

ORIGINAL ARTICLE

Exploring the Antimicrobial Efficacy and Phytochemical Profile of Solvent Extracts from *Eclipta* speciesPalaniyandi Velusamy¹, Sasikumar Pattabi², A. Emaya³, Periasamy Anbu⁴¹ Innovation and Incubation Centre for Health Sciences (IICHS), Sree Balaji Medical College and Hospital (SBMCH), Bharath Institute of Higher Education and Research (BIHER), Chromepet 600 044, Tamil Nadu, India.² Department of General Surgery, Sree Balaji Medical College and Hospital (SBMCH), Bharath Institute of Higher Education and Research (BIHER), Chromepet 600 044, Tamil Nadu, India.³ Department of Obstetrics and Gynecology, Saveetha Medical College & Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Thandalam, Chennai – 602 105, Tamil Nadu, India.⁴ Department of Microbiology, Saveetha Medical College & Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Thandalam, Chennai – 602 105, Tamil Nadu, India.

ABSTRACT

Introduction: *Eclipta* species are known for their rich phytochemical composition and medicinal properties. This study aimed to evaluate the extraction efficiency of ethyl acetate, and methanol as solvents for *Eclipta* sp. and to assess the antibacterial potential of the extracts against key bacterial pathogens. **Methods:** Extracts were prepared using ethyl acetate and methanol as solvents, and their yields were quantified. Phytochemical analysis identified bioactive compounds, including saponins, tannins, flavonoids, alkaloids, and terpenoids. Antibacterial activity was evaluated using the agar well diffusion method against *Pseudomonas aeruginosa*, *Bacillus subtilis*, and *Staphylococcus aureus* at a concentration of 150 µg/mL. **Results:** Phytochemical analysis revealed that ethyl acetate extracted the widest range of bioactive compounds. Antibacterial tests showed significant activity of *Eclipta* sp. extracts, with ethyl acetate exhibiting the most potent effects. The inhibition zones for *P. aeruginosa*, *B. subtilis*, and *S. aureus* were 16 mm, 11 mm, and 10 mm, respectively. The superior activity against Gram-negative *P. aeruginosa* suggests the extract's potential to target pathogens with varied cell wall structures. **Conclusion:** Ethyl acetate was the most effective solvent for extracting bioactive compounds from *Eclipta* sp., yielding extracts with notable antibacterial activity, particularly against *P. aeruginosa*. These findings highlight the potential of *Eclipta* sp. as a source of antibacterial agents and the efficacy of ethyl acetate as a solvent for phytochemical extractions.

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INTRODUCTION

The quest for new therapeutic agents is an ever-evolving field, where both recent drug discoveries and natural products play crucial roles in advancing medicine. Natural products, particularly those derived from plants and marine organisms, have historically been a rich source of medicinal compounds that significantly enhance human health and well-being (1-3). These natural substances have inspired the development of novel medicinal molecules and provided direct contributions to therapeutic practices.

Plants, through their production of secondary metabolites,

offer a diverse array of bioactive compounds that exhibit a broad spectrum of activities. These metabolites, which include alkaloids, flavonoids, saponins, and terpenoids, have demonstrated notable antibacterial, antifungal, antiviral, antineoplastic, and anticancer properties. Additionally, they can act as inhibitors of various enzymes and as plant growth promoters (4). The structural uniqueness of these compounds, such as the presence of halogen atoms and stereogenic centers, often derived from complex interactions with inorganic species in their natural habitats, enhances their potential for commercial applications (5).

Marine natural products have gained significant attention due to their distinctive biological properties and potential as bioactive substances. These compounds, originating from marine organisms, offer unique chemical structures and biological activities that are not typically found in terrestrial counterparts. Their strong activity and

selectivity make them ideal candidates for developing novel molecules with specific therapeutic qualities (6,7). The exploration of marine natural products has revealed promising candidates for pharmaceutical applications, serving either as lead compounds for drug development or as templates for the synthesis of new organic molecules.

The advancement of chemical compound research has furthered our understanding of these natural substances. The processes of isolation, biosynthesis, and structural elucidation of natural products have significantly impacted both agricultural and medical fields. Innovations in these areas have led to the creation of new chemicals with potential applications in managing diseases and pests. This ongoing research is critical for uncovering novel compounds that can address current challenges in health and agriculture, demonstrating the profound potential of natural products in shaping future therapeutic strategies (8). In this study, we assessed how effectively various solvents, including ethyl acetate, and methanol extract bioactive compounds from *Eclipta* sp. Additionally, we evaluated the antibacterial activity of these extracts against a range of bacterial strains to determine their potential as antimicrobial agents.

MATERIALS AND METHODS

Collection and identification of plant material

Fresh plant parts were collected from Thandalam, Kancheepuram District, Tamil Nadu. The plant material is washed in running tap water, air-dried, homogenized to fine powder; and then stored in air-tight bottles. The dried powder was used to check various compounds including alkaloids, flavonoids, tannin, phenol, and saponin.

Extraction of plant material

About 10 g of air-dried powder was taken in 100 ml of ethyl acetate or methanol in a conical flask, plugged with cotton wool, and then placed on a rotary shaker at 180 rpm for 24 h. After 24 h the supernatant was collected, and the solvent was evaporated to make the final one-fourth of the original volume. The sealed bottles were then refrigerated at 4°C.

Qualitative and quantitative estimation of the plant extracts

The plant extract was assessed using both qualitative and quantitative analysis to determine the presence of different components such as phenol, saponin, flavonoids, and alkaloids.

Preparation of bacterial suspension and antibacterial effect

A disc diffusion experiment was used to assess the antibacterial activity of plant extract against *Bacillus subtilis*, *Escherichia coli*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus* (9,10). To

produce bacterial suspensions, bacterial strains were cultivated in nutritional broth for 24 hours at 37 °C (180 rpm). The suspensions (1×10^6) were then evenly distributed on LB agar plates utilizing a sterile glass spreader. The inoculation plates were carefully topped with sterile filter paper discs (6 mm in diameter), onto which various quantities of plant extract (50, 100, and 150 mg/mL) and sterile water as a control were loaded onto each disc. Next, the inoculated plate was incubated at 37°C for 24 h. Following incubation, an estimate of the inhibitory zone using various plant extract concentrations was made.

RESULTS

Extraction and Yield

The extraction process of *Eclipta* sp. using ethyl acetate and methanol revealed significant differences in yield. Ethyl acetate yielded the highest extractable material at 7.3 mg/g (Table I), showcasing its superior extraction

Table I: Different solvents were used for the extraction.

| S. No. | Solvents | Yield in (mg/g) |
|--------|---------------|-----------------|
| 1 | Ethyl acetate | 7.3 |
| 2 | Methanol | 5.6 |

efficiency compared to methanol.

Phytochemical Profiling

Phytochemical analysis demonstrated that ethyl acetate was highly effective in extracting various bioactive compounds, including saponins, tannins, flavonoids, alkaloids, and terpenoids (Table II). Notably, saponins were exclusively extracted by ethyl acetate, indicating their capability to capture a broader spectrum of phytochemicals.

Antibacterial Activity

The antibacterial potential of *Eclipta* sp. extract was assessed against *B. subtilis*, *P. aeruginosa*, and *S. aureus* using the agar diffusion method at different sample concentrations (50, 100, and 150 µg/mL). The diameter of the *Eclipta* sp.-produced inhibitory zone surrounding

Table II: List of compounds extracted from the *Eclipta* sp. using various solvents.

| Compounds | Ethyl acetate Extract | Methanol Extract |
|------------|-----------------------|------------------|
| Saponins | + | - |
| Tannins | + | + |
| Flavonoid | + | + |
| Alkaloids | + | + |
| Terpenoids | + | + |

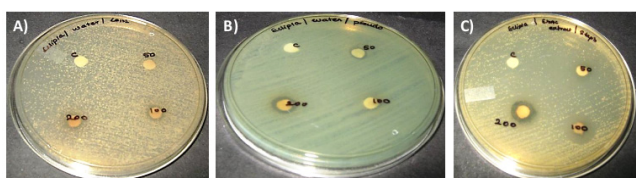


Figure 1: Inhibitory zone of plant extract. A) *B. subtilis*, B) *P. aeruginosa*, C) *S. aureus*.

the disc is displayed in Figure 1. At a concentration of 150 $\mu\text{g/mL}$, the extract exhibited the highest inhibitory activity against *P. aeruginosa* with a 16 mm inhibition zone, followed by *B. subtilis* (11 mm) and *S. aureus* (10 mm). Lower concentrations (50 $\mu\text{g/mL}$) resulted in reduced activity, with *S. aureus* showing no inhibition at this level (Figure 1 and 2). The extract also exhibited significant antibacterial activity against *B. subtilis* and *S. aureus*, with inhibition zone diameters of 11 mm and 10 mm, respectively, at the 150 $\mu\text{g/mL}$ concentration. At the lower concentration of 50 $\mu\text{g/mL}$, *S. aureus* did not exhibit any inhibitory activity. In contrast, *P. aeruginosa* and *B. subtilis* still displayed some level of inhibition at this lower concentration.

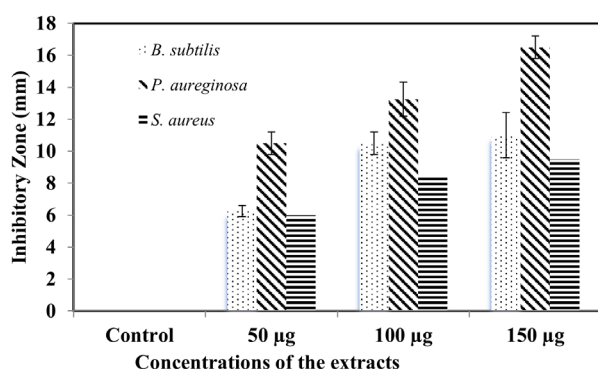


Figure 2: Antibacterial Activity of *E. alba* Extracts at Different Concentrations

DISCUSSION

Ethyl acetate's high extraction yield of 7.3 mg/g reflects its efficiency as a solvent, attributed to its unique polarity, which balances the solubilization of non-polar and semi-polar compounds. The solvent's unique polarity makes it particularly effective for capturing various bioactive constituents found in *Eclipta* sp., which are known for their potential health benefits. Consequently, the high yield of extractable material not only emphasizes the solvent's efficiency but also suggests that ethyl acetate may facilitate the isolation of bioactive compounds with significant therapeutic potential, reinforcing its value in phytochemical studies and applications (11).

Ethyl acetate's effectiveness in extracting a wide range of bioactive compounds, particularly saponins, highlights its versatility. The absence of saponins in methanol extracts underscores ethyl acetate's unique ability to solubilize certain phytochemicals. The presence of

these compounds is significant, as they are often linked to various health benefits, including anti-inflammatory, antioxidant, and antimicrobial activities. This finding aligns with previous research that emphasizes ethyl acetate's efficacy in extracting a wide range of bioactive compounds, suggesting its critical role in enhancing the therapeutic value of plant extracts (12).

Several studies have documented the plant extract's ability to effectively suppress pathogenic bacterial strains due to its rich bioactive and antibacterial components (13-16). The highest inhibition zone was observed against *P. aeruginosa* at 150 $\mu\text{g/mL}$, with a diameter of 16 mm. This indicates a strong antibacterial effect, suggesting that the extract contains potent bioactive compounds capable of effectively inhibiting this Gram-negative bacterium. *P. aeruginosa* has a relatively thin peptidoglycan layer surrounded by an outer membrane containing lipopolysaccharides, which can sometimes facilitate the entry of antibacterial agents into the cell, making them more susceptible to antimicrobial compounds (17,18). The thicker peptidoglycan layer in *B. subtilis* and *S. aureus* provides additional protection and makes it more challenging for antibacterial agents to penetrate and disrupt cellular functions (19-21). Despite this, the *Eclipta* sp. extract managed to inhibit both Gram-positive strains, though to a lesser extent compared to the Gram-negative strain.

CONCLUSION

This study establishes ethyl acetate as the most effective solvent for extracting bioactive compounds from *Eclipta* sp. Ethyl acetate's superior performance is attributed to its ability to solubilize a broad range of non-polar and semi-polar phytochemicals, including saponins, tannins, flavonoids, alkaloids, and terpenoids, which are less efficiently extracted by methanol. Furthermore, the antibacterial activity of the ethyl acetate extract was particularly noteworthy, showing strong inhibition against *P. aeruginosa* with an inhibition zone of 16 mm at 150 $\mu\text{g/mL}$. This suggests the extract contains potent antibacterial compounds that are effective against this Gram-negative bacterium. Additionally, the extract displayed significant activity against *B. subtilis* and *S. aureus* (Gram-positive bacteria), although with less potency, likely due to the thicker peptidoglycan layers in these bacteria. This study offers valuable insights into optimizing the extraction of plant-derived bioactive substances, which could lead to more effective applications in the medicinal and pharmaceutical industries.

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