Phytochemical and GCMS Analysis of Methanolic Leaf Extract of Bauhinia Variegate

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Herbal remedies are the most widely utilized medical therapy in most underdeveloped nations. Medicinal plants have been utilized for millennia due to their broad availability. Bauhinia variegata is a medicinal plant that grows in India and is thought to have medicinal properties due to specific bioactive components. The present work evaluated the phytochemical composition of the methanol extract of Bauhinia variegata's leaves. Further, three different assays were used to test the antioxidant activity of leaf extracts of Bauhinia variegata at varying doses. The antioxidant activity of several extracts of plant was shown to be more promising as a consequence of our research. The components of the more potent phytochemicals of Bauhinia variegata were further investigated using gas chromatography and mass spectrometry. Our results reveal that the presence of specific phytochemicals warrants the Bauhinia variegata's use in health-promoting properties.

Keywords: Antioxidant Activity; Bauhinia Variegata; Leaf Extract; Methanol Extract.

Medicinal plants are used as a source of raw materials for various drugs. Nearly 80% of the world's population uses a traditional system of medicines for primary health care, which has a significant economic advantage for treating various human disorders ¹. Phytochemicals in medicinal plants such as leaves, fruits and seeds act as a natural disease preventative and resistance mechanism. Chlorophyll, proteins, and simple sugars are primary components, whereas secondary chemicals include terpenoids, alkaloids, flavonoids, saponins, and phenolic compounds². The Western Ghats and Himalayas are rich in plant species in India, one of the world's wealthiest countries in terms of plant biodiversity ³. Phytochemicals and medicinal plants have long been India's most

great health care and life enhancement source. The country has the most traditional herbal plants and their prescriptions ⁴.

Bauhinia variegata is a flowering plant that belongs to the Fabaceae family of legumes. It is endemic to a region that stretches from China through Southeast Asia and then to the Indian subcontinent ⁵. In tropical places, the tree is frequently cultivated as an ornamental, hedge plant, or street tree because of its attractive flowers. It is regarded for its fragrant, beautiful pink, purple, or white blooms ⁶. The leaves are obcordate in form, 3.9–7.9 inches long and wide, rounded, and bilobed at the base and tip. *Bauhinia variegata* is valued for its therapeutic benefits, and the juice extracted from the plant's bark is used to treat

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amoebic dysentery, diarrhoea, and other stomach ailments ⁷. The dried buds are used to cure piles, dysentery, diarrhoea, and worms, while the root acts as a snake poison antidote. Further, the bark of Bauhinia variegata has long been used as a tonic and to cure ulcers. This herb is widely utilised in traditional medicine to treat a variety of disorders, including inflammatory problems. Residents in hilly parts in Punjab's northern east have utilised young flowers as a vegetable to treat a number of disorders, including liver illnesses ⁷.

Wild plant extracts play a significant part in tribal people's complex cultural system for healing a range of diseases, and the Bauhinia variegata has been employed in traditional knowledge⁸. According to study, many edible wild plants are abundant in phytochemicals, which may have health-promoting characteristics. Several researchers have been paying attention to this plant and its therapeutic usefulness in recent days, owing to its broad effect ⁷. Since, natural antioxidants are very effective in preventing the damaging processes caused by oxidative stress, which is responsible for a variety of human disorders, the purpose of this study, as well as its major aim, is to report phytochemical composition and pharmacological validity of leaf extracts in terms of antioxidant activity.

MATERIALS AND METHODS

Extraction

The plant "Bauhinia variegata" was collected in Trivandrum, Kerela, India (08°292 153 N, 76°572 93 E). Plant taxonomist Professor PV Anto, Department of Botany, Thomas College, Thrissur, India, identified the plant samples on the field based on morphology and taxonomy. Leaves were cleaned and dried in the shade for ten days at room temperature (37 °C). The dried plant components were powdered, and the sample was extracted using petroleum ether, chloroform, ethyl acetate, and methanol in order. The extracts were filtered through Whatman filter paper with a pore size of 8im, and only pure refined extracts were used in this investigation. The chemicals and reagents of AR grade purchased from Spectrum, Merck, Nice and HiMedia were used for the present study.

Phytochemical screening

Standard phytochemical analysis was used to check for the presence and absence of secondary metabolites in plant extracts ⁹. The presence of phytoconstituents such as alkaloids, cardiac glycosides, anthraquinones, steroids, triterpenes, and reducing sugars was determined using a methanolic extract of the *Bauhinia variegata* plant's leaf. Other phytochemical tests were performed on several classes of phytoconstituents, including the shinoda test for flavonoids, the froath test for saponins, the alkaloids test for alkaloids, and the legal's test for glycosides.

DPPH assay

The capacity of the plant extracts to scavenge free radicals was determined using the 1,1 diphenyl 1–2 -picryl-hydrazyl (DPPH) assay ¹⁰. Plant doses (20 to 100 mg/ml) were incubated in the dark with DPPH solution (2 ml) (30 min). The test sample was measured at 517 nm and compared to a blank combination (DPPH). Gallic and ascorbic acid were employed as a standard or positive control for comparison.

Hydrogen Peroxide assay

The method of using hydrogen peroxide as an oxidant scavenger was employed (Ruch et al., 1989). In phosphate buffer (50 Millimolar) with a pH of 7.4, a 4 mM hydrogen peroxide solution was prepared. Place the mixture in plant concentrations to reach a volume of 4 ml in the shaking incubator for 10 minutes. At 230 nm, the absorbance of the blank (phosphate buffer), test samples, and standards were measured ¹¹.

FRAP (Ferric Reducing Antioxidant Power Assay) assay

The Benzie and Strain technique was used to determine the reducing activity of phenolic acids ¹². The following was done to prepare the FRAP reagent: 12.5 mL of pH 3.6 0.3 mol/L acetate buffer was combined with the same amount of methanol. For each phenolic acid, measurements were taken five times. The findings of the tests were given as a reaction rate in total antioxidant units per mol of material.

Analysis by gas chromatography and mass spectrometry (GCMS)

The GC-MS apparatus (Shimadzu GC-MS; Model Number: QP2010S) was used to identify the Tetramethyl heptadecan derivatives. The GC system employed an HP-5 MS capillary column. At a flow rate of 1 mL/min, helium was used as the carrier gas. The injection was 2 litres in volume. The mass spectroscopy detector used ionisation energy EI of 70 eV. GCMS Solutions Libraries operated the GC-MS apparatus (NIST 11 & Mass spectral library). The injector and detector were both set to 250°C. Compound identification was accomplished using standard procedures.

RESULTS

Phytochemical analysis

Phytochemical study of methanolic extract of *Bauhinia variegata's* leaves was

Table 1. List of the bioactive compounds
from the methanolic leaf extract of Bauhinia
variegata.

Secondary Metabolites	Results
Alkaloids	++
Flavonoids	++
Cardiac Glycosides	-
Terpenoids	++
Phenolics	++
Proteins	++
Carbohydrates	++
Steroids	-
Coumarins	+

performed to determine the presence and absence of carbohydrates, alkaloids, flavonoids, phenols, proteins, steroids, cardiac glycosides and terpenoids (**Table 1**). The presence of alkaloids, flavonoids, terpenoids, phenolics, carbohydrates and proteins at a higher level and the absence of steroids and cardiac glycosides in the leaves have been observed from the table. Coumarins are found in moderate amounts in the leaves of the plants. The fact that isolated chemicals could be employed for important biological activities is supported by these findings. The literature examines the significance of each chemical in biological action ¹³.

Antioxidant activity

Three different antioxidant techniques were applied to quantify the free radical scavenging activity of methanol extracts of *Bauhinia variegata* in this investigation (**Table 2**). Leaf extracts of *Bauhinia variegata* were shown to have significant antioxidant activity in DPPH, hydrogen peroxide, and FRAP antioxidant experiments. The presence of active components such as flavonoids and phenols is considered to confer antioxidant potential to medicinal plants. Excessive reactive oxygen species generation or a lack of antioxidant resistance causes oxidative stress in the human body, which can lead to degenerative disorders.

Table 2. Antioxidant activity measured from three different assays using methanolic leaf extract of *Bauhinia variegata*

Concebtration(µg/ml)		Percentage of inhibition	
DPPH assay		Ascorbic acid Standard	
10	15	24.5	
20	28	37.5	
30	36	46.3	
40	48	58.2	
50	59	60.3	
H2O2 assay	Quercetin Standard		
10	45	59	
20	53	64	
30	61	71	
40	68	79	
50	76	85	
FRAP assay	Ascorbic acid Standard		
10	15.1	30.2	
25	26.4	45.3	
50	46.3	59.5	
75	65.4	74.1	
100	78.2	84.6	

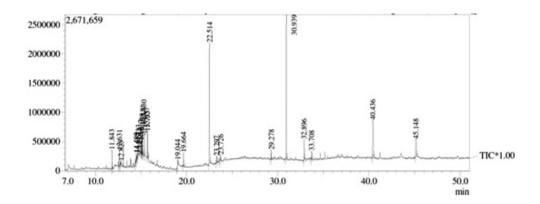


Fig. 1. Gas Chromatography & Mass Spectroscopy analysis of Bauhinia variegata methanolic extract

Antioxidant activity regulates a number of processes, including scavenging free radicals, decomposing peroxides, increasing reducing capacities, preventing transition metal ion binding, and inhibiting chain initiation. The current study's findings are consistent with prior findings for the leaf analyses of *Bauhinia variegata*.

Gas chromatography and mass spectrometry (GCMS)

The results of GC-MS analysis of methanolic extract of Bauhinia variegata's leaf is shown in figure 1 and allowed to the elucidation of compounds such as Caryophyllene, beta-copaene, alpha-muurolene, 5-Ethyl-1,3-dioxane-5-methanol, tert-butyldimethylsilyl ether, cadinol, tau-muurolol, beta-caryophyllene epoxide neoclovenoxidalkohol, cedrol, mome inositol, geranyl hexanoate, ascorbic acid 2,6-dihexadeanoate, hexadecanoic acid, 5,8-octadecadienoic acid, phytol, lycoxanthin, tetracosanoic acid, vitamin E and beta-sitosterol respectively. A notable observation from the analysis is the presence of vitamin E. It is reported that vitamin E supplements may help to avoid coronary heart disease, boost immunity, reduce inflammation, improve eye health, and cut cancer risk. Mome inositol, geranyl hexanoate, phytol and bis (2-ethylhexyl) phthalate are main constituents which present about an average of 15 % of the total area. Phytol aids in cholesterol metabolism and has anti-inflammatory properties, reduces the risk of heart disease and chronic inflammatory disorders.

DISCUSSION

Pharmaceuticals are more costly and cause side effects than herbal remedies. Traditional medicinal plant applications have an ancient legacy of value, and were largely employed to recover from sickness by our predecessors. The recent trend of avoiding natural rather than artificial sources of disease prevention, on the other hand, is disheartening. The antimicrobial resistance and synthetic drug adverse effects have been reported all over the world. As a result, creating a safer synthetic medicine substitute has become a major priority. In this case, we concentrated on a medicinal plant that has been used as a vegetable in south India for centuries ¹⁴. Our findings show that the leaves of Bauhinia variegata have a better medicinal value as a source of carbohydrates and proteins, in this study. Despite the fact that there are several reports demonstrating the medicinal effects of Bauhinia variegata (leaves), we give a foundation for further research into the antioxidant activity of Bauhinia variegate.

CONCLUSION

The presence of certain phytochemicals justifies the *Bauhinia variegata*'s usage in healthpromoting characteristics, according to this research. The therapeutic activity of the plant could be due to the presence of particular phytochemicals. Furthermore, the research demonstrates that the plant in question is a strong source of certain phytochemicals, making it a medicinal plant.

REFERENCES

- Atanasov AG, Zotchev SB, Dirsch VM, et al. Natural products in drug discovery: advances and opportunities. *Nat Rev Drug Discov* 2021; 20: 200–216.
- Olatunji TL, Adetunji AE, Olisah C, et al. Research Progression of the Genus Merremia: A Comprehensive Review on the Nutritional Value, Ethnomedicinal Uses, Phytochemistry, Pharmacology, and Toxicity. *Plants* 2021; 10: 2070.
- Subashree K, Dar JA, Karuppusamy S, et al. Plant diversity, structure and regeneration potential in tropical forests of Western Ghats, India. *Acta Ecol Sin* 2021; 41: 259–284.
- 4. Fierascu RC, Fierascu I, Baroi AM, et al. Selected Aspects Related to Medicinal and Aromatic Plants as Alternative Sources of Bioactive Compounds. *Int J Mol Sci* 2021; **22**: 1521.
- Kansal M, Shukla P, Shukla P. a Boon To Human Health-Bauhinia Variegata. *Int J Pharmacogn* 2020; 1: 155–161.
- Kulkarni YA, Laddha AP. Bauhinia variegata leaf extract: An effective management option for diabetic cardiomyopathy. *South African J Bot* 2020; 132: 50–58.
- 7. Sharma K, Kumar V, Kumar S, et al. Bauhinia

variegata: a comprehensive review on bioactive compounds, health benefits and utilization. *Adv Tradit Med* 2021; **21**: 645–653.

- Gul H, Awais M, Saddick S, et al. Quantification of biochemical compounds in Bauhinia Variegata Linn flower extract and its hepatoprotective effect. *Saudi J Biol Sci* 2021; 28: 247–254.
- 9. Evans WC. Trease and Evans Pharmacognosy. 15th edtion. *Edinburgh, Saunders*.
- 10. Sanja SD, Sheth NR, Patel NK, et al. Characterization and evaluation of antioxidant activity of Portulaca oleracea. *Int J Pharm Pharm Sci* 2009; **1**: 74–84.
- Ruch RJ, Cheng S, Klaunig JE. Prevention of cytotoxicity and inhibition of intercellular communication by antioxidant catechins isolated from Chinese green tea. *Carcinogenesis* 1989; 10: 1003–1008.
- Benzie IFF, Strain JJ. The Ferric Reducing Ability of Plasma (FRAP) as a Measure of "Antioxidant Power": The FRAP Assay. *Anal Biochem* 1996; 239: 70–76.
- 13. Altemimi A, Lakhssassi N, Baharlouei A, et al. Phytochemicals: Extraction, Isolation, and Identification of Bioactive Compounds from Plant Extracts. *Plants* 2017; **6**: 42.
- Saravanan KM, Zhang H, Senthil R, et al. Structural basis for the inhibition of SARS-CoV2 main protease by Indian medicinal plant-derived antiviral compounds. *J Biomol Struct Dyn* 2022; 40: 1970–1978.