018). Recently, extensive studies conducted on the biological activities of *Convolvulus* species such as *C. arvensis* had important antioxidant and antibacterial properties, which may be due to their richness in potentially active compounds such as carvacrol and thymol. (Salamatullah; 2022) , and antioxidant effects of the *C. prostratus* (Singh and Vora; 2017) . The genus Convolvulus was found to be wealthy in flavonoids, steroids, terpenoids, carbohydrates, amino acids, anthraquinones (Al-Rifai et al.,2017), anthocyanidins, phenylpropanoids, coumarins, lignans, resins (Chen et al .,2018) tannins, saponins, alkaloids, lipids, and fatty acids (Salehi et al.,202)

Furthermore a new concern in resin glycosides caused by the discovery of novel biological activities for C. scammonia such as cytotoxicity toward cancer cells (Fan et al.,2019), anti-bacterial (Pereda-Miranda et al,2010), anti-viral (Ono, 2017), anti-inflammatory (Yoshikawa et al .,2016), and multidrug resistance modulatory (Corona-Castaneda et al; 2016). Saqmonia is a gum-resin of *Convolvulus scammonia* L., it has been widely used in Unani system of medicine from the ancient times by the great Unani scholars for the treatment of infectious as well as chronic diseases like skin diseases *Pityriasis*, Vitiligo, Tinea, Sciatica, arthritis, gastro-intestinal disorders, dropsy, anasarca, intestinal worm infestations, resolvent and blood purifier. Recent scientific studies have showed the anticancerous activity of scammoni resin. Saqmonia exhibit the medicinal properties due the presence of active constituents like jalapin, and scammonin (scammonin I and II).(Zahid et al.,2020) Quadrupole-Orbitrap Mass Spectrometer were applied to carry out qualitative and quantitative determination A total of 80 components were identified, among which 79 indicated hydroxy C16 fatty acid as main aglycone that was in accordance with phytochemistry study on *Convolvulus scammonia* . In addition, aglycone with hydroxy C17 fatty acid was discovered in the plant,(Yin et al;2022). In recent years, GC-MS has solidified its position as a key technical platform for secondary metabolite profiling in both plant and non-plant species.(Velmurugan and Anand; 2017). GC–MS is a reliable technique for the identification of various compounds such as alkaloids, flavonoids, organic acids, amino acids etc. from plant extracts (Razack et al.;2018) Hence, the goal of this study was aimed to find the phytochemicals / bioactive compounds that are exiten in the (flower ; leaf and root) of *Convolvulus scammonia* L.by Gas chromatography and mass spectrometry analysis; however, To the best of our knowledge, no report on the of C. scammonia L. has been published up to date.

2. Materials and Methods

2.1. **Plant Material:** The three parts of the plant (root, leaf, flower) were collected from Selnfi (in Syria) ; the town is located 50 Km east of lattakia and is located in the herrt of the coastal Mountains of Syria , they are collected at summer of 2021.

2.2.Preparation of Plant Extracts: Freshly collected roots; leaves and flowers of *C.scammonia* L. were washed with running tap water. dried under shade for two weeks. The properly dried roots; leaves and flowers were powdered separately in an electric blender. The coarsely powdered materials were kept in airtight containers to avoid the effect of humidity and then stored at room temperature until use. About 500 g of dry powdered samples were extracted with ethanol (95%) by continuous hot percolation using Soxhlet extractor for 24 h. The extract was filtered through Whatman no.41 filter paper separately, and the extracts were concentrated in vacuum at 60 ºC using a rotary evaporator to evaporate the ethanol from it. To evaporate the remaining solvent, the extracts were kept in an oven at a temperature of 40-50 ºC for 8 h. The residues were kept separately in airtight containers and stored at 4 ºC for further use

2.3.Preliminary Phytochemical Analysis: The ethanolic extracts were used for qualitative identification of various secondary metabolites were carried out by using standard methods.

2.4.Gas Chromatography-Mass Spectroscopy (GC-MS) Analysis: Gas Chromatography-Mass Spectrometry (GC-MS) plays a key role in the analysis of unknown components of plant origin. The chemical composition of root; leaf and flower of *C.scammonia* L. were subjected to GC-MS.

GC-MS analysis of these extracts were carried out using the equipment GC Clarus 500 Perkin-Elmer system comprising a AOC–20i autosampler and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following condition: Equipped with a column Elite-1, fused silica capillary column (30 m × 0.25 mm ID × 1µm df, composed of 100% dimethyl polysiloxane), operating in electron impact mode at 70 eV; helium gas (99.999%) was used as carrier gas at a constant flow rate of 1 ml/min and an injection volume of 2 µl was employed (split ratio of 10:1). The injector temperature is set at 250 °C, and the ion-source temperature is 280 °C. The oven temperature was programmed from 110 °C (isothermal for 2 min), with an increase for 10 °C/min, to 200°C/ min, then 5°C/ min to 280 °C/min, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan-interval of 0.5 seconds, and fragments from 45 to 450 Da. Total GC running time was 36 min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Software adapted to handle mass spectra, and chromatograms was a Turbo mass ver. 5.0.

Interpretation on mass spectrum of GC-MS was conducted using the database of National Institute of Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight, and structure of the components of the test materials were ascertained and the results obtained have been tabulated (Merlin et al ;2009)

3. RESULTS AND DISCUSSION

The results of the chemical analysis by GC-MS of the roots, leaves and flowers of *C.scammonia* L. showed the presence of a wide spectrum of biologically active chemical compounds as shown in Tables (1-2-3). Twenty-one compounds were detected in the root of *C.scammonia*, and the hydrocarbons were more diverse, reaching eleven compounds, the most important of which were: Eicosan (C20H42) (15.92%), 2-tetradecene (3.11%). octadecene (2.43%), 1-Hexadecen (0.47%). Three fatty acids appeared, the most important of which was n-Hexadecanoic (palmitic) acid (27.09%). As for esters: 1,2 Benzenedicarboxylic acid, mono (2-ethylhexyl) ester(13.69 %) , aldehydes and alcohols, they were in varying proportions (2.05, 0.17). % Respectively. Figure(1)

Table (2) and Figure (2) the results of the

analysis of the leaf extract of the studied species, which shows that it contains a mixture of active compounds, which amounted to 18 different compounds, as hydrocarbons, esters, fatty acids, ketones and terpenoids recorded percentages (15.04, 14.04, , 7.37 ,10.63,1.32%), respectively. It should be noted It indicated the presence of Eicosane (9.60 %), Stearic acid, 3-(octadecyloxy)prop yl ester (5,73%), Oxalic acid, isobutyl tetradecyl ester (7.33 %) , and PHYTOL (10.63%) in the leaves of C. scammonia), which is one of the common terpenoid compounds. We noted from the table (3) the presence of three compounds belong to the group of hydrocarbons, which were dominant in the flowers of *C.scammonia* L. Octasiloxan (0.05%), . octadecene (0.04 %) Eicosane (0.06%). , Aldehydes were represented by the compound 2-undecenal (0.14%), followed by fatty acids with two compounds: chloropropionic acid (2.02%) and palmitic acid (1.16%) Figure (3) The results of the chemical analysis by using GC-MS technology showed a great diversity in the chemical compounds of C.scammonia plant parts. These results consistent with previous research stating that the genus convolvulus possesses oxygenated sesquiterpenes, sesquiterpenes, hydrocarbons, and the compound Octasiloxane is an important antimicrobial (Hassine et al..;2014) This study also consistents with previous research on the root of the species Convolvulus althaeoides, which contains four basic compounds, the most important of them was n-hexadecenoic acid(29.77%) (Hrichi, et al.; 2022), *Among the fatty acids, n*-Hexadecanoic (palmitic) acid (C16H32O2) was the most abundant compound, This has several biological activities: antimicrobial, antieczematous, antiseborrheic, sclerosing, antihypoxic, antimutagenic, fibrinolytic, anti-inflammatory, antisecretory, cytoprotective and anesthetic (. Karthikeyan et al.;2019) and anti oxidant activities (Kala et al.,2011) . In fact, palmitic acid is one of the primary metabolites produced during microbial degradation because of the oxidized end of the molecule is used as the initial site for the β-oxidation process, whereas microbial oxidation of saturated hydrocarbons (n-hexadecane) cannot be initiated as easily as for n-hexadecanoic acid (Zyakun et al;2012). the resulting Esters are important organic compounds with increasing commercial applications (Foresti et al; 2005). These compounds are largely used in fragrances, cosmetics detergents, flavors and pharmaceuticals. Esters (ethyl oleates) may also be used as plasticizers and lubrificants; biological additives and hydraulic fluids (Hazarika et al.; 2002) Phytol is an extremely common terpenoid, found in all plants esterified to Chlorophyll to confer lipid solubility. Phytol is commonly used as the basic raw materials for the manufacture of synthetic forms of vitamin E (Netscher, 2007) and vitamin K1 (Daines *et al*., 2003) Thus, because of the richness of this plant in the above-mentioned compounds, we can appreciate its presence in our nature. It is used in various pharmaceutical and industrial applications, and we must protect it from extinction, as it is present in small numbers and in specific areas.

Table 1.Compound Identified In the Roots of C.Scammonia

No.	Amount (%)	The Name of Compound	Compound Class
1	0.14	1,1-Dodecanediol, diacetate	Hydrocarbons
2	0.98	Oxirane, tetradecyl-	
3	2.29	Cyclotetradecane	
4	3.11	2-Tetradecene,	
5	0.58	Heptacosane,	
6	0.31	Cyclohexadecane	
7	2.43	Octadecane	
8	15,92	Eicosane	
9	0.48	5-Eicosene,	
10	0.32	Heptadecane	
11	0.47	-1-Hexadecene	
12	0.11	Heptafluorobutyric acid,	*Fatty Acids*
13	27.09	n-Hexadecanoic acid	
14	0.12	3-hydroxy-2-methylbutyric acid	
15	13.69	1,2 Benzenedicarboxylic acid,- mono (2-ethylhexyl) ester	Esters
16	0.65	Decenal, (E)--2	Aldehydes
17	0.41	2,4-Decadienal, (E,E)-	
18	0, 73	2-Undecenal	
19	0.08	3,5-di-tert-Butyl-4	
20	0.18	Hydroxybenzald	
21	0.17	2-Decen-1-ol, (E)-	Alcohols

Table 2.Compound Identified In the Leaves Of C.Scammonia

No.	Amount (%)	the Name of Compound	Compound Class
1	2.36	Bicyclo[3.1.1]heptane	Hydrocarbons
2	0.12	2-Hexadecene	
3	0.40	1,4-Eicosadiene	
4	0.56	Cyclohexadecane	
5	9.60	Eicosane	
6	0.59	3-Eicosene	
7	0.12	Octasiloxane	
8	1.19	Heneicosane	
9	0.70	Nonadecane	
10	0.77	cyanomethyl)-, ethyl ester -Benzeneacetic acid, .alpha.-cyano	Esters
11	5,73	Stearic acid, 3-(octadecyloxy)prop yl ester	
12	7.33	Oxalic acid, isobutyl tetradecyl ester	
13	0.21	Bromoacetic acid, hexadecyl ester	
14	3.46	Tridecanoic acid	Fatty Acids
15	3.91	Tetradecanoic acid	
16	10.63	Phytol	Terpenenoides
17	1.32	Pentadecanone-2	Ketones
18	1.27	3]-1)-4-Cyano-1,2,3,4-tetrahydrona	Other Compounds

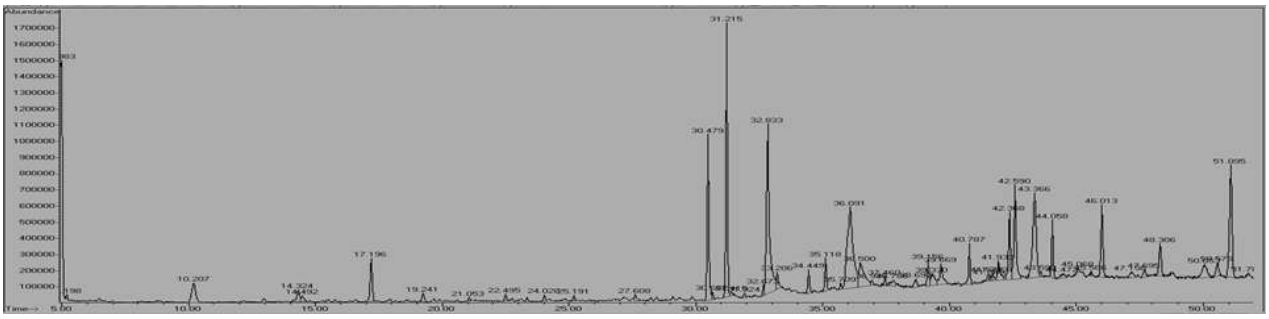

Figure 1.GC-MS Chromatogram of C.Scammonia Roots

Figure 2.GC-MS Chromatogram of C.Scammonia Leaves

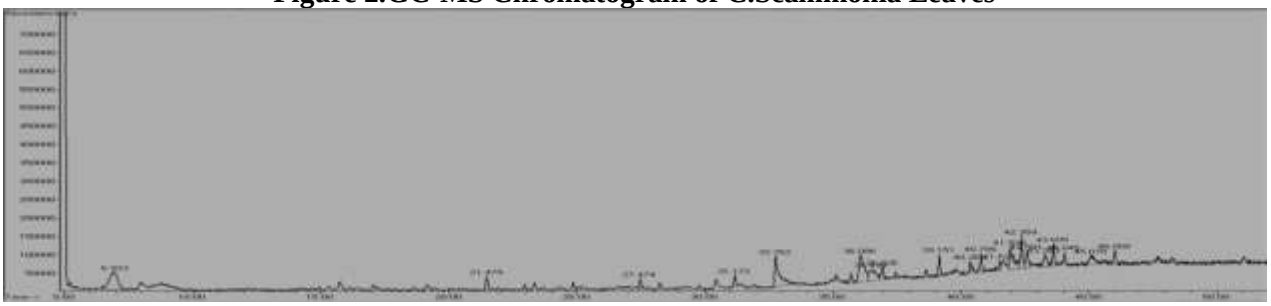

Figure 3: GC-MS Chromatogram of C. Scammonia Flowers

Table 3: Compound Identified in The Flowers Of C. Scammonia

No.	Amount (%)	the Name of Compound	Compound Class
1	0.06	Eicosane	Hydrocarbons
2	0,04	Octadecane	
3	0,05	Octasiloxane	
4	0.14	2-Undecenal	Aldehydes
5	2.02	Chloropropionic acid	Fatty Acids
6	1.16	n-Hexadecanoic acid	

4 .CONCLUSION

The present research investigates for the first time the chemical profiles of C. scammonia L. roots, leaves and flowers and their biological activities.. The largest number of biochemical compounds that we got was recorded from the roots, followed by the leaves, where as the flowers were the least diverse in the active chemical compounds. The group hydrocarbons were recorded as the most present in all plant parts of the species,. Among The fatty acids n-hexadecenoic acid (palmitic) acid was the most abundant compound, which has several biological activities .Therefore, we recommend conducting future studies to isolate and purify these compounds, test their effectiveness, and study our applications.

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