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GC-MS Analysis of Bioactive Components of *Shepherdia argentea* (Pursh.) Nutt. from Ukrainian Flora

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Gas chromatography mass spectrometry (GC-MS) analysis revealed the presence of 20 compounds in leaves of *Shepherdia argentea* (Pursh.) Nutt. In GC-MS analysis, some of the phytocomponents screened were phytol, squalene and n-hexadecanoic acid. The compounds were identified by comparing their retention time and peak area with that of literature and by interpretation of mass spectra. Many of them have antioxidant, anti-inflammatory, antimicrobial and anticancer actions.

Keyword: *Shepherdia argentea* (Pursh.) Nutt., Gas Chromatography-Mass Spectrometry (GC-MS).

1. Introduction

Medicinal plants have been used by human being since early ages in traditional medicine due to their therapeutic potential and the search on medicinal plants has led to the discovery of novel drug candidates used against diverse diseases. Higher plants as sources of bioactive compounds continue to play a dominant role in the maintenance of human health [1].

Plants are a rich source of secondary metabolites with interesting biological activities. In general, these secondary metabolites are an important source with variety of structural arrangements and properties.

In recent years Gas chromatography – Mass Spectrum (GC-MS) studies have been increasingly applied for the analysis of medicinal plants as this technique has proved to be a valuable method for the analysis of essential oil, alcohols, acids, esters, alkaloids, steroids, amino and nitro compounds etc.

Shepherdia Nutt is a small genus of the family *Elaeagnaceae Juss* and possesses only 3 species which are widely distributed in more northern and cooler parts of North America [2]. species *Shepherdia argentea* (Pursh.) Nutt. cultivated in Ukraine [3, 4]. These species have been a keen interest in phytochemical and pharmacological research due to their excellent medicinal values. *Shepherdia argentea* (Pursh.) Nutt. berries were traditionally used by Native Americans to treat stomach troubles. Tannins from the leaves of *Shepherdia argentea* (Pursh.) Nutt. have

demonstrated the capacity to inhibit HIV-1 reverse transcriptase.

2. The Aim of the Study:

The aim of the present study is to identify the phytocomponents of *Shepherdia argentea* (Pursh.) Nutt. with the use of Gas chromatography – Mass Spectrum analysis. In the present study, volatile organic matter of the leaf sample of plant was analyzed for the first time. This work will help to identify compounds, which may be used in therapeutic value.

3. Materials and Methods

The objects of the study were leaves of *Shepherdia argentea* (Pursh.) Nutt. harvested in the ripening stage in the summer 2011 in M. M. Gryshko National botanical garden NAS of Ukraine. GC-MS analyses were performed on a Agilent Technologies 6890 capillary gas chromatograph directly coupled to a mass spectrometer system (model 5973). A fused silica capillary column 5% phenyl-poly-dimethyl-siloxane (DB-5 capillary column (30 m * 0.25 mm inner diameter, with 0.25 mm film thickness) was used under the following conditions: oven temperature program from 50 °C (3 min hold) to 320 °C at 4 °C/min; injector temperature 250 °C; carrier gas He, flow rate 1,2 mL/min; the volume of injected sample was 1.5µl; splitless injection technique.

Individual components were identified by comparison of their mass spectra using both "NIST-MS Library 05" and "Wiley GC-MS Library 2007".

Relative percentage amounts of separated compounds were calculated automatically from peak areas of the total ion chromatograms (TIC).

This is done in order to determine whether these plant species contains any individual compound or group of compounds, which may substantiate its current commercial and traditional use as the herbal medicine. Further it helps to determine the most appropriate methods of extracting these compounds. These results will consequently be discussed in the light of their putative biological or therapeutic relevance.

4. Results and Discussion

The components present in leaves of *Shepherdia argentea* (Pursh.) Nutt. were identified by GC-MS analysis (Figure 1). The identified compounds are listed in Tabl.1, 2.

The GC-MS analysis of *Shepherdia argentea* (Pursh.) Nutt. leaves revealed presence of twenty compounds. Identified compounds possess many biological properties. Among the identified phytochemicals is an n-hexadecanoic acid may have a role in antioxidant, hypocholesterolemic and anti-inflammatory effects. Pentacosane, heptacosane, nonacosane, tricosane, pentadecane exhibited antibacterial activity [5, 6].

Squalene has properties of an antioxidant and immunostimulant it acts as antibacterial, antitumor and cancer preventive agent [1].

Phytol is a key acyclic diterpene alcohol that is a precursor for vitamins E and K1 and an antioxidant and a preventive agent against epoxide-induced breast cancer carcinogenesis [7].

Linalool is a principal component of many essential oils known to have several biological activities such as antibacterial [8], sedative [9], anti-inflammatory [10].

Previous studies have demonstrated that α-terpineol possesses pharmacological activities, such as, anticonvulsant [12], sedative [13], antinociceptive [14], anticancer [15], hipotensive [16], gastroprotective [17] and antifungal [18].

Medicinal plants have been used by human being since early ages in traditional medicine due to their therapeutic potential and the search on medicinal plants has led to the discovery of novel drug candidates used against diverse diseases. Higher plants as sources of bioactive compounds continue to play a dominant role in the maintenance of human health [1].

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valuable method for the analysis of essential oil, alcohols, acids, esters, alkaloids, steroids, amino and nitro compounds etc.

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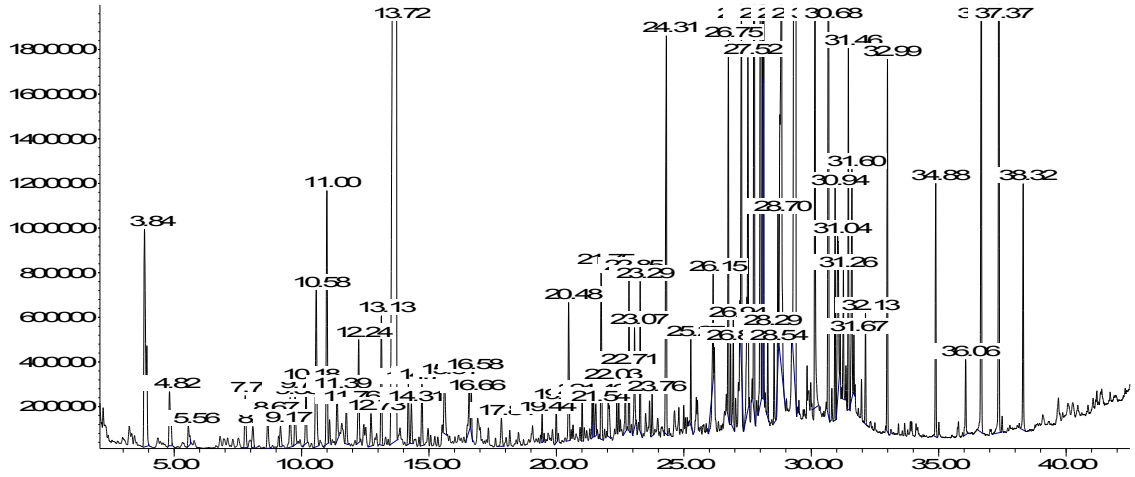

Fig 1: GC-MS chromatogram of the leaves of *Shepherdia argentea* (Pursh.) Nutt.

Table 1: GC-MS analysis revealed the presence of phytochemical components in leaves of *Shepherdia argentea* (Pursh.) Nutt.

S. No	R/T	Name of the Compound	Molecular Formula	MW	Peak Area %
1	10.58	3,7-dimethylocta-1,6-dien-3-ol	$C_{10}H_{18}O$	154	4.9
2	11.00	Nonanal	$C_9H_{18}O$	142	6.3
3	14.72	Alpha-Terpineol	$C_{10}H_{18}O$	154	1.6
4	19.44	Pentadecane	$C_{15}H_{32}$	212	0.5
5	21.45	α: (3E)-4-(2,6,6-Trimethylcyclohex-2-en-1-yl)but-3-en-2-one	$C_{13}H_{20}O$	192	0.7
6	21.54	Hexadecane	$C_{16}H_{34}$	226	0.7
7	22.85	β: (3E)-4-(2,6,6-Trimethylcyclohex-1-en-1-yl)but-3-en-2-one	$C_{13}H_{20}O$	192	2.4
8	26.15	Tetradecanoic acid	$C_{14}H_{28}O_2$	228	1.2
9	28.70	Palmitic acid, ethyl ester	$C_{18}H_{36}O_2$	284	4.2
10	28.83	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	18.1
11	30.14	Phytol	$C_{20}H_{40}O$	296	9.4
12	30.94	Tricosane	$C_{23}H_{48}$	324	3.7
13	31.04	Cis-9-Octadecenoic acid	$C_{18}H_{34}O_2$	282	5.2
14	31.26	9,12-Octadecadienoic acid	$C_{18}H_{32}O_2$	280	2.2
15	31.46	Ethyl linoleate	$C_{20}H_{36}O_2$	308	5.4
16	31.60	9,12,15-Octadecatrienoic acid	$C_{18}H_{30}O_2$	278	4.8
17	32.99	Pentacosane	$C_{25}H_{52}$	352	5.9
18	34.88	Heptacosane	$C_{27}H_{56}$	380	4.2
19	36.67	Nonacosane	$C_{29}H_{60}$	408	3.9
20	37.37	Squalene	$C_{30}H_{50}$	410	14.7

Table 2: GC-MS analysis showed phytochemical compounds, their nature and biological activities of leaves *Shepherdia argentea* (Pursh.) Nutt.

S. No	R/T	Name of the Compound	Compound Nature	Activity
1	10.58	3,7-dimethylocta-1,6-dien-3-ol	Oxygenated monoterpen	antibacterial [8], sedative [9], anti-inflammatory [10]
2	11.00	Nonanal	Aldehyde	Antidiarrhoeal [11]
3	14.72	α-terpineol	Terpene	anticonvulsant [12], sedative [13], antinociceptive [14], anticancer [15], hipotensive [16], gastroprotective [17], antifungal [18]
4	19.44	Pentadecane	Aliphatic hydrocarbon	antibacterial [5]
5	21.45	α: (3E)-4-(2,6,6-Trimethylcyclohex-2-en-1-yl)but-3-en-2-one	Norterpenoide	No activity reported
6	21.54	Hexadecane	Aliphatic hydrocarbon	Antibacterial, antioxidant [5]
7	22.85	β: (3E)-4-(2,6,6-Trimethylcyclohex-1-en-1-yl)but-3-en-2-one	Norterpenoide	Antioxidant, anticancer, [19, 20, 21]
8	26.15	Tetradecanoic acid	Fatty acid	antibacterial, antifungal [22]
9	28.70	Palmitic acid, ethyl ester	Ester	Inhibits phagocytosis [23]
10	28.83	n-Hexadecanoic acid	Fatty acid	Antioxidant, hypocholesterolemic, anti-inflammatory, antibacterial [1]
11	30.14	Phytol	Diterpene	Antimicrobial, anticancer, cancer preventive, diuretic anti-inflammatory [1]
12	30.94	Tricosane	Aliphatic hydrocarbon	antibacterial [6]
13	31.04	Cis-9-Octadecenoic acid	Fatty acid	Antitumor, antibacterial [24]
14	31.26	9,12-Octadecadienoic acid	Fatty acid	Anti-inflammatory, hypocholesterolemic, antibacterial, hepatoprotective, antihistaminic, antiarthritic, anticoronary [1]
15	31.46	Ethyl linoleate	Ester	antisclerotic [25]
16	31.60	9,12,15-Octadecatrienoic acid	Fatty acid	Anti-inflammatory, hypocholesterolemic, cancer preventive, hepatoprotective, antibacterial, antiarthritic, anticoronary [1]
17	32.99	Pentacosane	Aliphatic hydrocarbon	antibacterial [6]
18	34.88	Heptacosane	Aliphatic hydrocarbon	antibacterial [6]
19	36.67	Nonacosane	Aliphatic hydrocarbon	antibacterial [6]
20	37.37	Squalene	Triterpene	Antibacterial, antioxidant, antitumor, cancer preventive, immunostimulant [1]

Amongst diverse and potent biological activities of fatty acids and their derivatives is the ability to kill or inhibit bacterial growth. Their broad spectrum activity, non-specific mode of action and safety makes them attractive as antibacterial agents for various applications in medicine, agriculture and food preservation, especially where the use of conventional antibiotics is undesirable or prohibited.

These reports are in accordance with the result of this study.

Therefore, GC-MS method is a direct and fast analytical approach for identification of terpenoids and fatty acids and only few grams of plant material is required. The importance of the study is due to the biological activity of some of these compounds. The present study, which reveals the presence of components in *Shepherdia argentea* (Pursh.) Nutt. suggests that the contribution of these compounds in the pharmacological activity should be evaluated. So it is recommended as a plant of phytopharmaceutical importance. However further studies will need to be undertaken to ascertain fully its bioactivity.

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