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Qualitative and Quantitative Phytochemical Screening of *Acanthospermum hispidum* D.C. Leaf Extracts

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ABSTRACT
Phytochemicals have great potency as therapeutic agents. There is continuous and urgent need to discover new therapeutic compounds with diverse chemical structures and novel mechanism of action because there has been an alarming increase in the incidence of new and re-emerging infectious diseases. Hence, the present investigation was carried out to assess the phytoconstituents of various leaf extracts of *Acanthospermum hispidum*. The leaves were collected from its wild habitats, washed, air dried and then powdered. The solvent extracts of the respective leaves were prepared using Soxhlet apparatus with acetone, benzene, chloroform, distilled water, hexane and methanol. The extracts were subjected to qualitative and quantitative phytochemical analyses as per standard procedures. The results showed that alkaloids, steroids and terpenoids were detected in maximum number of solvent extracts (five among six solvent extracts analysed) followed by triterpenoids (in four solvent extracts). Saponins and quinones were absent in all the six solvents used for qualitative phytochemical analyses. Maximum number (six) of compounds were identified in methanol and chloroform, and least (four) in hexane acetone, benzene and distilled water leaf extracts of *Acanthospermum*. Findings of quantitative analyses highlighted that maximum content of alkaloids (3.623 %) were determined in aqueous extract, flavonoids (0.867 mg QE/g) in distilled water, phenols (4.367 mg GAE/g) in methanol, tannins (1.290 mg TAE/g) in methanol and terpenoids (3.531%) in chloroform extracts. The result of this study is encouraging further clinical studies to determine the potential effectiveness of particular phytochemical *in vivo*.

Keywords: Phytochemical analyses, *Acanthospermum hispidum*, Leaf extracts.

1. INTRODUCTION

Plant-derived substances have recently become of great interest owing to their versatile applications. Medicinal plants are a group of species that accumulate different active principles, useful in treating various human or animal diseases. They are the richest bio-resource of drugs of traditional systems of medicine, modern medicines, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs [1].

Phytochemicals are naturally occurring in different parts of the medicinal plants that have defense mechanism and protect from various diseases [2]. The medicinal plants are useful for healing as well as for curing of human diseases because of the presence of phytochemical constituents which produce definite physiological action on the human body and these bioactive substances include alkaloids, carbohydrates, terpenoids, steroids, flavonoids, tannins, etc. [3].
Plants with prospective medicinal activity have recently come to the attention of scientists and researchers because of their bioactive potential. Preliminary screening of phytochemicals is a valuable step in the detection of the bioactive principles present in medicinal plants and subsequently may lead to drug discovery and development. Due to the significance in this above perspective, such preliminary phytochemical screening of plants is the need of the hour in order to discover and develop novel therapeutic agents with improved value. Thus, the present study was aimed to assess the various phytoconstituents present in the six different leaf extracts of *Acanthospermum hispidum*.

2. MATERIALS AND METHODS

2.1. Plant sample collection

The healthy leaves of *Acanthospermum hispidum* belongs to the family Asteraceae and locally called as Kaandhaarimull in Tamil were collected from their natural habitats from coastal region near Athoor (78.0824º E longitude and 8.6106º N latitude) in Thoothukudi district of Tamil Nadu, India, and brought to the laboratory. The leaves were washed thoroughly with tap water and shade dried at room temperature to attain constant weight. The air dried samples were powdered in an electric blender and stored in plastic bags for further analysis. The plant was botanically confirmed and authenticated as per APG IV classification [4].

2.2. Preparation of plant extract

The dried powder material was extracted sequentially in six different solvents viz., acetone, benzene, chloroform, distilled water, hexane and methanol. 15 g of the dried and powdered plant material were separately extracted with 150 ml of acetone, benzene, chloroform, distilled water, hexane and methanol using Soxhlet apparatus for 6-8 hours at a temperature not exceeding the boiling point of the solvents. The obtained crude extracts were filtered by using Whatman No. 1 filter paper and then concentrated under vacuum at 40° C by using a rotary evaporator and later stored at 4° C for further use.

2.3. Qualitative phytochemical analysis

Preliminary phytochemical analyses were carried out on the leaf extracts of *Acanthospermum hispidum* in order to determine the presence of different phytochemicals like alkaloids, flavonoids, glycosides, phenols, quinones, reducing sugars, saponins, steroids, tannins, terpenoids and triterpenoids by subjecting standard procedures [5-8]. The qualitative results were expressed as (+) for the presence and (-) for the absence of phytochemical.

2.4. Quantitative phytochemical analysis

The content of alkaloids, flavonoids, phenols, tannins and terpenoids were determined as per the methodology of Harborne [7] and the results were expressed as % for alkaloids and flavonoids, mg Gallic Acid Equivalent (GAC)/g for phenols, mg Tannic Acid Equivalent (TAC)/g for tannins and percentage (%) for terpenoids.

3. RESULTS AND DISCUSSION

The results regarding the qualitative phytochemical screening of *Acanthospermum hispidum* leaf, alkaloids, glycosides, phenols, steroids, terpenoids and triterpenoids were found to be present in methanol extract, flavonoids, glycosides, reducing sugars, steroids, terpenoids and terpenoids were present in chloroform extract, alkaloids, flavonoids, glycosides, steroids and terpenoids were present in hexane extract, alkaloids, phenols, steroids and tannins were present in acetone extract, alkaloids, steroids, terpenoids and triterpenoids were present in benzene extract and alkaloids, phenols, terpenoids and triterpenoids in distilled water extract. All the six extracts showed the absence of quinones and saponins (Table 1). Most number of phytochemicals (six) were found to be present in chloroform and methanol extract, followed by hexane extract (five) and least (four each) with acetone, benzene and distilled water extracts of *Acanthospermum hispidum* leaf (Fig. 1).

Table 1: Qualitative phytochemical screening of *Acanthospermum hispidum* leaf extracts

Phytoconstituents	Solvent extracts					
	Acetone	Benzene	Chloroform	Aqueous	Hexane	Methanol
Alkaloids	+	+	–	+	+	+
Flavonoids	–	–	+	–	+	–
Glycosides	–	–	+	–	+	+
Phenols	+	–	–	+	–	+
Quinones	–	–	–	–	–	–
Reducing sugars	–	–	+	–	–	–
Saponins	–	–	–	–	–	–
Steroids	+	+	+	–	+	+
Tannins	+	–	–	–	–	–
Terpenoids	–	+	+	+	+	+
Triterpenoids	–	+	+	+	–	+

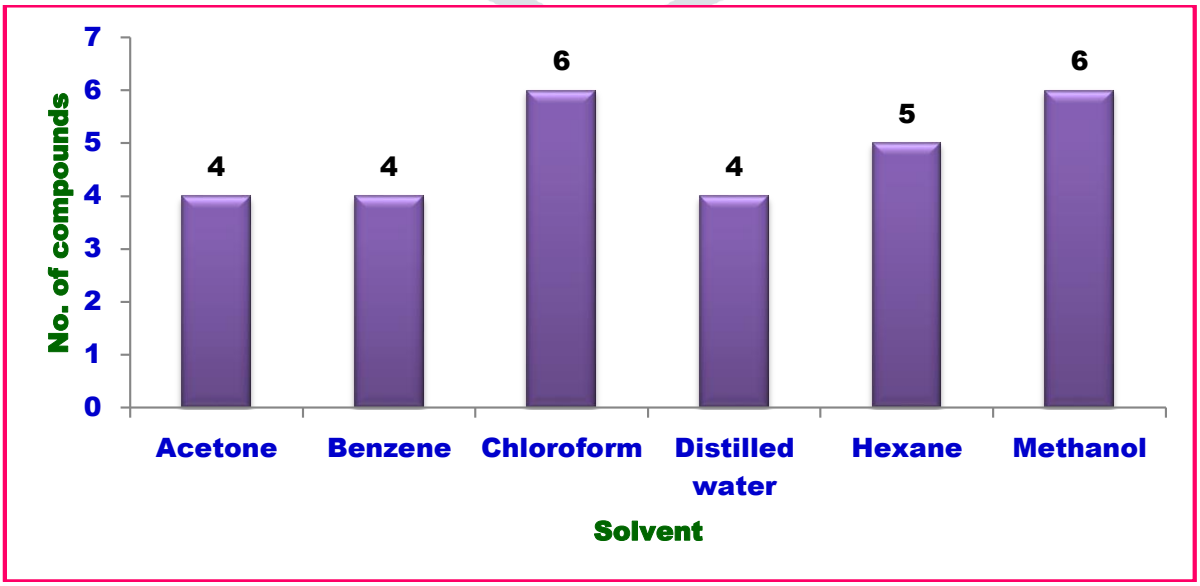

Fig. 1: Number of phytoconstituents detected in six different solvents

From the findings of qualitative phytochemical analyses, it was clearly known that most of phytoconstituents were detected in methanol and chloroform extracts than that of other solvent extracts subjected for present study. This can be attributable to the higher solubility of the phytocompounds of plant material in methanol than other solvents. Also, the recovery of phytochemical from plant sample could be influenced by dielectric constant, chemical structure of solvents used, and as well as chemical properties of phytochemicals [9].

For quantitative analyses, four different solvents (acetone, chloroform, distilled water and methanol) were selected and subjected as they were shown maximum number of phytoconstituents present in *Acanthospermum hispidum* leaf. The results of quantitative phytochemical analyses revealed that maximum (3.623 %) alkaloid content was observed in aqueous leaf extract of *Acanthospermum hispidum*, followed by methanol (3.553 %) and the least alkaloid content (2.733 mg/g) was detected in acetone extract. In case of flavonoids, highest (0.867 mg QE/g) content was exhibited in distilled water leaf extract and lowest (0.112 mg QE/g) in acetone extract. The methanol extract was found to be had the highest phenol content (4.367 mg GAE/g) and acetone extract with lowest phenol content (2.534 mg GAE/g). The outcome of present study also indicated that about 1.290 mg TAE/g was determined as maximum tannin content in methanolic extract. Next to this, 3.658 mg TAE/g was detected in chloroform extract and 0.569 mg TAE/g in acetone extract as minimum. Among terpenoid content, in chloroform extract, it was found to be exhibited at the level of 3.531 %, which was more than that recorded in other solvent extracts (Table 2).

Table 2: Quantitative phytochemical screening of *Acanthospermum hispidum* leaf extracts

Phytoconstituents*	Solvent extracts*			
	Acetone	Chloroform	Distilled water	Methanol
Alkaloids (%)	2.733	3.450	3.623	3.553
Flavonoids (mg QE/g)	0.112	0.397	0.867	0.811
Phenols (mg GAE/g)	2.534	3.658	3.003	4.367
Tannins (mg TAE/g)	0.569	0.116	1.103	1.290
Terpenoids (%)	2.837	3.531	3.090	2.358

*Values are mean of 3 replicates

The quantitative phytochemical screening of *Acanthospermum hispidum* leaf extracts showed that they own their phytoconstituents and such phytochemicals have several important biological activities. It was reported that alkaloids have the pharmacological activities like antimicrobial [10], antiarrhythmic, analgesic [11] and antihyperglycemic [12] activities. It was known that flavonoids possess alpha-glucosidase activity [13], antioxidant activity [14] and anti-inflammatory activity [15]. Phenolic compounds are also known for their anti-inflammatory [16,17], antimicrobial [18-20], and antioxidant [21,22] effects. Tannins have been reported to have various physiological effects like anti-irritant, antiparasitic effects [23]. Terpenoids have been found to be useful in the prevention and therapy of several diseases including cancer and possess antimicrobial, anti-parasitic, antiviral, anti-allergenic, antispasmodic, anti-hyperglycemic, anti-inflammatory, and immunomodulatory properties [24,25]. All these research evidences strongly justify the medicinal usage of leaves of *Acanthospermum hispidum* as they contained the above mentioned biologically important phytochemicals detected by the present study.

4. CONCLUSION

Further chromatographic studies should be carried out on the phytochemical compounds present in leaves of *Acanthospermum hispidum* to isolate, identify, characterize and elucidate the structure of the bioactive compounds. Biological efficacies of the isolated compounds should also be tested using animal models.

5. REFERENCES

1. Ncube NS, Afolayan AJ & Okoh AI. Assessment techniques of antimicrobial properties of natural compounds of plant origin: current methods and future trends. African Journal of Biotechnology 2008; 7(12):1797-1806.
2. Krishnaiah D, Sarbatly R & Bono A. Phytochemical antioxidants for health and medicine: A move towards nature. Biotechnology and Molecular Biology Reviews, 2007; 1:97-104.
3. Nostro A, Germanò MP, D'angelo V, Marino A & Cannatelli MA. Extraction methods and bioautography for evaluation of medicinal plant antimicrobial activity. Letters of Applied Microbiology, 2000; 30: 379-384.
4. APG IV. An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants. Botanical Journal of the Linnean Society, 2016; 181(1): 1-20.
5. Trease GE & Evans WC. Pharmacognosy. In: Soundera WB. (Ed.), 4th ed., New York Scientific Press, USA, 1996: 243-283.
6. Brain KR, Turner TD. The Practical Evaluation of Phytopharmaceuticals. 1st Edition, Wright Science Technical, Bristol Britain. 1975.
7. Harborne JB. Phytochemical methods: A Guide to Modern Technique of Plant Analysis. Champman and Hall Publishers. London. 1998.
8. Mukherjee PK. Quality Control of Herbal Drugs. Business Horizons Pharmaceutical Publishers, New Delhi. 2010.
9. Felhi S, Daoud A, Hajlaoui H, Mnafgui K, Gharsallah N, Kadri A. Solvent extraction effects on phytochemical constituents profiles, antioxidant and antimicrobial activities and functional group analysis of *Ecballium elaterium* seeds and peels fruits. Food Science and Technology (Campinas), 2017; 37(3):483-492.
10. Cushnie TP, Cushnie B, Lamb AJ. Alkaloids: An overview of their antibacterial, antibiotic-enhancing and antivirulence activities. International Journal Antimicrobial Agents, 2014; 44(5):377-386.
11. Raymond SS, Jahr JS, Pitchford JMW. The essence of Analgesia and Analgesics. Cambridge University Press, Cambridge. 2010; 82-90.
12. Qiu S, Sun H, Zhang AH, Xu HY, Yan GL, Han Y, Wang XJ. Natural alkaloids: basic aspects, biological roles, and future perspectives. Chinese Journal of Natural Medicines, 2014; 12(6):401-406.

13. Geng P, Yang Y, Gao Z, Yu Y, Shi Q, Bai G. Combined effect of total alkaloids from *Feculae bombycis* and natural flavonoids on diabetes. Journal of Pharmacy and Pharmacology, 2007; 59(8):1145-1150.
14. Gil MI, Ferreres F, Tomas-Barberan FA. Effect of post-harvest storage and processing on the antioxidant constituents (flavonoids and vitamin C) of fresh-cut spinach. Journal of Agricultural and Food Chemistry, 1999; 47:2213-2217.
15. Panthong A, Kanjanapothi D, Tuntiwachwuttikul P, Pancharoen O, Reutrakul V. Anti-inflammatory activity of flavonoids. Phytomedicine, 1994; 1:141-144.
16. Naczk M, Shahidi F. Phenolics in cereals, fruits and vegetables: Occurrence, extraction and analysis. Journal of Pharmaceutical and Biomedical Analysis, 2006; 41:1523-1542.
17. Giftson JS, Jayanthi S, Nalini N. Chemopreventive efficacy of gallic acid, an antioxidant and anticarcinogenic polyphenol, against 1, 2-dimethyl hydrazine induced rat colon carcinogenesis. Investigational New Drugs, 2010. 28:251-259.
18. Deng Y, Zhao Y, Padilla-Zakour O, Yang G. Polyphenols, antioxidant and antimicrobial activities of leaf and bark extracts of *Solidago canadensis* L. Industrial Crops and Products, 2015; 74:803-809.
19. Popa VI. Wood bark as valuable raw material for compounds with biological activity. Celuloza Si Hârtie, 2015; 64:5-17.
20. Tanase C, Cosarca S, Toma F, Mare A, Man A, Miklos A, Imre S, Boz I. Antibacterial activities of beech bark (*Fagus sylvatica* L.) polyphenolic extract. Environmental Engineering and Management Journal, 2018; 17:877-884.
21. Subramanian R, Raj V, Manigandan K, Elangovan N. Antioxidant activity of hopeaphenol isolated from *Shorea roxburghii* stem bark extract. Journal of Taibah University for Science, 2015; 9:237-244.
22. Zaiter A, Becker L, Petit J, Zimmer D, Karam MC, Baudelaire É, Scher J, Dicko A. Antioxidant and antiacetylcholinesterase activities of different granulometric classes of *Salix alba* L. bark powders. Powder Technology, 2016; 301:649-656.
23. Naveen Prasad R, Viswanathan S, Renuka Devi J, Vijayashree N, Swetha VC, Archana R, Parathasarathy N, Johanna R. Preliminary phytochemical screening and antimicrobial activity of *Samanea saman*. Journal of Medicinal Plants Research, 2008; 2(10):268-270.
24. Wagner KH, Elmadfa I. Biological relevance of terpenoids. Overview focusing on mono-, di- and tetraterpenes. Annals of Nutrition and Metabolism, 2003; 47:95-106.
25. Rabi T, Bishayee A. Terpenoids and breast cancer prevention. Breast Cancer Research and Treatment, 2009; 115:223-239.