International Journal of Pharmacy and Pharmaceutical Research (IJPPR)
Volume 30, Issue 8, August 2024 pp 158-163. **ijppr.humanjournals.com** ISSN: 2349-7203

Phytochemical Profile Screening and Assessment of Antioxidant Properties of *Artemisia herba-alba* from Oriental Iraq

Sakeyna Mohammed Mahdi[1], Laith B. Alhusseini[2]*

1. Department of Ecology, College of Science, Kufa University, Najaf, Iraq

Received: 2024-07-01 Revised: 2024-08-05 Accepted: 2024-09-01

ABSTRACT

Artemisia herba-alba is a plant from Waist City-Iraq offering a diverse range of pharmacological, cosmetic, and agro-ecological uses. However, *Artemisia herba-alba* has not been well exploited. So, the present study is to evaluate the antioxidant activities and Phytochemical analysis of extracts. Antioxidant activity was assessed using the Quantification assay. The chemical composition of the extracts was determined by GC-MS. In addition, the total phenol, flavonoid, saponens, tarpenat, and tannin content was determined using colorimetric methods. The results of current study showed that the GS-MS analysis of the ethanolic extract revealed the presence of 12 compounds, Carbamic acid, monoammonium salt , Acetic acid, 2-Propanone, 1-hydroxy-,Benzaldehyde, 2-hydroxy-6-methyl-,3-Deoxy-d-mannoic lactone, Hexanoic acid, 2-ethylhexyl ester, beta.-D-Glucopvranose, 4-O-,beta.-D-, E-4-(3-Hydroxyprop-1-en-I-yl)-2-metho, Nonylamine, N,N-di(allyl)-, 3,5a,9-TrimethvI-2-oxo-2,3,3a, 5,5a,6,7,9a, According to this study, the dominant compounds of *Artemisia herba-alba* are Acetic acid, mercapto (95.64%), Hexanoic acid, 2-ethylhexyl ester (1.67%), Benzaldehyde, 2-hydroxy-6-methyl- (0.77%), and alpha –methyl mannofuranoside (0.42%). Moreover, total content of Total Chlorophyll (0.308 mg/L), Total carotene (84.27 mg/L), and Total protein test (0.472 mg/L) was significantly in the aqueous extracts. The findings show that *Artemisia herba-alba* extract is a plant that can be used as a source of antioxidant.

Keywords: *Artemisia herba-alba*; antioxidant activities; extract; GC-MS analysis

Introduction

Oxidation is a chemical and aerobic reaction process to produce free radicals, ROS are one of the groups that occur in aerobic life as natural properties of partial oxygen derivatives the interaction of which causes cell damage when the antioxidant capacity of the body is exceeded, causing oxidative stress and causing many diseases (1). Redox reactions have a strong connection with energy metabolism, so physiology is indispensable for oxidation and reduction processes. In cells, the process of producing and getting rid of ROS takes place through controlled in oxidation and reduction signals (2). About 250 plant families with a medicinal role, including the Asteraceae, have biologically important properties, such as anti-inflammatory, anti-microbial, anti-cancer, high antioxidant properties, and many other antioxidants and compounds such as phenols, flavonoids, and high activity of DDPH and FTIR (3), in plants, antioxidants are in the form of secondary metabolites known as phytochemicals with properties that protect against diseases by preventing and slowing the oxidation process of other molecules (4,5). Removing the radical intermediates ends Continuous reactions and prevent oxidation reactions (3). These substances are classified as primary components: amino acids, sugars, pyrimidines from nucleic acids, chlorophyll, proteins, and so on (6). The constituent substances are classified as flavonoids and terpenes. Peel phenols. Creminis, saponins, glucose, phenols, and plant steroids are a minor component (7). Recently, many scientists have discovered that natural substances from plants have interesting biochemical and biological properties (8), including antifungal, antioxidant, antidiabetic, anticancer, and anti-inflammatory activities (9).

Consequently, there is a need to find genuine biological solutions, including natural antifungal substances that are safe and beneficial for the environment (10). Indeed, many of these plants (aromatic or medicinal) have biologically interesting properties and are used for a variety of purposes, including medicine, pharmacy, cosmetics, and agriculture (11). In Arabic, *A. herba-alba* is commonly known as “chih” (12). It is renowned for its therapeutic and medicinal virtues. Previous research into the medicinal characteristics of *A. herba-alba* has revealed multiple biological and pharmaceutical interests, including antioxidant (13), and natural antibacterial activities (14). In current sense, the present study is to evaluate the antioxidant activities and Phytochemical analysis of extracts.

International Journal of Pharmacy and Pharmaceutical Research (IJPPR)
Volume 30, Issue 8, August 2024 pp 158-163. ijppr.humanjournals.com ISSN: 2349-7203

Materials and methods

Plant material

The sample of wormwood herb (*Artemisia herba alba* from Family Asreraceae), was collected from Waist governorate/Iraq during 2023, it classified by the assistant prof. Dr. Nisreen Sabbar Hishaim, Faculty of Education for Pure Science, Biology Department/University of Diayala, the plant parts of the herb were taken and cleaned, drying at room temperature for 21 days (Dark condition), and pulverized via mechanical grinder to a fine powder, and kept in a refrigerator at 4C and then placed in paper bags and kept in conditions away from moisture until use.

Extracts preparation

It was extracted using 50 g of dried and finely ground plant into 500 ml of distilled water (ethanol, n-hexane, and ethyl acetate at a ratio of 1:10) (15). Following adding solvents, the boiling flask was placed on the hot plate at 22-26°C for 72 hours. The duration of the solvent extraction was fixed at for 72 hours, 8 hours a day. Subsequently, the extracts underwent filtering and dried after pouring it into petri dishes inside an oven and baking at a temperature of 40 °c until dry.

Phytochemical screening

Phytochemical screening of *A. herba-alba* was carried out according to the standard modalities (16) to determine the following categories: flavonoids, alkaloids, tannins, terpenoids, saponosides, Terpenes, Phenols, Resins and glucosides. Results are read by visual observation of color change or precipitate formation after the addition of specific reagents.

Physiological Indices

Quantification of total pigment, protein content, and total antioxidant content. The antioxidant activity of plant extracts was assessed as the following, Table 1.

Table 1: The antioxidant activity of plant extracts.

Agents	O.D	solution
Chlorophy A	645	acetone
Chlorophy B	663	
Carotens A	424	
Carotenes B	429	
SOD	420	phosphate buffer solution
Ascorbic acid	520	
The Total Proteins	550	
The Total Proteins	555	Blue reagent

- **Total pigments Content**

By mix the 1g of the Plants extract with ml of Aston.

- **Total protein Content**

By mix the 1g of the Plants extract with 10 ml blue reagent.

- **Total antioxidant Content**

By mix the 1g of the Plants extract with10 ml of phosphate buffer solution.

A. Enzymatic Estimation in Plants

The 1superoxide Dismutase (SOD) enzyme activity determination in plants: the reaction mix, consists of 0.5 ml of Pyragallol (0.2 Mm) that absorbs light at 420 nm, 2 ml of tris buffer, and 50 µl crude enzyme extract (17).

International Journal of Pharmacy and Pharmaceutical Research (IJPPR)
Volume 30, Issue 8, August 2024 pp 158-163. ijppr.humanjournals.com ISSN: 2349-7203

B. Non-Enzymatic Estimation in Plants extract

1. Chlorophy A, B Total Content

Through the technique of spectrophotometry in O.D 645,663 respectively, Chloroph A, B concentrations are measured. by filtering a known and specific amount of water from the sample after grinding it in an acetone solution, followed by treatment and analysis. the estimated time for treatment is (1-5) minutes. The detection limit is (0.08 mg/6 liters) using a cell with a size of 1 cm (18).

2. A and B Carotenes Content Estimation in Plants extract

The carotenoid content was calculated using the (19). technique a fresh plant extract weighing 1g was obtained, and 10 ml of acetone with a concentration of 80% was used to homogenize it in a mill and pestle. the findings are given in mg/l fresh weight, and the following equation was used to get the values of A, B, And Total Carotenes Concentration (mg/L).

3. The Total Proteins

The total proteins were isolated and quantified using the methodology described by (20). Estimating the protein content by weight involved crushing 1 g of fresh plant leaves with a pestle in a chilly mortar and 10 ml of a phosphate buffer solution (pH = 5.6). Using a spectrophotometer, the absorbance of protein samples was determined at 555 nm. Using the standard curve for albumin from cows. A standard curve was used to calculate the total protein content.

Gas chromatography-mass spectrometry (GC-MS) analysis

The labs of the Phi nanoscience center, the GC-MS equipment was used for analyzing the chemical components found in the silt, gas chromatograph: Agilent hp-5ms ultra unit (30 M Length X 250 µm Inner Diameter X 0.25 µm Film Thickness), 1 µl Injection Volume, 11.933 Psi Pressure, and GC Inlet Line are the Analytical Column Measurements for The Agilent (7820A) USA GC Mass Spectrometer. Temperature: 250 ˚C, Carrier Gas: He 99.99%, Temperature: 300 ˚C for The Aux Heaters. Using The Past Software to Analyze the Hierarchical Cluster, The Degree of Similarity Between the Tested Species Was Ascertained.

Results and discussion

Phytochemical screening

Qualitative phytochemical analysis was carried out using standard techniques to determine the active metabolites of *A. herba-alba*. The results of detection of the presence and absence of secondary metabolites in the aqueous extract of *A. herba-alba.* are presented in Table 2.

Table 2: Quantitative analysis to detect the main groups of secondary metabolites in the aqueous extract of *A. herba-alba.*

No.	compounds	Detection guide	
1.	Glucosides	+	Reddish brown ring
2.	Terpenes	+	A very clear reddish brown layer
3.	Tri_Tribounds	-	The red color didn't appear
4.	Tannins	+	A very clear greenish _blue color appears
	Ferric chloride Lead acetate	+	The appearance of a gelatinous precipitation layer
5.	Rategenat	+	The appearance of turbidity is very clear
6.	Flavonoids	-	No yellow appeared
	potassium hydroxide sulfuric acid	+	The appearance of a dark brown ring
7.	Saponins	+	The appearance of foam higher than 1cm_3cm
8.	Alkaloids/ formaldehyde	+	The appearance of turbidity when adding the regent
		+	The appearance of a black precipitate
9.	Phenols	+	Appearance of a bluish green color

International Journal of Pharmacy and Pharmaceutical Research (IJPPR)
Volume 30, Issue 8, August 2024 pp 158-163. **ijppr.humanjournals.com** ISSN: 2349-7203

GC-MS identification

The volatile compounds of the *A. herba-alba* plant studied are separated by gas chromatography (GC)), followed by an analysis using mass spectroscopy (MS). The results of this analysis are summarized in Table 3.

Table 3. The results of the *A. herba-alba* plant separated by gas chromatography.

P.K.	RT	Area%	Library/ ID	High	High %
1	0.889	0.36	Carbamic acid, monoammonium salt	13680	0.60
2	0.958	95.64	Acetic acid, mercapto-	2155714	93.76
3	1.125	0.33	Acetic acid	240329	1.05
4	1.341	0.08	2-Propanone, 1-hydroxy-	71339	0.31
5	7.720	0.77	Benzaldehyde, 2-hydroxy-6-methyl-	186856	0.81
6	8.645	0.24	3-Deoxy-d-mannoic lactone	34077	0.15
7	88.848	1.67	Hexanoic acid, 2-ethylhexyl ester	442850	1.93
8	9.021	0.25	beta.-D-Glucopvranose, 4-O-,beta.-D-	75446	0.33
9	9.137	0.42	alpha –methyl mannofuranoside	92444	0.40
10	9.579	0.11	E-4-(3-Hydroxyprop-1-en-I-yl)-2-metho	72866	0.32
11	9.643	0.07	Nonylamine,N,N-di(allyl)-	36236	0.16
12	11.829	0.110.07	3,5a,9-TrimethvI-2-oxo-2,3,3a, 5,5a,6,7,9a	44934	0.20

Chromatogram showed a peak 3 with retention time of 1.125 min which is corresponding to the molecular ion peak at m\e 136. The Identification of the compound was performed by comparing their mass spectra with a database library of National Institute of Standard and Technology (NIST08) and also compared with the available references. Chromatogram showed a peak 6 with retention time of 8.645min which is corresponding to the molecular ion peak at m\e 134. The Identification of the compound was performed by comparing their mass spectra with a database library of National Institute of Standard and Technology (NIST08) and also compared with the available references.

Chromatogram showed a peak 7 with retention time of 8.848 min which is corresponding to the molecular ion peak at m\e 154. The Identification of the compound was performed by comparing their mass spectra with a database library of National Institute of Standard and Technology (NIST08) and also compared with the available references. Compounds variability are coupled with environmental acclimatization and play vital biological roles. Several factors, such as environmental and edaphic conditions, geographical regions, season of collection, harvesting time, genotype, and ecotype influence the quantitative and qualitative composition (21,22). These chemical composition differences could be due to developmental and environmental factors that affect plant metabolism. Climate change may be to blame for the differences in chemical composition. When comparing studies, however, the following factors were accounted for: genotypic and environmental differences within species, sample extraction time, and the extraction technique used to obtain the ***Artemisia herba-alba*** extract, the chemical variability was recorded by sampling the plants in different regions, with differing climates and soil types and compositions, which makes it hard to distinguish accurately between genetic and environmental factors as causing the composition variability (23). The phytocompounds analyzed via GC-MS in previous studies are comparatively less and totally different than the phytocompounds in the present study, which may be due to different solvent nature, compounds' solubility and polarity, extraction method, different growing locations and climate of the plant, and time of harvest (mature or immature).

Antioxidant Activity

Based on the results obtained, it is evident that each extract exhibits an anti-free radical effect that is reliant on the administered dose (Table 4). All fractions in this study showed antioxidant effects, although the antioxidant strength varied as different water compounds have different redox properties, which make them reducing agents, hydrogen donors, and singlet oxygen quenchers.

Table 4: Antioxidant activity of plant extracts.

No.	Tests	Plant Extracts
1.	Chlorophyll a	0.231 mg/L
2.	Chlorophyll b	0.094 mg/L
3.	Total Chlorophyll	0.308 mg/L
4.	α carotene	45.063 mg/L
5.	βcarotene	39.26 mg/L
6.	Total carotene	84.27 mg/L
7.	Proline	47.118 mg/L
8.	Ascorbic acid	0.836
9.	(SOD) enzyme activities	57.18 mg/unit of enzyme* sec
10.	Total protein test	0.472 mg/L
11.	Antioxidant Scavenging %	80.074%

Conclusions

The present study was carried out on the *A. herba-alba* plant from Waist–Iraq Phytochemical evaluation of *Artemisia herba-alba* it proved to contains secondary metabolite like Glycosides, Terpenes, Tannins, Resins, Flavonoid, Saponins, Alkaloids, Phenols. Evaluation of extract of *Artemisia herba-alba* by GC/MS quantitatively and qualitatively reveled different active compounds it proved antioxidant.

Acknowledgements We are grateful for the useful comments and suggestions from anonymous referees.

Ethical approval: The conducted research is not related to either human or animal use.

References

1. Medrano-Macías J, Flores-Gallegos AC, Nava-Reyna E, Morales I, Tortella G, Solís-Gaona S, Benavides-Mendoza A. Reactive Oxygen, Nitrogen, and Sulfur Species (RONSS) as a Metabolic Cluster for Signaling and Biostimulation of Plants: An Overview. Plants (Basel, Switzerland). 2022;11. doi: 10.3390/plants11233203. Cited: in: : PMID: 36501243.
2. Lima JT, Almeida J, Mota KSL, Lúcio A, Câmara CA, Barbosa-Filho JM, Silva BA. Selective spasmolytic effect of a new furanoflavoquinone derivative from diplotropin on guinea-pig trachea. J Chem Pharm Res. 2011;3:249–258.
3. Nwozo OS, Effiong EM, Aja PM, Awuchi CG. Antioxidant, phytochemical, and therapeutic properties of medicinal plants: A review. Int J Food Prop. 2023;26:359–388.
4. Coulibaly AY, Hashim R, Sulaiman SF, Sulaiman O, Ang LZP, Ooi KL. Bioprospecting medicinal plants for antioxidant components. Asian Pac J Trop Med. 2014;7:S553–S559.
5. Goodarzi S, Rafiei S, Javadi M, Khadem Haghighian H, Noroozi S. A review on antioxidants and their health effects. J Nutr Food Secur. 2018;3:106–112.
6. Awuchi CG, Twinomuhwezi H. The medical, pharmaceutical, and nutritional biochemistry and uses of some common medicinal plants. Med Aromat plants world. 2021;1–32.
7. Zahnit W, Smara O, Bechki L, Bensouici C, Messaoudi M, Benchikha N, Larkem I, Awuchi CG, Sawicka B, Simal-Gandara J. Phytochemical profiling, mineral elements, and biological activities of Artemisia campestris L. grown in Algeria. Horticulturae. 2022;8:914.
8. Bouyahya A, Abrini J, Bakri Y, Dakka N. Screening phytochimique et évaluation de l'activité antioxydante et antibactérienne des extraits d'Origanum compactum. Phytothérapie. 2017;15:379.
9. Mrabti HN, Doudach L, Kachmar MR, KHALI Z, MRABTI NN, BENRAHOU KB, HARRAQUI K, ZENGİN G, BOUYAHYA A, FAOUZI MEA. Phenolic content, antibacterial, antioxidant, and toxicological investigations ofErodium guttatum (Geraniaceae) collected from the Northeast of Morocco. Turk J Botany. 2021;45:739–749.
10. Talibi I, Askarne L, Boubaker H, Boudyach EH, Oumar AA Ben. In vitro and in vivo antifungal activity of eight medicinal plants against citrus sour rot agent Geotrichum candidum. Acta Hortic. 2015;1065:1563–1570.
11. Elshafie HS, Camele I, Mohamed AA. A comprehensive review on the biological, agricultural and pharmaceutical properties of secondary metabolites based-plant origin. Int J Mol Sci. 2023;24:3266.
12. Bertella A, Benlahcen K, Abouamama S, Pinto DCGA, Maamar K, Kihal M, Silva AMS. Artemisia herba-alba Asso. essential oil antibacterial activity and acute toxicity. Ind Crops Prod. 2018;116:137–143.

13. Ouchelli Y, Dahmani N, Addi Y, Hechiche N, Baaliouamer A. Chemical characterization of volatile extract of Artemisia herba-alba and study of its antioxidant, antimicrobial and antifungal activities and its inhibitionory effect on corrosion of aluminum in hydrogen chloride solution. J Microbiol Biotechnol food Sci. 2022;11:e4889–e4889.
14. Mrabti HN, El Hachlafi N, Al-Mijalli SH, Jeddi M, Elbouzidi A, Abdallah EM, Flouchi R, Assaggaf H, Qasem A, Zengin G. Phytochemical profile, assessment of antimicrobial and antioxidant properties of essential oils of Artemisia herba-alba Asso., and Artemisia dracunculus L.: Experimental and computational approaches. J Mol Struct. 2023;1294:136479.
15. Burman S, Bhattacharya K, Mukherjee D, Chandra G. Antibacterial efficacy of leaf extracts of Combretum album Pers. against some pathogenic bacteria. BMC Complement Altern Med. 2018;18:1–8.
16. Dib MEA, Allali H, Bendiabdellah A, Meliani N, Tabti B. Antimicrobial activity and phytochemical screening of Arbutus unedo L. J Saudi Chem Soc. 2013;17:381–385.
17. Marklund S, Marklund G. Involvement of the superoxide anion radical in the autoxidation of pyrogallol and a convenient assay for superoxide dismutase. Eur J Biochem. 1974;47:469–474.
18. Goodwin CW. Process interaction and soil temperature near Point Barrow, Alaska. University of Michigan; 1976.
19. Maclachlan S, Zalik S. Plastid structure, chlorophyll concentration, and free amino acid composition of a chlorophyll mutant of barley. Can J Bot. 1963;41:1053–1062.
20. Bishop, M., Dben-Von Laufer, J., & Fody E. Thirty-three Contributors. Clinical Chemistry Principles, Procedures & Correlations,. 1985;181–182.
21. Morshedloo, M. R., Salami, S. A., Nazeri, V., Maggi, F., & Craker L. Essential oil profile of oregano (Origanum vulgare L.) populations grown under similar soil and climate conditions. Ind Crops Prod. 2018;183–190.
22. Karimi, D., Dou, H., Warfield, S. K., & Gholipour A. Deep learning with noisy labels: Exploring techniques and remedies in medical image analysis. Med Image Anal. 2020;101759.
23. Sadeh, N., Spielberg, J. M., Logue, M. W., Hayes, J. P., Wolf, E. J., McGlinchey, R. E., Milberg, W. P., Schichman, S. A., Stone, A., & Miller MW. Linking genes, circuits, and behavior: network connectivity as a novel endophenotype of externalizing. Psychol Med. 2019;49:1905–1913.

How to cite this article:
Laith B. Alhusseini et al. Ijppr.Human, 2024; Vol. 30 (8): 158-163.

Conflict of Interest Statement: All authors have nothing else to disclose.