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PHYTOCHEMICAL STUDY OF HYDROALCOHOLIC EXTRACT OF *Caesalpinia bonducella* LEAVES

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ABSTRACT

Medicinal plants are a primary source of medications and medical supplies. Our work aims to examine the anti-urolithiasis properties of the phytoconstituents in *Caesalpinia bonducella* leaf extracts. Phytochemicals are bioactive substances derived from the plant's secondary metabolites, and all parts of the plant naturally synthesize them. In the Malabar regions, traditional Siddha practitioners treat psoriasis with *Caesalpinia bonducella*, also included in Pakistan's traditional medicine program. As a purgative, tree bark effectively treats kapha and vata disorders, gynecological disorders, skin infections, intestinal issues, abdominal distension, piles, and ulcers. *Caesalpinia bonducella* denotes various parts of the plants that are utilized for multiple purposes, but are explicitly described by the local community as medicine. The seeds are used for their anti-inflammatory and analgesic properties to cure the damaged epithelial cells. Our study described a phytochemical screening of the hydroalcoholic extract of *Caesalpinia bonducella* leaves, traditionally used for psoriasis scar in Southeast Asian Countries. The hydro alcohol extract of *Caesalpinia bonducella* was exposed to primary phytochemical screening by standard procedures. A Soxhlet Extraction was performed in our study using hydro alcohol for 12 hours. The concentrated extract was then analyzed using various analytical techniques, including ultraviolet-visible spectroscopy, gas chromatography, infrared spectroscopy, and high-performance liquid chromatography preparative. This study showed the presence of functional groups characteristic of phytochemicals in the UV-Vis and IR spectroscopy data. The presence of several compounds was identified by the preparative isolation technique.

Keywords: *Caesalpinia bonducella*, Hydroalcohol Extract, Yield Extract, Phytochemical Analysis, Infra-red, Gas Chromatography, Preparative.

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INTRODUCTION

Humans have been using herbal plants as medications since the beginning, although only 5% of the world's plant species have been thoroughly investigated for medical purposes. In industrialized nations, plants constitute the source of more than 30% of prescribed medications.[1] The traditional medical systems of Ayurveda, Unani, Homeopathy, and Siddha mostly rely on natural products. Individuals living in poverty in Asia and Africa's emerging and underdeveloped nations are experiencing health issues linked to eating grains and cereals tainted with mycotoxin. Throughout a patient's life, kidney stone disease, also known as nephrolithiasis, continues to be a substantial health concern for adults. Over 80% of urinary calculi are calcium oxalate combined with calcium phosphate. Urolithiasis is a condition that is extremely common throughout the world.[2]

Nephrolithiasis is currently treated medically; however, it is expensive and has negative effects. Nephrolithiasis therapy involving invasive procedures carries a high risk of serious complications and a financial burden on the healthcare system.[3] A key component of diet and medication, phytochemicals help keep people healthy and prevent disease. Medicinal plants are employed in complicated concoctions to treat

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life-threatening disorders and home remedies for specific conditions.[4] Numerous phytotherapy methods have discussed medicinal plants in treating infectious disorders due to their accessibility, low toxicity, and lack of adverse effects. *Caesalpinia bonducella* (*C. bonducella*) is widely utilized in the traditional Indian medical system, Ayurveda, for its anti-periodic, pyretic, anti-inflammatory, anthelmintic, and malarial qualities.[5] Our main objective is to collect the plant and authentication, followed by extracting the crude from *C. bonducella* by using the Soxhlet apparatus. Subsequently, analyze the extracted compounds using ultraviolet-visible (UV), gas chromatography (GC), high-performance liquid chromatography (HPLC), and infrared (IR) spectroscopy. Moreover, we have examined the traditional medicinal values of *C. bonducella* and the microbiological effects of the isolated compound.

EXPERIMENTAL

Materials and Methods

The raw materials used in our study include *C. bonducella* leaves, which are collected from the Melur village, Madurai district, Tamil Nadu, India. It was authenticated in St. Joseph's College (Autonomous), Tiruchirappalli. Hydroalcohol solution, Hexane, Ethanol, Ammonium acetate, Acetonitrile, Methanol, Dimethylsulfoxide (DMSO), and Nitrogen gas was employed in our study.

General Procedure

Coarse powder of *C. bonducella* leaves was extracted with Hydroalcohol by the hot percolation technique employing a Soxhlet apparatus. The pigment was then removed with hexane. The extraction was conducted for 12 hours. A concentrated extract was obtained by distilling the solvent after extraction. Following vacuum drying of the concentrated extract, the dry extract was kept in an airtight container for additional phytochemical research.[6] The percentage of the extract was calculated for the hydro alcohol extract of *C. bonducella,* and it was exposed to preliminary phytochemical screening by standard procedures.[7]

Determination of UV and IR

Using a blank solution of 70% ethanol to calibrate the spectrophotometer by setting the baseline absorbance to zero at the desired wavelength. Take the diluted sample (0.02%) and place it in a quartz cuvette. Make sure the cuvette is clean and dry. Place the cuvette in the sample holder of the spectrophotometer and record the absorbance at the maximum wavelength. Plot a graph of absorbance versus concentration of the samples to obtain the calibration curve. Determine the maximum absorption wavelength for the *C. bonducella* plant extract by analyzing the UV absorption spectra obtained from the spectrophotometer.[8]

In IR spectroscopy, isopropyl alcohol is commonly used to clean the sample plate and Nernst glower in IR spectroscopy to remove any contaminants that may interfere with the analysis. The crude sample was analyzed to determine the functional group or structural characteristics. Every structural feature or functional group of a substance has a unique vibrational frequency, which was identified by the absorption peaks observed in the IR spectrum. By comparing the IR spectrum of an unknown sample to reference spectra of known compounds, the functional groups present in the sample can be recognized, allowing for the identification of the molecule.[9]

Determination of GC and HPLC

GC analysis was performed on a crude extract dissolved in methanol. The injector temperature was set at 220°C, and a 1uL sample of the extract was injected into the instrument. The oven temperature was programmed to start at 40°C for 0.2 minutes and then ramp up to 300°C. The total time of the program was 44 minutes. The column had a length and inner diameter of 25.0 m and 0.32 mm, respectively, with a maximum temperature of 300°C. The film thickness of the column was 5 µm. The appearance time, height, width, and area of the peaks in the chromatogram can be measured to acquire quantitative data about the components in the crude extract.[10]

Dissolve the crude extract in methanol. Inject 20 µL of the solution into an HPLC vial and place it in a temperature-controlled environment (322°C). Use a Shim-pack GIST column (250*4.6mm5µm) and a mobile phase comprising 10 mM ammonium acetate in water (A) and acetonitrile (B) to separate the components of the crude extract. Analyze the resulting peaks using HPLC and isolate the highest peaks. Weigh the lyophilized material and test it for the presence of the isolated peaks to identify the compounds. *C. bonducella* crude extract was analyzed and purified.[11,12]

RESULTS AND DISCUSSION

Extracted Hydroalcoholic Plant

C. bonducella leaf was obtained from Melur village, Madurai district, Tamil Nadu, India. It was authenticated in St. Joseph's College (Autonomous), Tiruchirappalli. The coarse powder of *C. bonducella* leaves was extracted with Hydro alcohol. The extraction was conducted for 12 hours, and the solvent was distilled out to attain a concentrated extract (Fig.-1). The concentrated extract was vacuum-dried, and the dry extract was stored in an airtight container. Then the extract with the solvent was collected.[13] The remaining solvent was evaporated by a solvent evaporation method. The extract was collected, and the percentage yield was calculated for the ethanolic extract of *C. bonducella*.

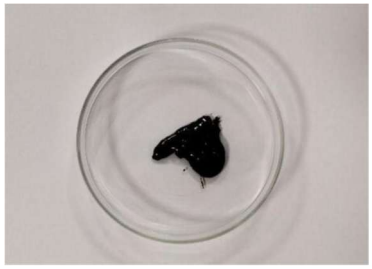

Fig.-1: Extraction of *C. bonducella*

UV and IR Analysis

The crude extract sample was dissolved in 70% Ethanol, and it was sonicated to analyze the blank to run peak analysis. Then, the 0.02% solution was filtered and used, and the results are exemplified in Fig.-2. The peak is indicated by the wavelength and low absorbance of the chemical compound's identification. In this case, peaks were identified at 665.0 nm, 265.0 nm, and 354.0 nm. IR spectroscopy analysis of a crude extract sample, using the frequencies observed in the spectrum to propose the presence of various functional groups includes, 1068.59 wavenumber: C-O stretching, 1368.77 wavenumber: N-O stretching, C-H bonding, O-H bonding, 1605.76 wavenumber: C=C stretching, 1733.59 wavenumber: C=O stretching, 2918.51 wavenumber: C-H stretching, and 3323.54 wavenumber: O-H stretching, N-H stretching (aliphatic primary amine or secondary amine). Different functional groups absorb IR radiation at diverse characteristic frequencies, which can be used to identify the existence of these groups in a sample (Fig.-2).

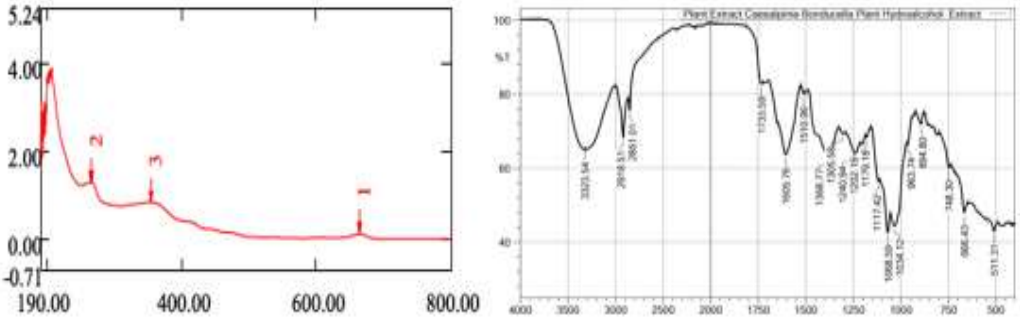

Fig.-2: UV and IR Spectral Analysis

Gas and HPLC

GC analysis of the hydroalcohol extract of leaves of *C. bonducella* revealed various compounds.[12] A total of seventeen most abundant compounds were identified. Initially, the GC analysis was performed for a black sample, which is shown in Fig.-3. GC chromatogram of the seventeen peaks of the compounds detected. The two samples show the seventeen peaks of the gas chromatography, and the concentrated level is very low, and it has a low area % level. Further tests were carried out to analyze the compounds. Because the temperature will be changed to 40 minutes. For 40 minutes, the temperature was maintained at 400°C. The sample was purified to analyze the compounds and their structure. Based on the HPLC preparative analysis and lyophilization, two peaks were identified as Peak 1 and Peak 2 with retention times of 16.87 min and 19.45 min, respectively (Fig.-4). Peak 1 has an area of 2,220,680 and 95.08%, while Peak 2 has an area of 3,242,569 and an area of 96.96%.

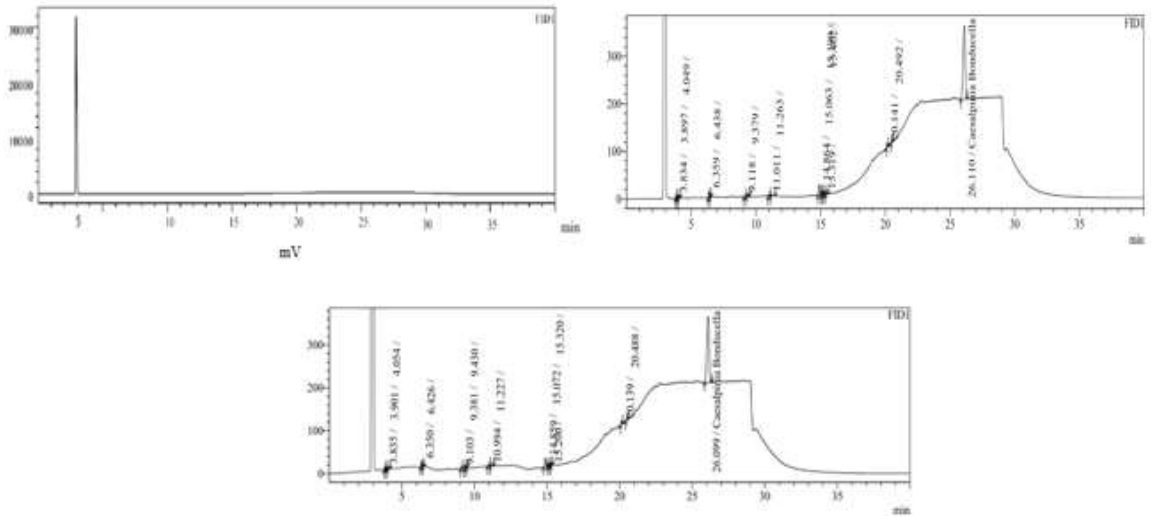

Fig.-3: Blank Sample, *C. bonducella* GC Sample 1 and 2 Peaks

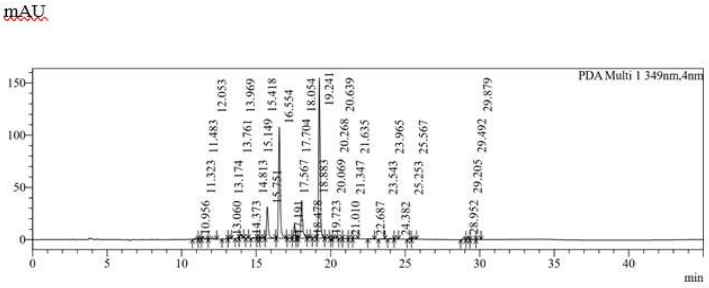

Fig.-4: HPLC Preparative Crude Sample

CONCLUSION

C. bonducella is a useful medicinal plant based on the phytochemical analysis of its leaves. Keeping this in mind, we have employed the hydroalcoholic extract of *C. bonducella* leaves, which are then characterized. In this case, the qualitative analysis of UV and IR shows some functional groups present in *C. bonducella* leaf extract. The crude extract was dissolved in methanol, and it was examined by LC-MS/MS. The LC-MS/MS shows that two high-area peaks and their masses are identified. The peak 1 and the peak 2 masses are 740 and 564. In preparative, the two peaks are isolated. The two isolated peaks are distilled and lyophilized. The lyophilized plant materials are analyzed by NMR spectroscopy. The NMR results and their masses based on the structures and the compounds were identified. The two compounds identified were previously unknown in this plant, and their potential pharmacological uses and microbiological activities need to be explored further. It is concluded that the aqueous extracts of *C. bonducella* leaf have a more inhibitory impact on CaOx crystallization and thus may be helpful in the therapy of urolithiasis. The compound identification is classified through further tests. However, there is a prerequisite for a thorough examination of expounded preclinical experiments and clinical tests to begin the usage of plants as anti-urolithiasis agents.

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CONFLICT OF INTERESTS

The authors declare that they have no conflict of interest.

AUTHOR CONTRIBUTIONS

All the authors contributed significantly to this manuscript, participated in reviewing/editing and approved the final draft for publication. The research profile of the authors can be verified from their ORCID ids, given below:

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REFERENCES

1. P. Ezhilarasan, S. Sivakrishnan and S.V. Anbiah, *Research Journal of Pharmacy and Technology,* **14(11)**, 5891(2021), https://doi.org/10.52711/0974-360X.2021.01024
2. A. Kumar, M.K. Nandi, B. Kumar, A. Kumar, R. Kumar, A.K. Singh and M. Singh, *International Journal of Pharmaceutical Investigation*, **11(3)**, 306(2021), https://doi.org/10.5530/ijpi.2021.3.54
3. S. Sasidharan, K.P. Srinivasa Kumar, S. Kanti Das and J. Hareendran Nair, *Medicinal and Aromatic Plants*, **10(7)**, 394(2021), https://doi.org/10.35248/2167-0412.21.10.394
4. N. Chaachouay and L. Zidane, *Drugs and Drug Candidates,* **3(1)**, 184(2024), https://doi.org/10.3390/ddc3010011
5. S.E. El-Abeid, M.A. Mosa, M.A. El-Tabakh, A.M. Saleh, M.A. El-Khateeb and M.S. Haridy, *Journal of Nanobiotechnology*, **22(1)**, 28(2024), https://doi.org/10.1186/s12951-024-02307-9
6. L.S. Putra, J. Sukweenadhi, C. Nathania, E.S. Wibowo, G. Buschle-Diller and M.G. Purwanto, *Heliyon*. **8(2)**, (2022), https://doi.org/10.1016/j.heliyon.2022.e08951
7. S. Sasidharan, S. Kp, A. Bhaumik, S. Kanti Das and J.H. Nair, *Research and Reports in Urology*, **14(2022)**, 225(2022), https://doi.org/10.2147/RRU.S365598
8. M.J. Gadakh, R.S. Jadhav and D.N. Vikhe, *Journal of Drug Delivery and Therapeutics*, **10(3-s)**, 308(2020), https://doi.org/10.22270/jddt.v10i3-s.4059
9. V. Kandasamy and U. Balasundaram, *Journal of Ethnopharmacology*, **279**, 114375(2021), https://doi.org/10.1016/j.jep.2021.114375
10. N.U. Olivia, U.C. Goodness and O.M. Obinna, *Future Journal of Pharmaceutical Sciences*, **7(59)**, 1(2021), https://doi.org/10.1186/s43094-021-00208-4
11. O. Amamou, J.P. Denis, E. Heinen, T. Boubaker and S. Cardinal, *Molecules*, **28(21)**, 7294(2023), https://doi.org/10.3390/molecules28217294
12. N. Pournaghi, F. Khalighi-Sigaroodi, E. Safari and R. Hajiaghaee, *Iranian Journal of Pharmaceutical Research*, **20(1)**, 274(2021), https://doi.org/10.22037/ijpr.2020.112557.13824
13. D.K. Meena, A.K. Sahoo, P.P. Srivastava, N.P. Sahu, M. Jadhav, M. Gandhi, H.S. Swain, S. Borah, and B.K. Das, *Scientific Reports*, **11(1)**, 10656(2021), https://doi.org/10.1038/s41598-021-88710-w

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