PLANT SCIENCE TODAY
Vol 12(4): 1-7
https://doi.org/10.14719/pst.9594

eISSN 2348-1900

RESEARCH ARTICLE

Isolation and characterization of protocatechuic acid from *Pterospermum acerifolium* (L.) (F. Sterculiaceae) cultivated in Iraq

Duha Ahmed Abdullah* & Amjed Haseeb Khamees*

Department of Pharmacognosy and Medicinal Plants, University of Baghdad / College of Pharmacy, Baghdad 10001, Iraq

*Correspondence email - doha.ahmed2200@copharm.uobaghdad.edu.iq, amjed.haseeb.khamees@gmail.com

Received: 23 May 2025; Accepted: 15 July 2025; Available online: Version 1.0: 05 November 2025; Version 2.0: 18 November 2025

Cite this article: Duha AA, Amjed HK. Isolation and characterization of protocatechuic acid from *Pterospermum acerifolium* (L.) (F. Sterculiaceae) cultivated in Iraq. Plant Science Today. 2025; 12(4): 1-7. https://doi.org/10.14719/pst.9594

Abstract

*Pterospermum acerifoliu*m is a perennial plant known as the dinner plant tree, belonging to the Sterculiaceae family. All parts of the plant are rich in phytochemical compounds, which are known for their various pharmacological properties. This study aimed to identify, isolate and structurally elucidate bioactive polyphenolic compounds-specifically protocatechuic acid-from *Pterospermum acerifolium* cultivated in Iraq, using a combination of chromatographic and spectroscopic techniques. Initially, the removal of fat and wax from plant material was achieved through maceration in n-hexane solvent for three days, followed by extraction using a Soxhlet apparatus with 85 % methanol. The extract was subsequently partitioned with ethyl acetate, yielding 2.6 g of the resulting fraction. The ethyl acetate fraction was analyzed using High-Performance Liquid Chromatography (HPLC) to identify protocatechuic acid. A sharp peak appeared at a retention time of 9.00 min, corresponding to that of the standard protocatechuic acid analyzed under the same conditions. Confirmation of the compounds' identity was further supported by the spiking technique, in which the addition of the reference standard to the sample resulted in an enhanced peak at 9.09 min, confirming the presence of protocatechuic acid. Structural elucidation of the isolated compound was carried out using complementary spectroscopic techniques. FTIR analysis displayed distinct absorption bands indicative of functional groups characteristic of protocatechuic acid (3335, 1675, 1524, 1423, 1302) cm^{-1}. Additionally, LC-MS/MS analysis yielded molecular ion signals and fragmentation patterns that aligned with those of the authentic standard, further confirming the compounds' identity, m/z (154, 137, 109, 93 and 77). The analytical results confirmed the presence of protocatechuic acid in the ethyl acetate fraction. Given its known pharmacological significance, this compound is likely a key contributor to the therapeutic potential of *Pterospermum acerifolium*. These findings offer a valuable foundation for phytochemists and pharmacologists interested in isolating and developing novel bioactive agents from this medicinal plant.

Keywords : FTIR; HPLC; LC-MS/MS; phenolic compound; protocatechuic acid

Introduction

Medicinal plants are used to treat human diseases and have garnered significant interest, largely due to experiments and the transmission of knowledge across generations. However, the global use of medicinal plants faces numerous challenges, including concerns about their safety, unverified claims made by sellers and inadequate quality control. Despite these issues, early human populations were able to document and expand their understanding of natural medicines and edible substances. This eventually contributed to the advancement of new technologies and the discovery of novel drugs (1). Wild plants exhibit greater genetic diversity compared to cultivated ones, owing to their ability to withstand environmental stress and their higher nutritional value, which leads to notable differences in their morphology and chemical composition. Both wild and cultivated plants have been studied for their potential in treating various health conditions (2). Plants can synthesize a wide range of organic compounds, which are generally categorized into primary and secondary metabolites. Primary metabolites are essential for fundamental physiological processes such as growth and Development. In contrast, secondary metabolites are not directly required for these basic functions but are vital for the plants' adaptation and defence mechanisms against various abiotic and biotic stressors. These secondary metabolites are recognized as rich sources of bioactive compounds, with growing evidence supporting their significant health benefits. Therefore, advancing our knowledge of these plant-derived bioactive is crucial for both scientific and therapeutic applications (3).

Pterospermum acerifolium (L.) Willd, a member of the family Sterculiaceae, is a widely recognized deciduous tree commonly referred to as the 'Dinner Plate Tree' or 'Muchukunda'. This species is characterized by its substantial size, reaching heights of up to 24 m and a trunk girth of approximately 2.5 m, with a straight, unbranched bole extending up to 12 m (4). Research indicates that *Pterospermum acerifolium* in terms of its traditional and ethnomedicinal uses, chemical composition and

a comprehensive overview of its pharmacological properties and clinical applications. The aim is to encourage further research on this plant, which may lead to the improvement of novel pharmaceutical preparations with developed therapeutic potential and greater cost-effectiveness. All parts of *Pterospermum acerifolium* have a wide range of pharmacological activity. All parts exhibit various pharmacological activities, including antiulcer, analgesic, anti-inflammatory, wound-healing and antioxidant properties (4). The methanolic extract of *Pterospermum acerifolium* shows many chemical compounds, especially phenolic compounds and flavonoids (5). This study is considered the first conducted on *Pterospermum acerifolium* in Iraq, aiming to identify and extract phenolic compounds, such as protocatechuic acid, using various analytical techniques. Protocatechuic acid (PCA), identified chemically as 3,4-dihydroxybenzoic acid, is a naturally abundant phenolic compound found in a range of plant species. It exhibits structural similarity to other prominent antioxidant compounds, such as gallic acid, caffeic acid, vanillic acid and syringic acid, which are widely recognized for their potent antioxidant activities (6). Based on previous studies highlighting the phenolic content of *Pterospermum acerifolium* and the established pharmacological significance of protocatechuic acid (PCA), this research proposes that *P. acerifolium* grown in Iraq possesses measurable and isolatable quantities of PCA. It is further anticipated that this compound can be effectively separated and structurally confirmed using advanced chromatographic and spectroscopic approaches. These findings may reinforce the therapeutic potential of this plant as a natural source of bioactive constituents with medicinal value. The chemical structure of protocatechuic acid is shown in Fig. 1.

Protocatechuic acid has shown promising potential in the field of neurodegenerative disorders. Extensive research has highlighted its ability to exert neuroprotective effects through various mechanisms, including antioxidant, anti-inflammatory and anti-apoptotic activities. PCA has demonstrated its ability to mitigate oxidative stress, reduce neuroinflammation and enhance neuronal survival in several *in vitro* and *in vivo* studies. Further research is needed to fully understand its mechanisms of action and optimize its therapeutic potential (7). Protocatechuic acid attenuated liver injury (decrease AST, ALT, GGT, ALP, bilirubin and increase serum albumin) induced by injection of high doses of cisplatin (10 mg/kg). This improvement in markers of liver damage was associated with marked hepatic antioxidant effects (decreased lipid peroxidation, normalized GSH level and SOD activity and decreased RNS metabolites (NO)). Moreover, protocatechuic acid suppressed the cisplatin-induced elevation in IL-6 and TNF-α levels, as well as the NF-κB p65 subunit (8). Research indicates that PCA demonstrated high antioxidant activity. PCA also showed potential to induce the synthesis of type I collagen in human dermal fibroblasts and skin explants under laboratory conditions. It inhibited MMP-1 secretion from UVA-irradiated human dermal fibroblasts. An *in vivo* study showed that treatment with a lotion containing 0.02 % PCA for 8 weeks significantly reduced the percentage of all skin wrinkle parameters (9). PCAs' anti-asthma properties were investigated in mice that were ovalbumin (OVA) sensitized. PCA treatment reduced the airway hyperresponsiveness to breathed methacholine caused by OVA. PCA also reduced mucus hypersecretion and cell irritation. As a result, PCA may help treat asthma (10).

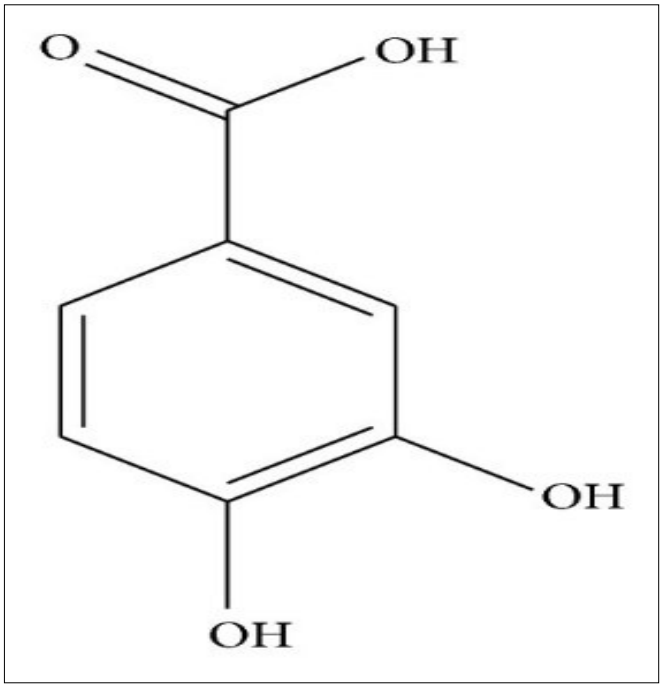

Fig. 1. Chemical Structure of protocatechuic acid.

Proteocatechuic acids' (PCA) chemopreventive qualities include its antioxidant effects, its capacity to chelate metals (particularly in ferroptosis), its ability to induce cell death (including apoptosis, pyroptosis and necroptosis), its anti-inflammatory properties and its beneficial regulatory effects on the p53 protein. Its interference with the three basic stages of chemically induced carcinogenesis highlights its capacity to prevent, inhibit, or reverse tumour formation in addition to exhibiting anti-proliferative, antiangiogenic and antineoplastic effects on various cell lines both *in vivo* and *in vitro* (11). Rats that had stomach ulcers caused by pyloric ligation, oral ethanol or aspirin treatment, or both, were used to study protocatechuic acid ethyl ester. PCA ethyl ester was given intraperitoneally at doses of 30 mg/kg and 60 mg/kg. Significant antiulcer properties were detected 30 min before ulcer induction and the ulcer index was considerably lower than in vehicle control animals. The mode of action of PCA ethyl ester could be attributed to its cytoprotective effects or to fortifying the stomach mucosa, which would improve mucosal defence. Likewise, protocatechuic acid may also possess antiulcer properties (12). Recent research has demonstrated that PCA exhibits antioxidant potential by enhancing the activity of endogenous antioxidant enzymes, including glutathione peroxidase (GSH-PX) and superoxide dismutase (SOD). Additionally, PCA is thought to be an excellent peroxyl radical scavenger in aqueous solutions' polar environment and a reasonably effective anti-radical protector in lipid solutions' nonpolar environment. By boosting GSH-PX and SOD activity, decreasing xanthine oxidase (XOD) and NADPH oxidase (NOX) activity and lowering malondialdehyde (MDA) concentrations, it can mitigate oxidative stress (6).

Materials and Methods

Plant preparation

The root and aerial parts of *Pterospermum acerifolium* from a grove in Baghdad in June 2024. Its identification and authentication was done in the College of Science, University of Baghdad. The plant *Pterospermum acerifolium* was washed and exposed to air at room temperature for a month and then ground in an electronic grinder to a semi-powder.

Extraction by soxhlet and fractionation

250 g of dried plant was weighed and soaked in hexane for three days to remove wax and fat. After that, the degreasing plant was treated with 85 % alcoholic methanol using a Soxhlet apparatus until the colour disappeared. Then, it was filtered and dried by a rotary evaporator machine. The active constituents were separated using solvents of varying polarity-petroleum ether, chloroform and ethyl acetate-applied sequentially in a separatory funnel. The ethyl acetate fraction is dehydrated using anhydrous sodium bisulphate, filtered and then evaporated using a rotary evaporator. It is weighed and set aside for further analysis.

Phytochemical test for phenol compounds

For the Ferric chloride test, take 2 mL of the ethyl acetate fraction and mix it with 5 drops of 5 % $FeCl_3$. The presence of a purple colour or dark blue-green indicates the presence of phenol (13-15).

Identification and isolation of protocatechuic acid by high performance liquid chromatographaphy

High-performance liquid chromatography (HPLC) is a highly accurate and widely employed

analytical technique for both qualitative and quantitative assessment of pharmaceutical products. The improvement and corroboration of HPLC approaches are fundamental to the pharmaceutical industry, as they support various stages of drug discovery, development and production by ensuring the reliability, efficacy and quality of pharmaceutical products (16, 17). High-Performance Liquid Chromatography (HPLC) analysis performed by using a C18-ODS column (250 × 4.6 mm, 5 µm particle size) under both isocratic and gradient elution modes to ensure accurate separation. In the isocratic method, mobile phase consisted of methanol and water in a 1:1 percentage, with the addition of 5 % formic acid. The investigation was carried out at a flow rate of 0.75 mL/min, with an injection volume of 20 µL and finding was conducted at a wavelength of 254 nm. For the gradient elution method, two solvents were used: Solvent A (95 % acetonitrile with 0.01 % trifluoroacetic acid) and Solvent B (5 % acetonitrile with 0.01 % trifluoroacetic acid). The gradient program was as follows: 10 % solvent A from 0 to 5 min, increased to 25 % from 5 to 7 min and then to 40 % from 7 to 12 min, followed by a return to the initial conditions. This method was performed at a flow rate of 1 mL/min, with an inoculation capacity of 100 µL. The discovery of phenolic compounds was approved by using a UV-Visible detector set at 278 nm (18).

Identification and characterization of protocatechuic acid by different techniques

Spiking analysis by HPLC

The spiking method involves adding a known quantity of the standard to the isolated compound and then the mixture is re-examined by HPLC. The isolated compound is identified by comparing its retention time to that of the known standard (19).

Fourier transform-infra red

Since Fourier transform infrared spectroscopy (FT-IR spectroscopy) examines the interaction of molecules and radiation in the infrared portion of the spectrum (IR region = 4000-400 cm^{-1}), it is utilized as a fingerprinting tool in phytochemical research (20). The analysis was done at the Ministry of Science and Technology, Department of Environment and Water.

Liquid chromatography-mass spectroscopy (LC/MS/MS)

LC-MS/MS analysis was performed at the Jordan University of Science and Technology using a Shimadzu system with a C18 column (100 × 4.6 mm, 5 µm) at 35 °C. The injection volume was 5 µL, the flow rate was 1 mL/min and the run time was 25 min. Detection used positive ESI mode (50-800 m/z, 5500 V). The isocratic mobile phase included water with 0.1 % formic acid and a 1:1 mixture of acetonitrile and methanol with 0.1 % formic acid (21).

Results

The quantity and percentage yield of the crude extract obtained from extraction using a Soxhlet apparatus, as well as the n-hexane and ethyl acetate fractions, are illustrated in Table 1 below, where the amount of the ethyl acetate fraction appears 2.6 g. The result of the phytochemical phenols test in the ethyl acetate fraction showed a deep green colour, which indicates the presence of phenols in the plant.

Identification and isolation of protocatechuic acid by using HPLC

The analysis of ethyl acetate fraction proves the presence of some phenolic and flavonoid compounds in *Pterospermum acerifolium* like protocatechuic acid, vanillic acid, gallic acid, quercetin, luteolin and rutin (Fig. 2).

Based on the retention time (9.00), this confirms the presence of protocatechuic acid in this fraction, as shown in Fig. 2, where isolated protocatechuic acid is depicted in Fig. 3 and the protocatechuic acid standard in Fig. 4.

By comparison, the figures below show that the peak of protocatechuic acid in the ethyl acetate fraction (9.00) matches the peak of the isolated compound (9.05) and the protocatechuic acid standard.

Table 1. Quantities and percentages of crude extract and fractions

Name of fraction	Amount (g)	Percent yield %
crude extract by Soxhlet	32 g	12.8 %
Hexane fraction	4 g	1.6 %
Ethyl acetate fraction	2.6 g	1.04 %

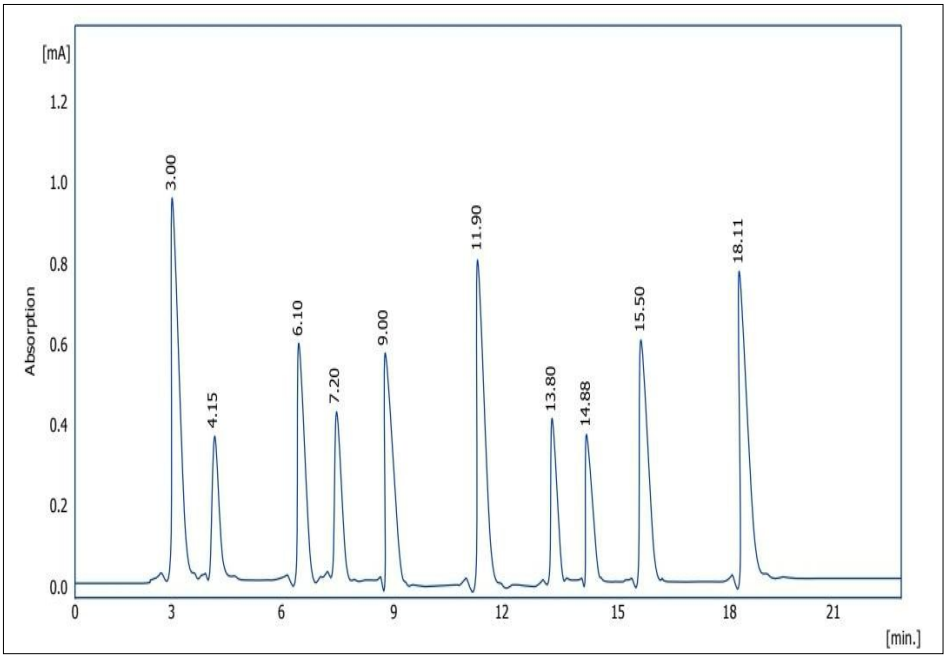

Fig. 2. Ethyl acetate fraction analyzed by HPLC.

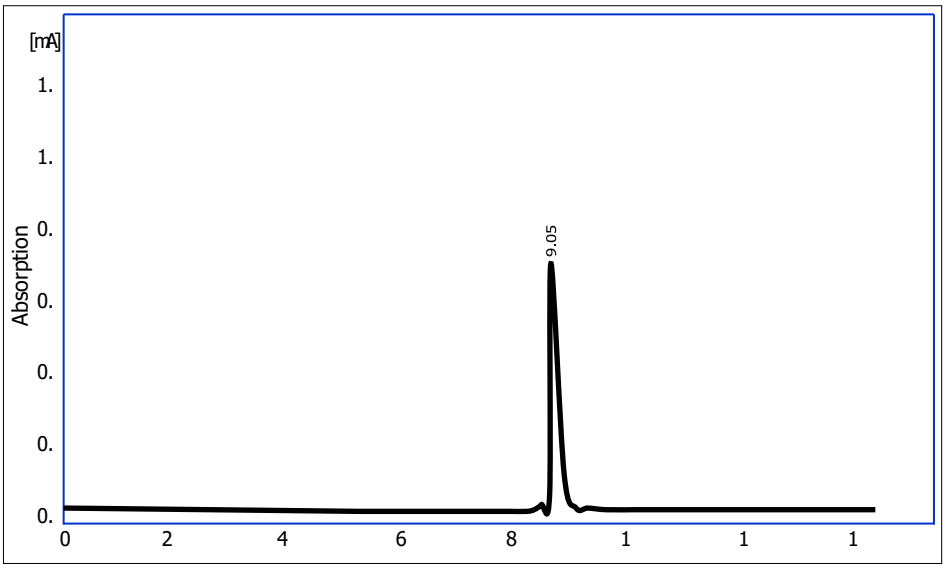

Fig. 3. HPLC for isolated protocatechuic acid.

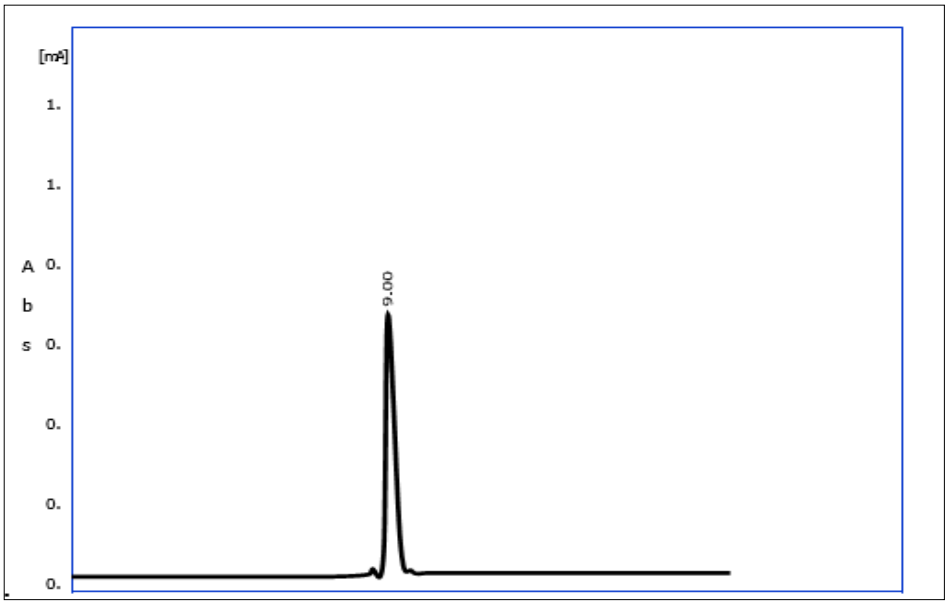

Fig. 4. HPLC chromatogram for protocatechuic acid standard.

Identification and characterization of isolated protocatechuic acid

Spiking by HPLC

By collecting the isolated compound which has retention time (9.05) with standard of protocatechuic acid which has a retention (9.00), an increase in the area peak was observed, it appear at (9.09) as illustrated in Fig. 5.

FTIR (Fourier transform-infra red)

The infrared spectra of protocatechuic acid that was separated from the ethyl acetate portion of the whole *Pterospermum acerifolium* plant are shown in Fig. 6 and Table 2. illustrated value of each peak with its functional group where the peak at 3335 cm^{-1} refer to OH stretching (broad) of phenolic and carboxylic groups, peak at 1675 cm^{-1} refer to C=O stretching of carboxylic acid group (COOH), peak at 1524 cm^{-1} refer to C=C stretching of aromatic benzene ring vibration, peak at 1423 cm^{-1} refer to C-H bending associated with aromatic skeletal vibration and peak at 1302 cm^{-1} refer to C-O stretching of phenolic group supports presence of 3,4-dihydroxy groups.

Liquid chromatography-mass spectroscopy (LC/MS/MS)

An LC/MS/MS study was conducted to detect and characterise isolated protocatechuic acid, where the molecular mass of 154 corresponds to the molecular ion peak of protocatechuic acid.

Most prominent fragment ion was observed at m/z: 137 ($C_7H_5O_3$) by loss of hydroxyl group, m/z: 109 ($C_6H_5O_2$) through decarboxylation, loss of carboxylic acid group, m/z 93 (C_6H_5O) result from breakdown of aromatic and m/z 77(C6H5) resulted from ring cleavage (Fig. 7).

Table 2. FTIR interpretation for isolated proto catechuic acid

Interpretation	FTIR bands
OH stretch	3335
C=O (carboxylic acid group)	1675
C=C (benzene ring)	1524
C-O stretch (phenolic hydroxyl)	1302
C-H bending (aromatic)	1423
O-H bending (out of plane)	765

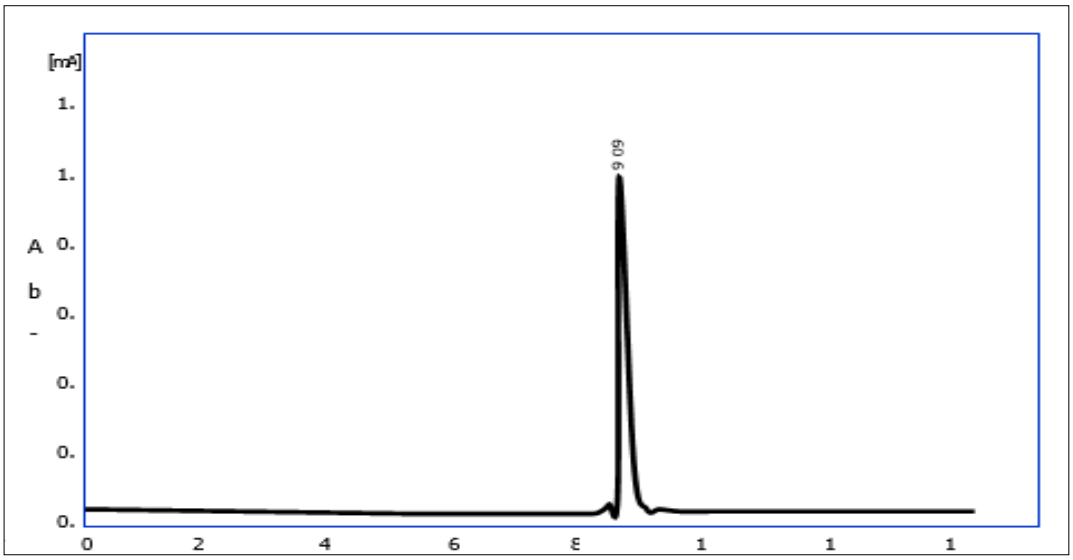

Fig. 5. Spiking analysis (mixing isolated and standard of protocatechuic acid).

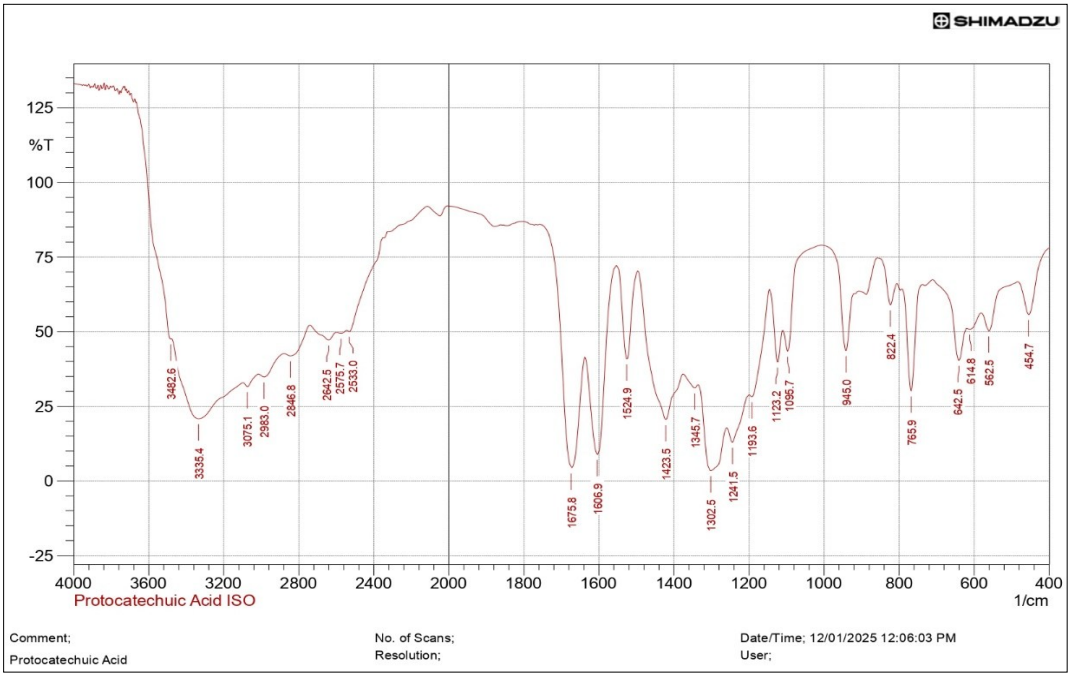

Fig. 6. FTIR for isolated protocatechuic acid.

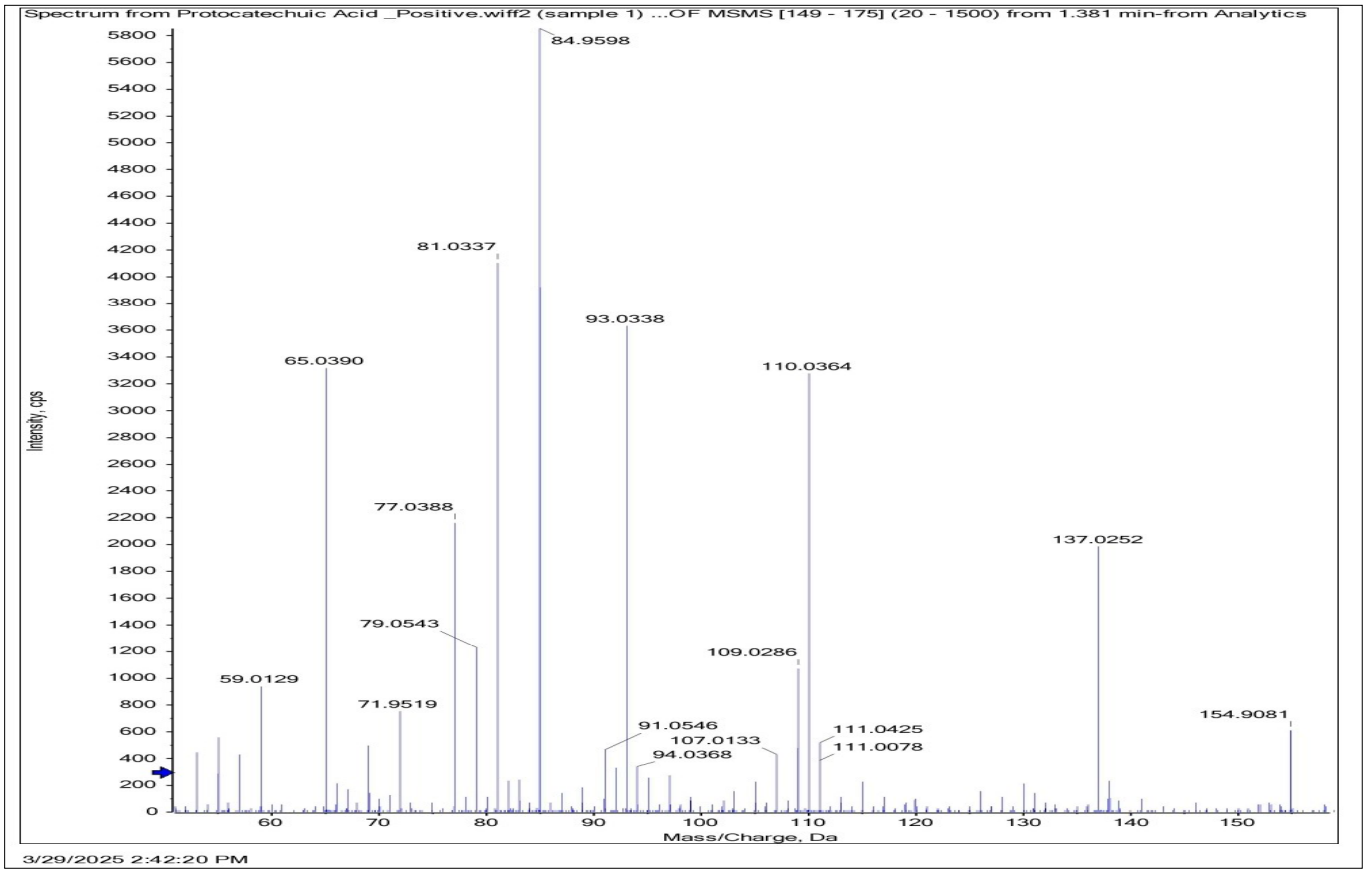

Fig. 7. LC-MS-MS of protocatechuic acid.

Discussion

Medicinal plants are currently essential in treating many diseases and in the discovery of new drugs. *Pterospermum acerifolium* has been shown to contain a variety of polyphenolic components, including flavonoids and phenolic acids, among other phytochemicals. Secondary metabolites, specifically polyphenolic compounds, possess antioxidant properties that enable plants to thrive in harsh conditions (22). In the present study, the ethyl acetate fraction was found to be positive for phenolic compounds during preliminary screening. Spectroscopic techniques were subsequently employed to elucidate the nature of these compounds. Protocatechuic acid is used as a standard reference in later analytical procedures due to its significant antioxidant potential.

Analytical HPLC results confirmed the successful identification of protocatechuic acid in the ethyl acetate fraction by matching its retention time with that of the respective standard under the same conditions. When the standard solution of protocatechuic acid was mixed with the ethyl acetate fraction, a sharp peak appeared at 9.00 min, verifying their presence, as shown in Fig. 4. These results provided a strong basis for proceeding with the isolation of this compound using HPLC under the previously established conditions.

The isolated protocatechuic acid was confirmed by PHPLC analysis, which showed an identical retention time to its standard at 9.00 min. To further validate its identity, spiking experiments were conducted, yielding an increased peak area when the isolated compound was mixed with the standard, as shown in Fig. 5. Moreover, FT-IR analyses provided additional confirmation, the broad O-H stretch (3335 cm^{-1}) and C=O (1675) are clear indicators of hydroxyl and carboxylic groups, on the other hand the aromatic peaks confirm the benzene ring structure and the C-O stretch at 1302 cm^{-1} fits the phenolic oxygen. The observed LC-MS/MS fragmentation pattern of protocatechuic acid, which predominantly exhibits losses of hydroxyl and carboxyl groups before the aromatic ring breaks down, is consistent with its structure. This supports the structural characterization of PCA using LC-MS/MS and validates its identity. The spectral characteristics and fragmentation patterns matched those previously reported for protocatechuic acid. This study was the first to isolate protocatechuic acid compound from the *Pterospermum acerifolium* extract which may demonstrate some of its therapeutic uses such as anticancer as it was confirmed through researchers that protocatechuic acid possesses the ability to inhibition of the generation of free radicals and their ability to scavenge and increase the catalytic activity of endogenous enzymes involved in the neutralization of free radicals (11). Our results confirm the presence of protocatechuic acid, a compound extensively documented in literature for its antioxidant potential. This finding provides a plausible explanation for the traditional use of *P. acerifolium* in managing oxidative stress-related conditions (13).

Conclusion

Pterospermum acerifolium was found to contain a wide variety of polyphenolic chemicals, according to phytochemical research. To fully utilize *P. acerifolium*s' therapeutic potential in Iraq, it is essential to extract and describe these substances due to their significant pharmacological capabilities, particularly in the treatment of various ailments. Protocatechuic acid, which exhibits anti-inflammatory, anticancer and antioxidant properties, was one of the molecules effectively isolated using

HPLC. Its authenticity was verified using a variety of analytical methods, such as FT-IR, LC-MS/MS and HPLC. Interpretation of FT-IR and LC-MS/MS supports successful identification of protocatechuic acid and good agreement with those reported in PCA literature.

Acknowledgements

The authors express their sincere gratitude to the College of Pharmacy, University of Baghdad, for its support and for providing the opportunity to conduct this work.

Authors' contributions

DAA was responsible for data collection, analysis, practical work and writing the manuscript. AHK designed this study and provided final approval and overall supervision of the research.

Compliance with ethical standards

Conflict of interest: The Authors do not have any conflicts of interest to declare.

Ethical issues: None

References

1. Fu XM, Zhang MQ, Shao CL, Li GQ, Bai H, Dai GL, et al. Chinese marine materia medica resources: status and potential. Mar Drugs. 201614(3):46. https://doi.org/10.3390/md14030046
2. Monari S, Ferri M, Salinitro M. Ethnobotanical review and dataset compiling on wild and cultivated plants traditionally used as medicinal remedies in Italy. Plants. 2022;11(15):2041. https://doi.org/10.3390/plants11152041.
3. Khade OS, K S, Sonkar RM. Plant secondary metabolites: extraction, screening, analysis and their bioactivity. Int J Herb Med. 2023;11 (2):1–17. https://doi.org/10.22271/flora.2023.v11.i2a.855
4. Jena J, Verma S, Rai JK, Tripathi R. *Pterospermum acerifolium* Linn: ethnomedicinal uses, phytochemistry and pharmacology: a review. J Pharmacogn Phytochem. 2023;12(1):708–15. https://doi.org/10.22271/phyto.2023.v12.i1f.14617
5. Dhani R. Extraction and chemical investigation of leaves of *Pterospermum acerifolium*. Int J Pharm Res Bio-Sci. 2012;1(6): 1–5.
6. Zhang S, Gai Z, Gui T, Chen J, Chen Q, Li Y. Antioxidant effects of protocatechuic acid and protocatechuic aldehyde: old wine in a new bottle. Evid Based Complement Alternat Med. 2021;2021:6139308. https://doi.org/10.1155/2021/6139308
7. Thapa R, Goyal A, Gupta G, Bhat AA, Singh SK, Subramaniyan V, et al. Recent developments in the role of protocatechuic acid in neurodegenerative disorders. EXCLI J. 2023;22:595. https://doi.org/10.17179/excli2023-5940
8. Habib SA, Suddek GM, Rahim MA, Abdelrahman RS. The protective effect of protocatechuic acid on hepatotoxicity induced by cisplatin in mice. Life Sci. 2021;277:119485. https://doi.org/10.1016/j.lfs.2021.119485
9. Shin S, Cho SH, Park D, Jung E. Anti-skin ageing properties of protocatechuic acid *in vitro* and *in vivo*. J Cosmet Dermatol. 2020;19 (4):977–84. https://doi.org/10.1111/jocd.13086
10. Wei M, Chu X, Guan M, Yang X, Xie X, Liu F, et al. Protocatechuic acid suppresses T cell activation by regulating NFAT and NF-κB transcription factors. Int Immunopharmacol. 2013;15 (4):780. https://doi.org/10.1016/j.intimp.2013.01.010
11. Cadena-Iñiguez J, Santiago-Osorio E, Sánchez-Flores N, Salazar-Aguilar S, Soto-Hernández RM, Riviello-Flores MD, et al. The cancer-protective potential of protocatechuic acid: a narrative review. Molecules. 2024;29(7):1439. https://doi.org/10.3390/molecules29071439
12. Kakkar S, Bais S. A review on protocatechuic acid and its pharmacological potential. Int Sch Res Notices. 2014;2014 (1):952943. https://doi.org/10.1155/2014/952943
13. Hasan HT, Kadhim EJ. Phytochemical investigation of leaves and seeds of *Corchorus olitorius* L. cultivated in Iraq. Asian J Pharm Clin Res. 2018;11(11):408–17. https://doi.org/10.22159/ajpcr.2018.v11i11.28408
14. Al-Jaberi AMZ, Al-Fadal SAM, Abdul-Jalil TZ, Al-Wafi H. HPLC isolation of rutin, hesperidin and quercetin from *Ruta chalepensis* extract growing in Iraq Pharmacogn J. 2023;15(4):606–11. https://doi.org/10.5530/pj.2023.15.127
15. Khamees AH, Fawzi HA, Sahib HB. Phytochemical investigation and assessment of the hypoglycemic activity of two herbal extracts from selected Iraqi medicinal plants in alloxan-stimulated diabetic rats: a comparative study. F1000Res. 2020;9:247. https://doi.org/10.12688/f1000research.22788.1
16. Singh R. HPLC method development and validation—an overview. J Pharm Educ Res. 2013;4(1):26–33.https://doi.org/10.7897/2230-8407.04536
17. Shah BP, Patel MA, Soni ST, Shruti J, Patel NM. Stability indicating HPLC method development: a review. Int J Pharm Sci Res. 2012;3 (9):2978–85. https://ijpsr.com/bft-article/stability-indicating-hplc-method-development-a-review/.
18. Ngamsuk S, Huang TC, Hsu JL. Determination of phenolic compounds, procyanidins and antioxidant activity in processed *Coffea arabica* L. leaves. Foods. 2019;8(9):389. https://doi.org/10.3390/foods8090389
19. Hamad MN. Detection and isolation of flavonoids from *Calendula officinalis* (F. Asteraceae) cultivated in Iraq. Iraqi J Pharm Sci. 2016;25(2):1–6. https://bijps.uobaghdad.edu.iq/index.php/bijps/article/view/343
20. Khamees AH, Kadhim EJ. Isolation, characterization and quantification of a pentacyclic triterpenoid compound, ursolic acid, in *Scabiosa palaestina* L. distributed in the north of Iraq. Plant Sci Today. 2022;9(1):178–82. https://doi.org/10.14719/pst.1398
21. Panchal H, Shah M. Development of a simultaneous LC-MS/MS method for the quantitation of apigenin, luteolin and quercetin in *Achillea millefolium* extract. Pharm Lett. 2017;12:72–86.
22. Abdulhussein AJ, Mutlag SH, Khamees AH, Sahib HB. Evaluation of antiangiogenic and antioxidant activity of *Harpagophytum procumbens* (devil's claw). Drug Invent Today. 2018;10(4):3542–5. https://doi.org/10.36468/pharmaceutical-sciences.469

Additional information

Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

Reprints & permissions information is available at https://horizonepublishing.com/journals/index.php/PST/open_access_policy

Publisher's Note: Horizon e-Publishing Group remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Indexing: Plant Science Today, published by Horizon e-Publishing Group, is covered by Scopus, Web of Science, BIOSIS Previews, Clarivate Analytics, NAAS, UGC Care, etc
See https://horizonepublishing.com/journals/index.php/PST/indexing_abstracting

Publisher information: Plant Science Today is published by HORIZON e-Publishing Group with support from Empirion Publishers Private Limited, Thiruvananthapuram, India.