Research Article

Comprehensive Study on the Bioactive Compounds and Biological Functions of *Senna hirsuta*

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ABSTRACT

India possesses one of the oldest, most diverse and rich traditions of medicinal plant use, with *Senna hirsuta* standing out as a robust, herbaceous plant known for its unique therapeutic properties. This study aimed to evaluate the phytochemical constituents, antioxidant capacity, and antibacterial potential of *Senna hirsuta*. Phytochemical analysis was performed using Soxhlet extraction with various solvents based on their polarity, revealing the presence of several bioactive compounds. The antibacterial activity of the ethanol extract was assessed in vitro using the disc diffusion method against five nosocomial bacterial strains: *Staphylococcus aureus* and *Bacillus subtilis* (Gram-positive), *Salmonella typhi*, *Escherichia coli*, and *Pseudomonas aeruginosa* (Gram-negative). The extract demonstrated inhibitory effects against all tested strains. Additionally, the antioxidant activity of the extract was evaluated, with results indicating a concentration-dependent increase in reducing power. These findings suggest that *Senna hirsuta* possesses significant antibacterial and antioxidant properties, which may contribute to its potential application in managing oxidative stress-related conditions and infections.

Keywords: *Senna hirsuta*, phytochemicals, antioxidant activity, antibacterial activity, nosocomial pathogens.

INTRODUCTION

Plant community represents one of the richest sources of bioactive compounds, which serves as the foundation for traditional medicine, modern pharmaceuticals, nutraceuticals, food supplements, and chemical intermediates for synthetic drugs [9]. The use of plant resources for the treatment of wide range of diseases dates back to ancient times that remains integral to primary healthcare, particularly in developing countries where approximately 80% of the population continues to rely on traditional medicine for their well-being [18]. Crude plant extracts containing phytochemicals with known antimicrobial and therapeutic properties are of increasing interest in the search for new and effective remedies. Phytochemicals, or phytoconstituents, are naturally occurring bioactive compounds produced by plants, often as part of their defense mechanisms against environmental stressors and pathogens[2]. These compounds, along with essential nutrients and fibers, contribute significantly to human health by enhancing resistance to disease and mitigating the effects of oxidative stress.

Phytochemicals are broadly classified into primary and secondary metabolites [20] based on their roles in plant physiology. Primary metabolites include sugars, amino acids, proteins, and chlorophyll, which are essential for plant growth and development. In contrast, secondary metabolites such as alkaloids, terpenoids, flavonoids, and steroids, although not directly

involved in growth, play critical roles in plant defense and have been recognized for their pharmacological significance[21]. Among these, antioxidants are vital due to their ability to neutralize free radicals—unstable molecules that can damage cellular components and contribute to the development of chronic diseases, including cancer and cardiovascular disorders. By scavenging free radicals, antioxidants help reduce oxidative stress, which is associated with cell damage and death. Consequently, antioxidants are widely used in dietary supplements and functional foods to promote health and prevent disease[15]. Due to the rising demand for natural therapeutic agents, the exploration of medicinal plants such as *Senna hirsuta* for their phytochemical content and bioactivities, including antioxidant and antimicrobial potential, holds significant promise for the development of novel pharmaceutical products.

MATERIALS AND METHODS

Collection of Plant Materials

Fresh samples of *Senna hirsuta* were randomly collected from Yercaud, Salem, Tamil Nadu, India. The plant materials were washed thoroughly with running water to remove soil and debris, then air-dried at room temperature. The dried materials were ground into a fine powder using a mechanical grinder and stored at 4°C in an airtight container until further use[3].

Preparation of Extracts

Soxhlet extraction was employed to obtain crude extracts from the powdered plant material. Approximately 20 g of *Senna hirsuta* powder was packed into a cellulose thimble and sequentially extracted with 250 ml each of hexane, ethyl acetate, and methanol. The extraction process was continued for 24 hours or until the solvent in the siphon tube appeared colorless[8]. Each solvent extract was

concentrated using a hot plate maintained at 30–40°C to evaporate the solvent. The resulting dried extracts were stored at 4°C for subsequent analysis.

Phytochemical Screening

Preliminary phytochemical screening of the hexane and ethanol extracts was conducted to detect the presence of bioactive compounds such as alkaloids, flavonoids, terpenoids, steroids, and others. The analysis followed standard protocols as described by Brain and Turner [5,3] .

Antimicrobial Assay

Test Organisms

Five bacterial strains commonly associated with nosocomial infections were selected for antimicrobial testing: *Staphylococcus aureus* and *Bacillus subtilis* (Gram-positive), and *Salmonella typhi*, *Escherichia coli*, and *Pseudomonas aeruginosa* (Gram-negative). All strains were obtained from the Microbial Type Culture Collection (MTCC), Institute of Microbial Technology, Chandigarh, India.

Preparation of Inoculum

Stock cultures were maintained on nutrient agar slants at 4°C. A loopful of each bacterial culture was inoculated into Mueller-Hinton Broth (MHB) and incubated at 37°C for 24 hours. The bacterial suspension was diluted with fresh MHB to achieve an inoculum density of approximately 2.0×10^6 CFU/mL, corresponding to the 0.5 McFarland standard. The antibacterial activity of the ethanol extract was evaluated using the disc diffusion method[4]. Mueller-Hinton Agar (MHA) was prepared by pouring 15 mL of molten medium into sterile Petri dishes. Once solidified, 0.1 ml of the bacterial suspension was uniformly swabbed onto the agar surface and allowed to dry for 5 minutes. Sterile 6 mm paper discs were impregnated with 40 mg of plant extract and placed on the inoculated plates. After allowing the extract to diffuse for 5 minutes, the plates were incubated at 37°C for 24 hours. Zones of inhibition around the discs were measured in millimeters using a transparent ruler.

Antioxidant Activity

Reducing Power Assay

The antioxidant activity of the extract was assessed based on its reducing power, following the method of (16). Briefly, the extract (or standard ascorbic acid) was mixed with 2.5 ml of phosphate buffer (0.2 M, pH 6.6) and 2.5 ml of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 minutes, then cooled and combined with 2.5 mL of 10% trichloroacetic acid. The mixture was centrifuged at 3000 rpm for 10 minutes. Five milliliters of the supernatant was mixed with 5 ml of distilled water and 1 mL of 0.1% freshly prepared ferric chloride. After a 10-minute reaction, absorbance was measured at 700 nm using a UV-Vis spectrophotometer. Higher absorbance values indicated stronger reducing power. Results were expressed as ascorbic acid equivalents (AAE) per gram of dry weight [13].

RESULTS AND DISCUSSION

Extraction Yield

Crude extracts of *Senna hirsuta* were obtained using ethanol, ethyl acetate, and hexane as solvents. Among the three, the ethanol extract yielded the highest quantity of phytoconstituents, approximately 20% more than the ethyl acetate and hexane extracts. Due to this higher yield and the known solvent efficacy of ethanol in extracting a broad spectrum of bioactive compounds, the ethanol extract was selected for further phytochemical, antibacterial, and antioxidant analysis.

Phytochemical Screening

Qualitative phytochemical analysis of the ethanol, ethyl acetate, and hexane extracts of *Senna hirsuta* revealed the presence of various secondary metabolites. The ethanol extract, in particular, showed the presence of alkaloids, flavonoids, steroids, tannins, phenols, and carbohydrates. These bioactive compounds are widely recognized for their therapeutic potential.

Flavonoids, detected in the ethanol extract, are known for their potent antioxidant properties, largely attributed to their role as free radical scavengers and water-soluble antioxidants [7,15]. They also possess anti-inflammatory activity, which supports their traditional use in treating wounds, burns, and ulcers. Steroids and Saponins, also present in the extract, are recognized for their antimicrobial, cardiotonic, and insecticidal properties [19] (Table 1). Phenolic compounds contribute significantly to antioxidative, antimicrobial, antidiabetic, and anticarcinogenic activities [1], highlighting the potential pharmacological value of *Senna hirsuta*.

Antibacterial Activity

The antibacterial efficacy of the ethanol extract of *Senna hirsuta* was assessed using the disc diffusion method against five pathogenic bacterial strains: *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Salmonella typhi*, and *Pseudomonas aeruginosa.* The results demonstrated significant antibacterial activity at higher extract concentrations, with *E. coli* being the most susceptible, followed by *S. aureus* (Fig 1a). No inhibition zones were observed at the lowest concentrations tested, indicating a dose-dependent antibacterial effect (Fig 1b).

The observed antibacterial activity could be attributed to the presence of bioactive compounds such as flavonoids, terpenoids, saponins, and phenols. These compounds are known to disrupt microbial cell membranes, inhibit nucleic acid synthesis, or interfere with metabolic processes. These findings support the potential of *Senna hirsuta* as a natural source of antibacterial agents and align with previous reports highlighting its use in the treatment of microbial infections [12].

Table 1: Phytochemical analysis

Phytochemicals	Extracts		
	Ethanol	Ethyl acetate	Hexane
ALKALOIDS			
Mayer's Test	+	-	-
Wagner's Test	+	-	-
FLAVONOIDS Lead Acetate Test	+	+	-
Sulphuric Acid Test	+	+	-
STEROIDS Liebermann –Buchard Test	+	-	+
TERPENOIDS Salkowski Test	-	-	-
ARTHROQUINONE Borntrager's Test	-	-	-
PHENOLS Ferric Chloride Test	+	-	-
Lead Acetate Test	+	-	-
SAPONIN	-	-	-
TANNIN	+	-	-
CARBOHYDRATE	+	+	+
OILS & RESINS	-	-	+

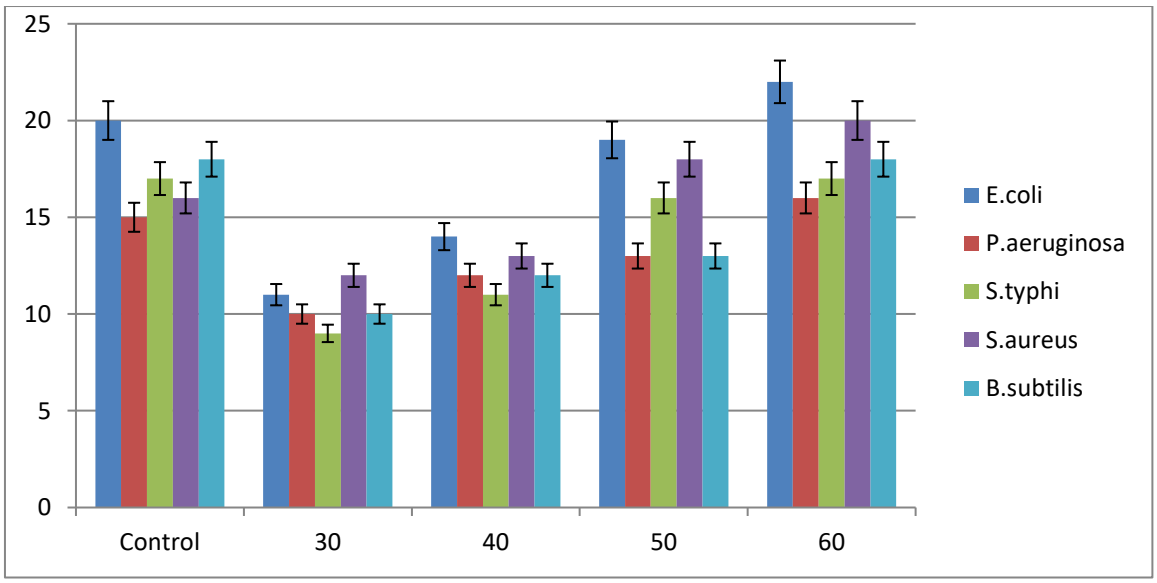

Figure 1a: Antibacterial activity of *Senna hirsuta*

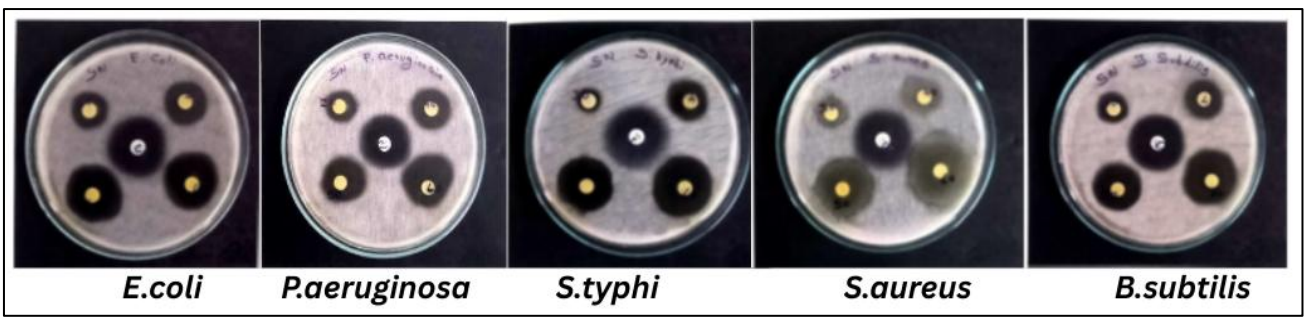

Figure 1b: Antibacterial activity of *Senna hirsuta*

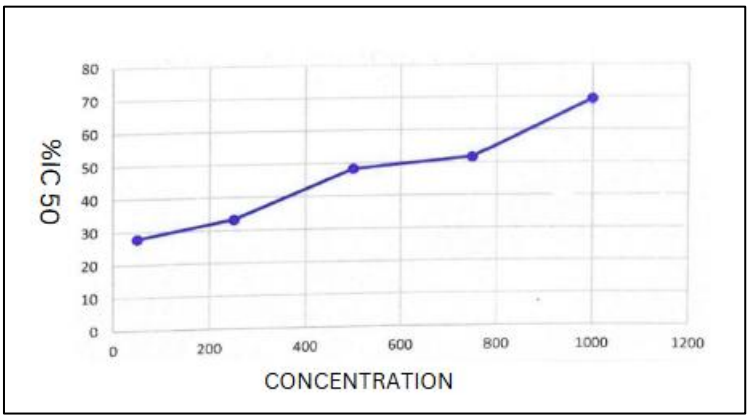

Figure 2: Reducing power assay of *Senna hirsuta* in ethanol extract

Antioxidant Activity

The reducing power assay was employed to evaluate the antioxidant potential of *Senna hirsute* ethanol extract. The assay demonstrated a concentration-dependent increase in absorbance at 700 nm, indicating higher reducing power with increasing extract concentration (Fig 2). The extract effectively reduced Fe^{3+} to Fe^{2+}, as evidenced by the formation of Perl's Prussian blue complex, confirming its electron-donating ability.

This result is consistent with the premise that antioxidant activity correlates with reducing power [11,14]. The extract's capacity to donate electrons suggests its potential to neutralize free radicals and inhibit oxidative processes such as lipid peroxidation [22]. Compounds exhibiting strong reducing power often function as primary and secondary antioxidants by halting free radical chain reactions and stabilizing oxidized intermediates [6,21].

These findings collectively underscore the pharmacological promise of *Senna hirsuta* and validate its traditional applications. Its ethanol extract, rich in phytoconstituents, displays substantial antibacterial and antioxidant activities, warranting further investigation into its active constituents and potential therapeutic applications[10].

The study has investigated the phytochemical composition, antibacterial potential, and antioxidant activity of *Senna hirsuta*, a medicinal plant traditionally used in various healing practices by ancient tribal people. Plant material was collected from the foothills of Yercaud, Tamil Nadu, shade-dried, and subjected to Soxhlet extraction using hexane, ethyl acetate, and ethanol as solvents. The resulting extracts were evaluated for the presence of phytochemical constituents using standard qualitative assays.

The ethanol extract's antibacterial activity was assessed using the disc diffusion method against five clinically relevant bacterial strains: *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Staphylococcus aureus*, and *Bacillus subtilis*. Among these, *E. coli* exhibited the highest susceptibility to the ethanol extract, while all strains demonstrated a dose-dependent response.

Antioxidant activity was examined using the reducing power assay, and results indicated that the ethanol extract possessed concentration-dependent antioxidant potential. This activity is likely ascribed to the presence of bioactive compounds such as flavonoids, phenols, and other reducing agents.

CONCLUSION

This study confirms that *Senna hirsuta* possesses significant pharmacological potential. Qualitative phytochemical screening of ethanol, ethyl acetate, and hexane extracts revealed that the ethanol extract contained a rich profile of secondary metabolites, including alkaloids, flavonoids, steroids, carbohydrates, and phenolic compounds. The ethanol extract exhibited notable antibacterial activity, with *E. coli* being the most susceptible strain. Additionally, the extract showed promising antioxidant capacity in a concentration-dependent manner.

The observed bioactivities can be imputed to the presence of specific phytochemicals, suggesting that *Senna hirsuta* may serve as a valuable source of natural compounds with therapeutic relevance. Further investigation into the isolation, purification, and structural elucidation of the active constituents is warranted to explore their full potential in pharmaceutical applications, particularly as antibacterial and antioxidant agents.

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REFERENCES

1. Abubakar, E.M. Antibacterial Activity of Crude Extracts of Euphorbia hirta against Some Bacteria Associated with Enteric Infections, *Journal of Medicinal Plants Research* July 2009;3(7):498-505.
2. Ahmad, I. and Beg, A.Z. Antimicrobial and Phytochemical Studies on 45 Indian Medicinal Plants against Multi-Drug and Resistant Human Pathogens. Journal of Ethnopharmacology, 2001;74:113-123.

3. Balkrishna, T., Ashwani, T., &Himani, S. Genetic Variability And Heritability In Growth And Leaf Associated Traits Of Selected *Dalbergia Sissoo* Genotypes. *Journal of Tropical Forest Science*, 2025;37(1):85-95.
4. Bauer AW, Kirby WM, Sherris JC, Turck M. Antibiotic susceptibility testing by a standardized single disk method. *American Journal of Clinical Pathology*, 1966 Apr:45(4):493-6.
5. Brain, K.R., and T.D. Turner. The practical evaluation of phytopharmaceuticals, Wright science technical, 1st Ed, Bristol Britain., 1975, 144.
6. Chanda, S. and Dave, R. In Vitro Models for Antioxidant Activity Evaluation and Some Medicinal Plants Possessing Antioxidant Properties An Overview. African Journal of Microbiology Research, 2009;3:981-996.
7. Doss A. Preliminary phytochemical screening of some Indian Medicinal Plants. Anc Sci Life. 2009 Oct;29(2):12-6.
8. Fereja,W. M., Geleta, W. D., Desalegn, E., Moaneanda, M., Abdissa, A., &Usamo, F. B. In vitro antioxidant and antibacterial activities of *Ajugainte grifolia* leaf extracts obtained with different solvents. BMC Complementary Medicine and Therapies 2024. Oct 12; 24(1):368-72.
9. Hammer KA, Carson CF, Riley TV. Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology. 1999 Jun; 86(6):985-90.
10. Köksal E, Tohma H, Kılıç Ö, Alan Y, Aras A, Gülçin İ, Bursal E. Assessment of Antimicrobial and Antioxidant Activities of Nepeta trachonitica: Analysis of Its Phenolic Compounds Using HPLC-MS/MS. Sci Pharm. 2017 May 15;85(2):24-9.
11. Manikandan S, Devi RS. Antioxidant property of alpha-asarone against noise-stress-induced changes in different regions of rat brain. Pharmacological Research. 2005 Dec;52(6):467-74.
12. Manjunatha, B.K. Antimicrobial Activity of *Pterocarpus santalinus*. Indian Journal of Parmaceutical Sciences, 2006;68:115-116.
13. Nicolae-Valentin, V., Simion, A., & Despina-Maria, B. Nutrient Status and Antioxidant Activity of the Invasive *Amaranthus retroflexus* L. Sustainability, 2025;17(11):5141-9.
14. Oktay, M., Gülçin, İ. and Küfrevioğlu, Ö. Determination of in Vitro Antioxidant Activity of Fennel (*Foeniculum vulgare*) Seed Extracts. LWT-Food Science Technology, 2003;36:263-271.
15. Okwu, D.E. Phytochemical and Vitamin Content of Indigenous Species of South- Eastern Nigeria, Journal of Sustainable Agriculture and the Environment. 2004;6(1):30-37.
16. Oyaizu M , Studies on product of browning reaction prepared from glucose amine. The Japanese Journal of Nutrition and Dietetics, 1986;44:307–315.
17. Pant, D. D., Tapwal, A., Tiwari, B., & Thakur, J. In vitro evaluation of antifungal potential of selected plant species against *Fusarium solani*. Asian Journal of Natural Product. 2023;21(2):51-57.
18. Rainer W Bussmann & Douglas Sharon Traditional medicinal plant use in Loja provin, Southern Ecuador. 2006, volume 2, 44-52.
19. Ravanappan Srinivasan Ramya, Akshaya A An Analysis on the Larvicidal Activity of Banana Peel Extract and its Silver Nanoparticles against *Culex* species Endemic In Adyar, Tamil Nadu, India JETIR December. 2024;11(12):2349-5162.
20. Salah W, Miller NJ, Pagauga G, Tijburg, Bolwell GP, Rice E, Evans C 1995. Polyphenolic flavonols as scavenger of aqueous phase radicals and chain-breaking antioxidants. Arch. Biochem.Biol. 1995; 2: 339-346.
21. Saranya VTK, Uma Gowrie S. Phytochemical analysis and in vitro studies on Antibacterial, Antioxidant and Anti-inflammatory activities using *Casuarina equisetifolia* bark extracts. Int J Pharm Pharm Sci. 2018 a; 10(1): 0975-1491.
22. Sreejayan N, Rao MNA (1997): Nitric oxide scavenging by curcuminoids. J Pharm Pharmacol. 1997;49: 105–107.