JETIR.ORG

ISSN: 2349-5162 | ESTD Year : 2014 | Monthly Issue JOURNAL OF EMERGING TECHNOLOGIES AND INNOVATIVE RESEARCH (JETIR)

An International Scholarly Open Access, Peer-reviewed, Refereed Journal

INVESTIGATION OF PHYTOCHEMICALS FROM ACETONE EXTRACT OF SEEDS OF PSORALEA CORYLIFOLIA

Ms. Lalita Kumari, Dr. Mahesh Chandra Sharma

ABSTRACT

Corylin (I), psoralidin (II), isopsoralen (III) and psoralen (IV) were obtained from acetone extract of seeds of *Psoralea Corylifolia* (Fabaceae). The structures of the compounds were confirmed by elemental analysis employing 1H, 13C NMR, IR and MS spectroscopy.

Key words: Psoralea corylifolia, acetone, extract, phytochemicals, chromatography.

INTRODUCTION

Psoralea corylifolia, also referred to as Babchi or Bakuchiol, is a medicinal plant that has long been valued for its curative qualities in traditional medical systems. This herbaceous plant, which is native to India and other areas of Asia, is a member of the Fabaceae family of legumes. Psoralea corylifolia is well-known for its wide range of pharmacological uses and has been used extensively in traditional Chinese medicine, Ayurveda, and other folk medicinal [1]. The plant's unique seeds, called Bakuchiol seeds, are rich in flavonoids, coumarins, and psoralens, among other bioactive substances that support the plant's therapeutic qualities [2]. Psoralea corylifolia has attracted the interest of scientists over time, resulting in an increasing amount of data indicating the plant's possible medicinal uses. Psoralea corylifolia is still being researched in the field of herbal medicine for a variety of purposes, including dermatological applications where its anti-inflammatory and antioxidant qualities are being investigated, as well as its usage in traditional treatments for ailments including vitiligo and skin problems [3]. Fig 1 illustrates Psoralea corylifolia plant image and its potential seeds image

Psoralea corylifolia is a plant that is deeply rooted in traditional medicine. Its seeds contain a wealth of bioactive chemicals that have drawn interest due to their wide range of possible medicinal uses. Psoralen and isopsoralen, two of these substances that are well-known furanocoumarins ^[4] exhibit notable photoactive properties, making them crucial components in the treatment of skin disorders such as psoriasis and vitiligo through phototherapy ^[5]. Bakuchiol, a meroterpene phenol which is abundant in the seeds, has shown to be a complex molecule with antioxidant, anti-inflammatory and anti-cancer activities ^[6].



Fig 1: - (a) Psoralea Corylifolia Plant (b) Seeds

Bavachin, a flavonoid, contributes to the overall medicinal profile of the seeds with its antioxidant and anti-inflammatory effects ^[7]. *Psoralea corylifolia* seeds also yield corylin, psoralidin, neobavaisoflavone, bakuchicin, and bakuchiphysalin ^[8], each presenting distinctive bioactivities such as anti-inflammatory, anti-cancer, and antioxidant actions. Consequently, *Psoralea corylifolia* seeds serve as a source of powerful phytochemicals, motivating continued study into their modes of action and possible uses in contemporary medicine.

Psoralea corylifolia seeds have pharmacological properties due to the combined action of these chemicals, which makes them useful in traditional medicine and encourages investigation into their uses in contemporary medicine. Psoralea corylifolia is a fascinating botanical specimen with a rich history and potential applications in modern healthcare, particularly as the research of alternative medicines gains steam.

MATERIAL AND METHODS

Experiment: - Using a melting points instrument, the compound(s)' melting point was ascertained. Using KBr, IR spectra were captured using the FTIR SHIMADZU 8400S spectrometer. The 1H-NMR and 13C-NMR spectra were recorded in CDCl3 at 300 MHz and 75.5 MHz, respectively, with TMS serving as the internal standard. The FAB gas utilized was argon/xenon, and spectra were recorded with a JEOL SX 102/DA-6000 mass spectrometer.

Collection of Plant material: - In the month of February, Seeds of *Psoralea corylifolia* were purchased from local market named Chiranji lal shop located in Chand Paul of Jaipur city of Rajasthan (India).

Preparation of plant material: -After being washed with tap water, seeds were allowed to dry at room temp. Before being converted into a fine powder using a electric mixer and afterwards being kept in airtight boxes. It was mixed with and refluxed with acetone for 72 hours. Then it was filtered. Filtrate was dried and used for further experiment.

Extraction and Isolation of the compounds: - For 48 hours, 150 g of plant material in the form of seeds was extracted using acetone. When obtained extract was concentrated below abridged force, crude extract was created. Acetone extract was dissolved in the least amount of acetone and adsorbed on silica gel to create a slurry On silica gel, column chromatography was applied to the dried slurry. The subsequent compounds were eluted from the column using a series of solvents in increasing polarity order, and then they were separated, purified, and characterized.

Isolation of Compound -I as Corylin: -Compound-I as corylin was obtained when column was eluted with petroleum ether and ethyl acetate in 7:3 ratio. After removal of solvent, a yellowish amorphous solid mass was obtained which on crystallization with methanol yellowish white crystal of the compound was observed. The melting point of this compound was found to be 229-230 °C. IR V_{max} (KBr): 3235 (OH), 2971, 1622, 1568 (C=C), 1498 cm⁻¹. 1H NMR (300 MHz, DMSO-d6 + CDCl₃), δ 8.16 (s, 1H, H-2), 7.98 (d, 1H, J = 8.7 Hz, H-5), 6.90 (d, 1H, J = 8.8 Hz, H-6), 8.39 (s, 1H, -OH at C-7), 6.83 (s, 1H, H-8), 7.24 (s, 1H, H-2'), 6.76 (d, 1H, J = 8.2 Hz, H-5'), 7.28 (d, 1H, J = 7.9 Hz, H-6'), 1.42 (s, 6H, -CH₃ at C-8'), 5.70 (d, 1H, J = 10.0 Hz, H-9'), 6.38 (d, 1H, J = 9.8 Hz, H-10'). 13C NMR (75.5 MHz, DMSO-d6 + CDCl₃): 102.0 (C-8) 115.0 (C-6) 115.5 (C-10) 116.7 (C-3') 120.5 (C-3) 121.7 (C-5') 124.3 (C-1') 130.7 (C-9') 126.7 (C-10'), 127.0 (C-5), 129.4 (C-2'), 130.8 (C-6'), 152.2 (C-4'), 146.8 (C-2), 157.4 (C-9), 162.5 (C-7), 174.6 (C-4). MS (m/z): 321 [M + H] +, 305, 247, 208, 137. Molecular formula calculated as $C_{20}H_{16}O_4$.

Isolation of Compound -II as Psoralidin: - Compound-II as psoralidin was obtained when column was eluted with n-hexane and ethyl acetate in 4:1 ratio. After removal of solvent, a pale-yellow solid mass was obtained which on crystallization with methanol and ethyl acetate yielded pale yellowish white crystal was observed. It revealed a single spot upon TLC analysis ($R_f = 0.35$) in acetone as a mobile phase. It was discovered that this compound's melting point was 290-291°C. IR (KBr, cm⁻¹): 3448, 3349 (-OH), 1720 (C=O), 1630, 1597, 1577 (C=C). 1H-NMR (300 MHz, DMSO-d6) δ 7.09 (s, 1H, H-5), 6.49 (s, 1H, H-8), 7.31 (s, 1H, H-4'), 6.62 (s, 1H, H-5'), 6.91 (s, 1H, H-7'), 8.12 (s, 2H, -OH at C-7 and C-6'), 3.32 (d, 2H, J = 2.4 Hz, H-1''), 5.82 (t, 1H, J = 9.6, 9.9 Hz, H-2''), 1.75 (s, 6H, H-4'' and H-5''). 13C-NMR (75.5 MHz, DMSO-d6) δ 117.2 (C-3), 160.6 (C-4), 127.5(C-5), 120.2(C-6), 156.4(C-7), 107.0(C-8), 148.0(C-9), 119.9(C-10), 156.3(C-2'), 114.6(C-3'), 121.0(C-4'), 108.4(C-5'), 151.4(C-6'), 97.0(C-7'), 21.5(C-1''), 115.9(C-2''), 132.7(C-3''), 17.9(C-4''), 22.8(C-5''), 157.5(C=O). MS m/z: 336, 281, 267, 250. Molecular formula calculated as $C_{20}H_{16}O_5$.

Isolation of compound-III as isopsoralen: Compound-III as isopsoralen was obtained when column was eluted with n-hexane and acetone in 4:1 ratio. After removal of solvent, a white amorphous solid mass was obtained which on crystallization with methanol and n-hexane white crystal of the compound was observed. The melting point of this compound was found to be 133-134 °C. IR V_{max} (KBr): 1712 (C=O), 1616 (C=C), 833, 743 cm⁻¹. 1H-NMR (300 MHz, CDCI₃) δ: 6.37 (d, 1H, J = 9.6 Hz, H-3), 7.79 (d, 1H, J = 9.5 Hz, H-4), 7.36 (d, 1H, J = 8.4 Hz, H-5), 7.42 (d, 1H, J = 8.7 Hz, H-6), 7.68 (d, 1H, J = 2.1 Hz, H-2'), 7.12 (d, 1H, J = 1.9 Hz, H-3'). 13C-NMR (75.5 MHz, CDCI₃) δ: 162.5 (C=O), 115.3 (C-3), 145.2 (C-4), 124.2 (C-5), 109.5 (C-6), 158.6 (C-7), 118.1 (C-8), 149.6 (C-9), 114.4 (C-10), 147.1 (C-2'), 105.3 (C-3'). MS m/z: 186[M]+, 158, 130, 102. Molecular formula calculated as $C_{11}H_6O_3$.

Isolation of compound-IV as psoralen: Compound-IV as psoralen was obtained when column was eluted with n-hexane and acetone in 3:2 ratio. After removal of solvent, a yellowish amorphous solid mass was obtained which on crystallization with methanol yellowish white crystal of the compound was observed. 155-156 °C. IR V_{max} (KBr) 3156, 3123 (furan), It was discovered that this compound's melting point was 1718 (C=O), 1633, 1577 (C=C) cm⁻¹; 1HNMR (300 MHz, CDCI₃) δ : 6.37 (d, 1H, J = 9.5 Hz, H-3), 7.79 (d,1H, J = 9.6 Hz, H-4), 7.67 (s, 1 H, H-5),

7.48 (d, 1H, J = 0.9 Hz, H-8), 7.72 (d, 1H, J = 2.4 Hz, H-2'), 6.81 (d, 1H, J = 2.1 Hz, H-3'). 13C-NMR (75.5 MHz, CDCI₃) δ : 160.8 (C=O), 114.5 (C-3), 144.0 (C-4), 124.8 (C-5), 106.3 (C-6), 156.3 (C-7), 119.8 (C-8), 151.9 (C-9), 115.3 (C-10), 146.8 (C-2'), 99.6 (C-3'). MS m/z: 186[M]+, 158, 130, 102. Molecular formula calculated as C₁₁H₆O₃.

RESULT AND DISCUSSION

Characterisation of Compound -I as *corylin*: The isolated compound was characterized by infrared spectroscopy (IR) and mass spectrometry (MS). The IR spectrum exhibited peaks at 3235 cm⁻¹ (-OH), 2971 cm⁻¹ (C-H), 1622 cm⁻¹, 1568 cm⁻¹, and 1498 cm⁻¹ (C=C), indicating the presence of hydroxyl groups and aromatic rings. The mass spectrum displayed a molecular ion peak at m/z 321 [M + H]+, consistent with the calculated molecular formula of $C_{20}H_{16}O_4$. Fragment ions at m/z 305, 247, 208, and 137 provided additional structural insights.

The compound was analyzed using 1H NMR spectroscopy (300 MHz, DMSO-d6 + CDCl₃) to elucidate its structural features. The spectrum revealed distinct signals providing valuable information about the proton environment in the molecule. The singlet at δ 8.16 ppm (s, 1H, H-2) indicates a proton at position 2, while the doublet at δ 7.98 ppm (d, 1H, J = 8.7 Hz, H-5) and the doublet at δ 6.90 ppm (d, 1H, J = 8.8 Hz, H-6) correspond to protons at positions 5 and 6, respectively, suggesting a potential aromatic system. The singlet at δ 8.39 ppm (s, 1H, -OH at C-7) confirms the presence of a hydroxyl group at position 7. Additional aromatic protons were observed at δ 6.83 ppm (s, 1H, H-8), δ 7.24 ppm (s, 1H, H-2'), δ 6.76 ppm (d, 1H, J = 8.2 Hz, H-5'), and δ 7.28 ppm (d, 1H, J = 7.9 Hz, H-6'). Notably, the methyl group at position 8' is evident from the singlet at δ 1.42 ppm (s, 6H, -CH₃ at C-8'). The coupling patterns observed at δ 5.70 ppm (d, 1H, J = 10.0 Hz, H-9') and δ 6.38 ppm (d, 1H, J = 9.8 Hz, H-10') further contribute to the characterization of the compound's structure. Overall, the 1H NMR data suggests the presence of an aromatic ring system, a hydroxyl group, and a methyl substituent, providing valuable insights into the molecular framework of the analyzed compound.

The structural characterization of the compound was further augmented through 13C NMR spectroscopy (75.5 MHz, DMSO-d6 + CDCl₃). The obtained spectrum provided valuable information about the carbon environments within the molecule. Notably, the signals at δ 102.0 ppm (C-8), δ 115.0 ppm (C-6), and δ 115.5 ppm (C-10) suggest the presence of aromatic carbons, corroborating with the aromatic protons observed in the 1H NMR spectrum. The resonance at δ 116.7 ppm (C-3') and δ 121.7 ppm (C-5') corresponds to aromatic carbons in the substituents, while the signal at δ 124.3 ppm (C-1') indicates the presence of a quaternary carbon in the aromatic ring. The resonances at δ 130.7 ppm (C-9') and δ 126.7 ppm (C-10') correspond to the methyl carbon in the side chain, further supporting the identification of the methyl group observed in the 1H NMR spectrum. The aromatic carbons C-5, C-2', C-6', and C-4' were identified at δ 127.0 ppm, δ 129.4 ppm, δ 130.8 ppm, and δ 152.2 ppm, respectively. Additionally, signals at δ 146.8 ppm (C-2), δ 157.4 ppm (C-9), and δ 162.5 ppm.

The above Compound-1's 1H and 13C NMR spectrum values were found to be similar to those of corylin [5]. Compound-I was recognized as corylin based on the description above and the spectrum information.

Compound-I

Characterization of Compound-II as *psoralidin*: The isolated compound was thoroughly characterized using infrared spectroscopy (IR) and mass spectrometry (MS). In the IR spectrum obtained with KBr, distinct peaks at 3448 cm⁻¹ and 3349 cm⁻¹ were attributed to the stretching vibrations of hydroxyl (-OH) groups, underscoring the presence of alcohol functionality. The absorption band at 1720 cm⁻¹ indicated the presence of a carbonyl group (C=O), further corroborating the IR data. Additionally, peaks at 1630 cm⁻¹, 1597 cm⁻¹, and 1577 cm⁻¹ were indicative of conjugated carbon-carbon double bonds (C=C) within the compound. The mass spectrum (MS) displayed prominent peaks at m/z 336, 281, 267, and 250, aligning well with the calculated molecular formula of $C_{20}H_{16}O_5$.

The isolated compound was subjected to detailed structural analysis using 1H NMR spectroscopy at 300 MHz in DMSO-d6. The spectrum revealed distinct signals, providing key insights into the compound's molecular structure. Notably, the singlets at δ 7.09 ppm (H-5) and δ 6.49 ppm (H-8) indicated the presence of aromatic protons, confirming the existence of an aromatic ring system. Additional aromatic protons were observed at δ 7.31 ppm (H-4'), δ 6.62 ppm (H-5'), and δ 6.91 ppm (H-7'), suggesting the presence of a substituted aromatic ring. The singlet at δ 8.12 ppm accounted for two protons and is attributed to hydroxyl groups at positions C-7 and C-6', providing valuable information about the compound's functional groups. Furthermore, the multiplets at δ 3.32 ppm (d, J = 2.4 Hz, H-1'') and δ 5.82 ppm (t, J = 9.6, 9.9 Hz, H-2'') indicated the presence of aliphatic protons in the side chain. The singlet at δ 1.75 ppm (s, δ H) corresponds to two methyl groups (H-4'' and H-5'') further supporting the overall structural elucidation. The combination of these 1H NMR signals provides a comprehensive view of the compound, confirming the presence of aromatic rings, hydroxyl groups, and aliphatic side chains.

The recorded 13C NMR spectrum (75.5 MHz in DMSO-d6,) exhibited distinctive signals, including resonances at δ 117.2 ppm (C-3), δ 160.6 ppm (C-4), δ 127.5 ppm (C-5), and δ 120.2 ppm (C-6), indicative of the aromatic ring carbons. The carbon resonances at δ 156.4 ppm (C-7) and δ 107.0 ppm (C-8) corroborated with the aromatic protons observed in the 1H NMR spectrum, confirming the aromatic nature of the compound. The peaks at δ 148.0 ppm (C-9) and δ 119.9 ppm (C-10) further supported the presence of aromatic carbons in the molecular framework. Additionally, resonances at δ 156.3 ppm (C-2'), δ 114.6 ppm (C-3'), δ 121.0 ppm (C-4'), and δ 108.4 ppm (C-5') corresponded to the carbons in the substituted aromatic ring. The signals at δ 151.4 ppm (C-6'), δ 97.0 ppm (C-7'), and δ 157.5 ppm (C=O) provided crucial information about the carbonyl carbon and additional aromatic carbons. Furthermore, aliphatic carbons in the side chain were identified at δ 21.5 ppm (C-1"), δ 115.9 ppm (C-2"), δ 132.7 ppm (C-3"), δ 17.9 ppm (C-4"), δ 22.8 ppm (C-5").

On the basis of above discussion this isolated compound was identified as psoralidin [9].

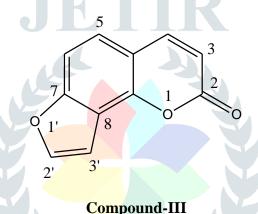
Compound-II

Characterization of compound-III as *isopsoralen*: The IR spectrum exhibited distinctive peaks at 1712 cm⁻¹ (C=O) and 1616 cm⁻¹ (C=C), indicative of a carbonyl group and conjugated double bonds, respectively. Additional bands at 833 cm⁻¹ and 743 cm⁻¹ further supported the presence of specific functional groups. The mass spectrum displayed peaks at m/z 186 [M]+, 158, 130, and 102, aligning precisely with the calculated molecular formula of $C_{11}H_6O_3$. These findings confirm the compound's molecular composition and suggest the presence of a conjugated aromatic system.

The 1HNMR spectrum (300 MHz, CDCl₃) displayed distinctive resonances, including a doublet at δ 6.37 ppm (d, 1H, J = 9.6 Hz, H-3) and doublets at δ 7.79 ppm (d, 1H, J = 9.5 Hz, H-4), δ 7.36 ppm (d, 1H, J = 8.4 Hz, H-5), and δ 7.42 ppm (d, 1H, J = 8.7 Hz, H-6), corresponding to aromatic protons in the molecule. The observed coupling constants (J values) of 9.6 Hz, 9.5 Hz, 8.4 Hz, and 8.7 Hz for H-3, H-4, H-5, and H-6, respectively, provided information about the coupling patterns within the aromatic ring. Additionally, signals at δ 7.68 ppm (d, 1H, J = 2.1 Hz, H-2') and δ 7.12 ppm (d, 1H, J = 1.9 Hz, H-3') represented protons in a substituted aromatic ring.

The recorded 13C-NMR spectrum (75.5 MHz, CDCl₃) revealed resonances at δ 162.5 ppm (C=O), indicating the presence of a carbonyl group in the molecule. The aromatic region displayed peaks at δ 115.3 ppm (C-3), δ 145.2 ppm (C-4), δ 124.2 ppm (C-5), δ 109.5 ppm (C-6), δ 158.6 ppm (C-7), δ 118.1 ppm (C-8), δ 149.6 ppm (C-9), and δ 114.4 ppm (C-10), suggesting the existence of an intricate aromatic ring system. Additionally, signals at δ 147.1 ppm (C-2') and δ 105.3 ppm (C-3') indicated the presence of carbons in a substituted aromatic ring.

Using the explanation from above, this isolated chemical was determined to be isopsoralen [9, 10].



Characterization of compound-IV as psoralen:

In the infrared spectrum (IR) obtained with KBr, characteristic peaks at 3156 cm⁻¹ and 3123 cm⁻¹ are indicative of furan functionalities, and peaks at 1718 cm⁻¹, 1633 cm⁻¹, and 1577 cm⁻¹ suggest the presence of a carbonyl group and conjugated double bonds. Mass spectrometry (MS) data revealed peaks at m/z 186 [M]+, 158, 130, and 102, aligning precisely with the calculated molecular formula of $C_{11}H_6O_3$. The 1H NMR spectrum (300 MHz) in CDCl₃ exhibited resonances, including a doublet at δ 6.37 ppm (d, 1H, J = 9.5 Hz, H-3) and doublets at δ 7.79 ppm (d, 1H, J = 9.6 Hz, H-4), δ 7.67 ppm (s, 1 H, H-5), and δ 7.48 ppm (d, 1H, J = 0.9 Hz, H-8), corresponding to aromatic protons, and signals at δ 7.72 ppm (d, 1H, J = 2.4 Hz, H-2') and δ 6.81 ppm (d, 1H, J = 2.1 Hz, H-3') indicative of a substituted aromatic ring. The 13C NMR spectrum (75.5 MHz, CDCl₃) displayed resonances at δ 160.8 ppm (C=O), δ 114.5 ppm (C-3), δ 144.0 ppm (C-4), and δ 124.8 ppm (C-5), affirming the carbonyl group and aromatic carbon environments. The identified furan moiety and the unique pattern of protons and carbons in the aromatic rings provide a comprehensive understanding of the compound's structure as psoralen [9,10].

a414

Compound-IV

CONCLUSION

New drugs are created using a variety of naturally occurring chemical components that have been extracted from plants. Motivated by these remarkable developments in plant chemistry, an inquiry was thus conducted to identify and characterize the active components of the medicinally important and widely used plants for the well-being of humankind.

REFERENCES

- 1. Nabi, N. G., & Shrivastava, M. (2017). Endangered medicinal plant Psoralea corylifolia: Traditional, phytochemical, therapeutic properties and micropropagation. *Pharmaceutical and Biosciences Journal*, 40-46.
- 2.Alam, F., Khan, G. N., & Asad, M. H. H. B. (2018). Psoralea corylifolia L: Ethnobotanical, biological, and chemical aspects: A review. *Phytotherapy Research*, *32*(4), 597-615.
- 3.Li, C. C., Wang, T. L., Zhang, Z. Q., Yang, W. Q., Wang, Y. F., Chai, X., ... & Li, Z. (2016). Phytochemical and pharmacological studies on the genus psoralea: a mini review. *Evidence-Based Complementary and Alternative Medicine*, 2016.
- 4.Mar, W., Je, K. H., & Seo, E. K. (2001). Cytotoxic constituents of Psoralea corylifolia. *Archives of Pharmacal Research*, 24, 211-213.
- 5. Zhao, L., Huang, C., Shan, Z., Xiang, B., & Mei, L. (2005). Fingerprint analysis of Psoralea corylifolia L. by HPLC and LC–MS. *Journal of Chromatography B*, 821(1), 67-74.
- 6.Ozyigit, I. I., Dogan, I., Hocaoglu-Ozyigit, A., Yalcin, B., Erdogan, A., Yalcin, I. E., ... & Kaya, Y. (2023). Production of secondary metabolites using tissue culture-based biotechnological applications. *Frontiers in Plant Science*, 14, 1132555.
- 7. Shaikh, H. S., & Shaikh, S. S. (2021). Babchi (Psoralea corylifolia): From a variety of traditional medicinal application to its novel roles in various diseases: A review. *Asian Journal of Pharmacy and Technology*, 11(3), 238-244.
- 8.Koul, B., Taak, P., Kumar, A., Kumar, A., & Sanyal, I. (2019). Genus Psoralea: a review of the traditional and modern uses, phytochemistry and pharmacology. *Journal of ethnopharmacology*, 232, 201-226.
- 9. Jiangning, G., Xinchu, W., Hou, W., Qinghua, L., & Kaishun, B. (2005). Antioxidants from a Chinese medicinal herb–Psoralea corylifolia L. *Food chemistry*, 91(2), 287-292.
- 10.Liu, R., Li, A., Sun, A., & Kong, L. (2004). Preparative isolation and purification of psoralen and isopsoralen from Psoralea corylifolia by high-speed counter-current chromatography. *Journal of Chromatography A*, 1057(1-2), 225-228.