Chemical Composition and Antioxidants of Senecio vulgaris (Asteraceae)

Karzan Omer Qader

Department of Biology, College of science, University of Sulaimani, Iraq.

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ABSTRACT

The study included the antioxidant and chemical compounds of methanolic extract of *Senecio vulgaris* in Iraq. *S. vulgaris* has a rich history in traditional medicine for treating different health problems. The results gas chromatography-mass spectrometry (GC-MS) chromatogram of *S. vulgaris* extract showed 19 peaks, which indicates the presence of 19 compounds (phytochemical constituents). Most of the chemical components extracted from *S. vulgaris* include oleic acid (40.46%), n-Hexadecanoic acid (23.40%), 5,9-dimethyl-2-(1-methylethyl)-1-cyclodecanone (6.52%), octadecanoic acid (3.57%) and 1,2-benzenedicarboxylic acid, bis(2-methylpropyl) ester (3.27%).

The antioxidant activity of overall methanol extract contents of *S. vulgaris* was determined by adding different concentrations of methanol extract to 2,2-diphenyl-1-picrylhydrazyle (DPPH). The inhibitory activity was evaluated by using five different concentrations of methanol extract of *S. vulgaris*. The results showed that the antioxidant activity in concentration 3 μ L/mL was 41.23%, while the highest antioxidant activity was 98.33% in 25 μ L/mL compared with vitamin C, which was 16% in 3 μ L/mL and recorded 91.97% in 25 μ L/mL. The total antioxidant capacity of *S. vulgaris* methanol extract was evaluated as vitamin C equivalents per gram. The DPPH radical scavenging potency with a minimum IC50 value in *S. vulgaris* was 5.64 μ L/mL, while vitamin C was 3.09.

Keywords: Antioxidant, GC-MS analysis, Senecio vulgaris, Soxhlet.

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INTRODUCTION

Senecio vulgaris sometimes referred to as "common groundsel," was also known as Sheikh Al-Rabee and AL-Kurresa in Iraq. The genus Senecio belongs to the family Asteraceae and it includes more than 1500 species.¹ S. vulgaris is known for its adaptability and ability to grow under a variety of environments; it is native to tropical regions, Mediterranean Europe, temperate Asia, and North Africa, and its habitats include gardens, waste sites, roadsides, and cultivated fields and is now widespread worldwide through human activity.^{2,3} The plant is endemic in Iraq which is distributed in lower Mesopotamia- Central alluvial plain district and eastern alluvial district.⁴ S. vulgais is a winter or summer annual weed and reproduces solely through seeds. The true leaves are 20 to 100 mm long and 5 to 45 mm wide, arranged alternately, and have oblong, blunt lobes with coarse and irregular teeth.^{3,5} The stems can be 100 to 700 mm tall, dark green in color, hairless, and hollow. They may stand erect or climb and have a smooth and fleshy texture. In Iraq, the plant's life cycle spans 6 to 8 months, starting from November in the first year and ending in May of the second year.⁶

S. vulgaris has been used historically to treat a range of medical conditions, including intestinal worms and toothaches, amenorrhea, and dysmenorrhea. It was known to have diaphoretic, antiscorbutic, purgative, diuretic, and anthelmintic properties. Additionally, the plant was utilized in traditional medicine to address women's disorders and regulate menstrual cycles. Externally, it was applied in compresses to relieve joint inflammation, treat boils, and help with foot conditions in diabetic patients.^{6,7} Numerous research works have reported the chemical composition of solvent extracts from S. vulgaris, indicating the presence of bioactive substances such as flavonoids. This study provides insight into its unique phytochemical profile. Furthermore, studies on the antibacterial activity of this plant may provide valuable information about its potential therapeutic applications against various pathogens.3

Natural products derived from plants have demonstrated promising antimicrobial activities against various pathogens, such as bacteria, fungi, and viruses. In this context, the research's findings regarding the antimicrobial activity of *Senecio vulgaris*, specifically against gram-positive bacteria *Bacillus subtilis* and *Staphylococcus aureus*, hold significant value. This exploration of *S. vulgaris*'s antimicrobial potential contributes to the ongoing efforts to develop new antimicrobial agents and address the challenges posed by antimicrobial resistance.

S. vulgaris L. is regarded as a dangerous plant because it contains many toxic pyrrolizidine alkaloids, which have been isolated from it. Such as alkaloids, flavonoids, and terpenes.⁸⁻¹⁰ This plant is widely recognized for its toxicity. Through phytochemical analysis of the entire plant, a novel coumarin called 3,7-dihydroxy-5,6 dimethoxycoumarin-7-O-β-D-glucoside was isolated, apart from isorhamnetin-3-O-β-Dglucoside isolation and the genus Senecio contains two commonly occurring alkaloids: senecionine and seneciphylline.¹¹ Tundis *et al.*, $(2012)^{12}$ study the chemical composition was characterized by the presence of some monoterpenes (borneol and terpinen-4-ol) and different sesquiterpenes, caryophyllene, ylangene, -humulene, aromadendrene, patchoulene, -patchoulene, -muurolene, nerolidol, -selinene, -cadinol, isobicyclogermacrene, neophytadiene, -copaene, elemene, epi-bicyclosesquiphellandrene, trans-farnesene, -gurjunene, -cadinene, -amorphene and sandaracopimaradiene and found the IC₅₀ was 0.16 mg mL. Four compounds were isolated from S. vulgaris; a new coumarin, 3,7-dihydroxy-5,6 dimethoxycoumarin7-O-β-D-glucoside, and a known flavone: isorhamnetin-3-O-β-D-glucoside from the EtOAc fraction and two known alkaloids of common occurrence in genus Senecio; senecionine and seneciphylline from the CHCl3 fraction.¹¹ The extract from S. biafrae included fourteen different chemicals, the most prevalent of which was 3-buten-2-one, 4-(2,6,6-trimethyl-1-cyclohexen-1-yl), with a peak area of 16.14%. Oleic acid and ficusin, with peak areas of 14.26 and 11.24%, respectively, were two more prevalent components.¹³

Antioxidant activities studied by Tundis *et al.*, $(2012)^{12}$ using two different *in-vitro* assays (DPPH) test and found the plant is rich in toxic pyrrolizidine alkaloids.

Various factors, including geographic location, can affect the chemical composition of plants. Environmental conditions, and extraction methods. Therefore, it is important to study the specific chemical composition of *Senecio vulgaris* in Iraq. So, the study aims to analyze the bioactive chemical composition of *S. vulgaris* from Northern regions of Iraq by GC-MS analysis and assess its antioxidant activity.

MATERIAL AND METHODS

Plant Collection

The whole plant of *S. vulgaris*, except its root was collected through field trips from different regions of Sulaimani city-Iraq in March 2023. The plants were identified according to the flora of Iraq. The plant was cleaned, air- dried at room temperature and then ground using the Moulinex mill. Then keep it in containers with the name of the plant written on it and the time of collection until used.

Extraction of S. vulgaris

Extraction of plant components executed utilizing the soxhlet apparatus with methanol extraction. About 65 gm of *S. vulgaris*

was weighed and packed in a filter bag, then placed in the soxhlet apparatus with 350 mL of 70% methanol and distilled water (150 mL). After 24 hours of completing the extraction process, the methanolic extract was obtained and added into petri dishes and let dry.⁶

Isolation and Diagnosis of Methanol Extract by GC-MC Analysis

GC-MS examination was done in Basrah University, Agriculture College, Iraq using Shimadzu GC-2010 ultra-gas chromatograph. GC oven temperature was set at 40 to 280°C, the rate of 15°C/min. Helium was employed a gas courier; 96.1 kPa inlet pressure and 47.2 cm/s of linear velocity. Column flow was 1.71 mL/min. injector temperature: 280°C injection mode: split. MS scan were conducted with an ion source temperature of 200°C interface temperature of 280°C with 0.70 + 0.10 kV detector gain. About 1250 of scan speed, starting at 50 m/z, and then raised to end 600 m/z.¹⁴ The components of the Senecio vulgaris were identified by comparing the spectra with those of known compounds stored in the NIST library (2005).¹⁵ The components of the phytochemicals were identified and verified by looking at their molecular formula, peak area, and retention time (Figure 1).

Determination of Antioxidant Activity

Using DPPH assay to determine the *S. vulgaris* antioxidant activity (Blois, 1958)¹⁶ method with some modifications. Various concentrations from *S. vulgaris* methanol extract was prepared 3, 6, 15, 20 and 25 μ L/mL diluted with methanol. 0.004 mg from A solution of DPPH was made in methanol (100 mL). The samples were incubated for half an hour at room temperature and in the dark before the examination process using a spectrophotometer for 30 minutes at ambient temperature with a 517 nm wavelength. DPPH (50 μ L/mL) was used as the control, vitamin C. Then the percentage of antioxidant activity was calculated according to the following equation:

Antioxidant activity (Inhibition) % = [(A control- A sample) / A control] x 100

A control = absorbance of the control reaction

A sample = absorbance in the presence of extract.

RESULTS AND DISCUSSION

Chemical Composition of S. vulgaris

The GC-MS chromatogram of *S. vulgaris* extract (Figure 1; Table 1) showed 19 compounds (phytochemical constituents). Most of the chemical components from *S. vulgaris* extract include oleic acid (40.46%), n-hexadecanoic acid (23.40%),5,9-dimethyl-2-(1-methylethyl)-1-cyclodecanone (6.52%), octadecanoic acid (3.57%) and 1,2-benzenedicarboxylic acid, bis(2-methylpropyl) ester (3.27%). Our results agreed with^{13,17,18} by found n-hexadecanoic acid, 9-octadecenoic acid (Z)-, methyl ester, oleic acid and linolic acid. Our results agreed with Omotehinwa *et al.*, (2023)¹³ that the main compound in *S. vulgaris* was oleic acid, but the percentage was higher at 4.46%.

Table 1: Chemical constituents of 5. vulgaris extract by using GC-MS analysis				
Peak	Chemical constituents	Formula	RT	Content (%)
1	Diethyl phthalate	$\mathrm{C}_{12}\mathrm{H}_{14}\mathrm{O}_{4}$	15.022	0.85
2	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester	$\mathrm{C_{16}H_{22}O_{4}}$	19.618	3.27
3	Neophytadiene	$C_{20}H_{38}$	19.729	1.90
4	9,12,15-octadecatrienoic acid, (Z,Z,Z)- Linolenic acid	$C_{18}H_{30}O_2$	20.557	0.89
5	Palmitoleic acid	$C_{16}H_{30}O_2$	20.642	2.55
6	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	20.858	23.40
7	13-Octadecenoic acid, methyl ester	$C_{19}H_{36}O_2$	21.944	1.75
8	9-Octadecenoic acid (Z)-, methyl ester	$C_{19}H_{36}O_2$	22.000	1.72
9	9-Octadecenoic acid, (E)-	$C_{18}H_{34}O$	22.104	3.01
10	Oleic Acid	$C_{18}H_{34}O_2$	22.252	40.46
11	Octadecanoic acid	$C_{18}H_{34}O_2$	22.423	3.57
12	Hexadecanoic acid, butyl ester	$C_{20}H_{40}O_2$	22.595	2.56
13	(9E,11E)-Octadecadienoic acid	$C_{18}H_{32}O_2$	22.771	0.65
14	Glycidyl palmitate	$C_{19}H_{36}O_{3}$	23.261	0.92
15	Dodecane, 1,1'-oxybis-	$C_{24}H_{50}O$	23.536	2.12
16	5,9-Dimethyl-2-(1-methylethyl)-1-cyclodecanone	$C_{15}H_{28}O$	23.625	6.52
17	Octadecanoic acid, butyl ester	$C_{22}H_{44}O$	23.922	1.61
18	9,12-Octadecadienoic acid (Z,Z)-, 2,3-dihydroxypropyl ester	$C_{21}H_{40}O_4$	24.274	1.28
19	Glycidyl (Z)-9-Heptadecenoate	$C_{22}H_{40}O_3$	24.422	0.95

Table 1: Chemical constituents of S. vulgaris extract by using GC-MS analysis

The compound n-hexadecenoic acid (palmitic acid act as an antioxidant, hypocholesterolemic, antiinflammatory, nematicide, pesticide, anti-androgenic flavor, hemolytic, 5-alpha reductase inhibitor, potent mosquito larvicide, anticancer and antimicrobial.^{19,20} 9-octadecenoic acid (Z), methyl ester or oleic acid have been found to have hepatoprotective, used as an alpha-glucosidase inhibitor properties, antibacterial, and antimicrobial properties against M. smegmatis, S. aureus and E. coli and apoptotic activities.¹⁹⁻²³ A native of Saudi Arabia, S. glaucus is important as a phenolic profile, cytotoxic activity, and antioxidant.²⁴ The S. rhizomatus extract inhibited the DPPH radical at 92.5%, which may be because it contains phenolic compounds and flavonoids,¹⁷ Sadgrove (2022).²⁵ When S. vulgaris plants' aerial parts were separated and collected into 30 essential oil samples, the primary constituents were α -humulene (57.3%), (E)- β -caryophyllene (5.6%), terpinolene (5.3%), are curcumin (4.3%), and geranyl linalool (3.4%). Malak et al. (2023)¹⁸ sourced S. cruentus DC. Ethanol extracts demonstrated the most potent cytotoxic extract IC_{50} = 7.63 μ g/mL and underwent GC-MS metabolite profiling. We suggest that changes in chemical compound number and amount could be related to environmental or genetic causes or as a result of the plant's varied nutritional and chemical composition.²⁶⁻²⁹

Antioxidant Assays of *S. vulgaris* Extract using DPPH Radical- Scavenging

The *S. vulgaris* extract's components' antioxidant activity was assessed utilizing an activity that scavenges free radicals

(DPPH) by using five different concentrations of the extract. This is done by using the conventional vitamin C complex, the antioxidant action of DPPH methanol extract that was assessed for positive comparison, as shown in (Figure 2). The outcomes of the current investigation and also that of extract concentrations lead to an elevation in the antioxidative effectiveness percentage; the results indicated that 25 μ L/mL concentration showed a radical scavenging activity stronger than lower concentrations (Figure 2). Antioxidant activity was lower for the extract of S. vulgaris at 41.23% at a concentration of 3 µL/mL, while the maximum level of antioxidant activity in 25 µL/mL was 98.33% compared with vitamin C, which was 16% in 3 µL/mL and recorded 91.972% in 25 µL/mL. One possible explanation for the rise in the rate of inhibition is the increase in compound concentration and the presence of several active compounds in the plant, which have an antioxidant effect.^{24,30} The chemical compounds found in plants in low concentrations reduce or stop the oxidation process by multiple mechanisms (Shahidi, 2008).³¹

The total antioxidant capacity of *S. vulgaris* plants was evaluated as Vitamin C equivalent per gram. The total antioxidant capacity of the test samples was calculated using the standard line of vitamin C (y = 2.403+42.561, $R^2 = 0.4437$). The standard calibration curve was used to determine the extract content of the test solutions. The research results indicated that *S. vulgaris* were (y = 2.4678x + 36.06, $R^2 = 0.9818$) (Figure 3). Arroyo-Acevedo *et al.*, (2021)¹⁷ reported that *Senecio* species possesses antibacterial, antioxidative, and anticancer properties.



Figure 1: Chromatogram of chemical compounds of S. vulgaris extract



Figure 2: The percentage inhibition of *S.vulgaris* extract by the antioxidant vitamin C

IC₅₀ Assay

The antioxidant property of various concentrations of S. vulgaris was presented by their IC₅₀ values. All data was compared with the IC50 value of standard Vitamin C, as shown in (Figure 4). The DPPH radical scavenging potency of with a minimum IC₅₀ value in S. vulgaris was (5.64 µL/mL), while Vitamin C was 3.09. The results go in line with.^{11,12} Ethanol extract is an important secondary metabolite, and it is as important as antioxidant, antiinflammatory and anticancer agents. Several studies have demonstrated the efficacy of ethanol extract and its potential to act as an antioxidants, which may be owing to these chemicals' ability to inhibit. The ethanoic extract of S. vulgaris was found IC_{50} be 19.9 μ g/mL. Free radical scavenging activity screening using DPPH assay results showed %inhibition at values of 23.31%.¹¹ Senecio clivicolus wednesday's phytochemical profile and possible antioxidant activity were investigated by Faraone et *al.* (2018), 32 who discovered an IC₂₅ value was four times lower than Ascorbic acid, demonstrating its effectiveness. Salinitro et al., (2020)³³ reported that S. vulgaris has polyphenols and flavonoids that has a good antioxidant. S. glaucus of Saudi



Figure 3: Calibration curve of percentage inhibition of the free radical DPPH by *S.vulgaris* extract



Figure 4: IC₅₀ values of the different methanolic extracts in DPPH scavenging assay

Arabia has antioxidant, cytotoxic activity, and phenolic of with an IC_{50} value of 41.8 µg/mL.²⁴ In contrast, Malak *et al.*, (2023)¹⁸ determined the LD₅₀ of *S. cruentus* ethanol extract as 1.5 g/kg.

CONCLUSION

The research findings highlight the potential of *S. vulgaris* as a source of bioactive compounds with antioxidant properties. The study contributes to the understanding of the phytochemical profile of *S. vulgaris* in Iraq and its potential therapeutic applications. Further research and exploration of this plant species may provide valuable insights for pharmaceutical and agricultural industries, and it has been a good source to produce many modern drugs and drug development.³⁴ *S.vulgaris* has biologically active chemicals of widespread pharmacological and medicinal properties; and the presence of phytochemicals inflorescence of *S.vulgaris* may be responsible for controlling diseases.

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