Research Article

Preliminary phytochemical investigations and Antibacterial activity of some Medicinal plants

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Abstract

Aim: This study evaluates the preliminary phytochemical screening and antimicrobial effects of petroleum ether extract of five selected medicinal plants of *Cardiospermum halicacabum* (L.) leaves, *Ervatamia divaricata* (L.) Burkill leaves, *Curcuma zedoaria* rhizome, *Clerodendrum inerme* (L.) Gaertn leaves and *Sida rhombifolia* L. leaves.

Methods: Dried plant materials of selected plants were extracted with petroleum ether in a Soxhlet apparatus for 3hr. The collected extracts were screened for phytochemicals and studied for their antibacterial activity using the agar disc diffusion technique.
Results: The result of the present study observed that extraction values of *C.inerme* (L.) Gaertn exhibit the highest yield at 6.4%, while the extract from the rhizome of *C.zedoaria* shows the lowest yield at 2.2%. The preliminary phytochemical investigation of petroleum ether extracts of the leaves of *C.halicacabum* (L.), *E.divaricata* (L.) Burkill, and *C.inerme* (L.) Gaertn as well as the rhizome of *C.zedoaria* and the leaves of *Sida rhombifolia* L., revealed that this plant tested for positive results were alkaloids, flavonoids, and terpenoids while active compounds of Tannin and saponins were absent. The medicinal plants of Curcuma zedoaria rhizome had good antibacterial activity against S. aureus and Bacillus subtilis.
Conclusion: The petroleum ether extracts of the chosen plants were examined in this study indicate that these extracts may serve as more effective for antimicrobial agents.

Keywords: Antimicrobial activity, plant extract, Gram positive, Gram negative

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1. Introduction

The planet hosts a remarkable diversity of 391,000 vascular plants, with at least 35,000 species potentially harboring medicinal properties. Medicinal plants have been used by indigenous physicians since pre-Hispanic times and are part of the traditional knowledge of humanity. Medicinal plants are species within the plant kingdom, whose flowers, leaves, roots, stems, fruits, or seeds are used directly or in preparations to treat medical conditions or diseases. Medicinal plants contain several secondary metabolites with valuable pharmacological properties such as antitumor, antiviral, antiepileptic, antibiotic, anti-inflammatory, and antinociceptive agents (Alonso-Castro *et al*., 2011). Moreover, plants are the source of 25% of the bioactive compounds used in modern medicines, and traditional herbal remedies remain the primary health care option for at least 80% of the world's population. The use of natural products for primary health care has increased worldwide over the last decade (Martins Ekor,2014). The global market for medicinal plants and plant-derived drugs was estimated at $25.6 billion in 2015. Many medicinal plants were lacking scientific evidence regarding their toxicity, chemical composition, and pharmacological effects. The present study investigated the phytochemicals and antibacterial activity of medicinal plant materials collected from a local herb shop in Tirunelveli, Tamil Nadu. The plant materials included leaves from *Cardiospermum halicacabum* (L.), *Ervatamia divaricata* (L.) Burkill, *Clerodendrum inerme* (L.) Gaertn, and *Sida rhombifolia* L., as well as the rhizome of *Curcuma zedoaria*.

2. Materials and Methods

2.1 Plant Materials

The plant materials of *Cardiospermum halicacabum* (L.), leaves, *Ervatamia divaricata* (L.) Burkill, leaves, *Curcuma zedoaria* Rhizome, *Clerodendrum inerme* (L.) Gaertn leaves and *Sida rhombifolia* L. leaves were collected from the local herb shop, Tirunelveli, Tamilnadu.

2.2 Extraction

Each plant materials were air dried and coarsely powdered. One hundred grams of dried powdered materials were extracted with 250 ml of petroleum ether at 40 °C for 2h in a Soxhlet apparatus and filtered and collect the extract. These extracts were excess solvents removed by rotatory evaporation at 40 °C till complete dryness occurs. The total extract was dissolved in petroleum ether at a concentration of 500 mg/ml and stored at -20 °C for further use.

2.3 Preliminary phytochemical screening

Petroleum ether extracts from the leaves of *C.halicacabum* (L.), *E.divaricata* (L.) Burkill, and *C.inerme* (L.) Gaertn, as well as the rhizome of *C.zedoaria* Rosc and the leaves of *S.rhombifolia* L., underwent preliminary phytochemical screening for using various method described. To assess the presence of alkaloids, 0.5 g of the crude extract was combined with 5 ml of 1% HCl on a steam bath. Mayer's reagent was added to 1 ml of the filtrate, followed by an equivalent amount of Dragendorff's reagent in another sample. The formation of turbidity or precipitation with both reagents indicated preliminary evidence of alkaloids. For saponins, 0.5 g of the crude plant extract mixed with water in a test tube, utilizing a mobile phase of chloroform, glacial acetic acid, methanol, and water, and a vanillin-H^2SO_4 spraying reagent for detection. The appearance of blue, blue-violet, red, or yellow-brown zones was considered a positive indication of saponins. To test for tannins, 10 ml of distilled water was combined with 0.5 g of the crude extract, followed by filtration. The addition of $FeCl_3$ reagent to the filtrate, resulting in blue, blue-black, green, or blue-green coloration or precipitation, was interpreted as a sign of tannins. For anthraquinones, 0.5g of the extract were mixed with 10ml of benzene and filtered. The filtrate was then mixed with 5ml of a 10% ammonium hydroxide solution and agitated; the presence of a pink, red, or violet color in the ammoniacal phase indicated anthroquinones. To detect polyphenols, 2 ml of the crude extract was treated with 3 drops of a solution containing 1 ml of 1% $FeCl_3$ and 1 ml of 1% $C_6N_6FeK_3$, with the formation of a green-blue color signifying their presence. Lastly, for flavonoids, 2 ml of the alcoholic solution of the crude extract was mixed with 4 drops of a 2% lead acetate solution, where the appearance of a yellow or orange tint indicated the presence of flavonoids.

2.4. Antimicrobial activity

2.4.1 Test organisms

Four types of bacteria were investigated by the four bacteria : Gram positive (*Staphylococcus aureus* and *Bacillus cereus*), and Gram negative (*Escherichia coli* and *Psudomonas auriginosa*).

2.3.1. Disc diffusion method

The inocula of the various bacteria were streaked over the Mueller Hinton agar plates to completely cover the plates and become uniform after incubation. Using a sterile cork borer, 10 mm diameter wells were made on Mueller-Hinton plates. Each well was filled with 100 µl of the test substances and the plates were left at room temperature for 2h. The plates were then incubated for 24h at 37 °C. The crude extract was prepared at concentration of 100 mg/ml by dissolving the dried extracts in petroleum ether. Gentamycin was used as a positive control at a concentration of 0.1 mg/ml. Petroleum ether served as a negative control. The diameter of the zones of inhibition was used to calculate the antibacterial activity of all samples (Andrew, 2001; Valgas et al.,2007). The data were entered into an excel spreadsheet using the Statistical Package for Social Science (SPSS)

3. Results and Discussion

3.1 Phytochemical analysis

3.3.1 Medicinal values

The medicinal values of the chosen medicinal plant uses are represented in the table-1. The Leaf materials are contained rich in essential bioactive compounds, including tocopherols, carotenoids, flavonoids, sterols, vitamins, tannins, lipids, and minerals, which have applications in the health, beauty, and food sectors. The isolation of bioactive compounds from medicinal plants, which is applicable in numerous formulations and products, exhibits a range of beneficial properties, including anti-aging, antibacterial, anticancer, anti-depressive, antifungal, anti-inflammatory, antioxidant, antiparasitic, antiviral, and anti-stress effects. These compounds are considered biorenewable high-value chemicals.

3.3.2 Extraction values

The yields of the extracts from five chosen medicinal plants are presented in Table 2. The extract of *Clerodendrum inerme* (L.) Gaertn exhibits the highest yield at 6.4%, while the extract from the rhizome of *Curcuma zedoaria* shows the lowest yield at 2.2%.

Earlier studies, acetone extract of Hypericum roeperianum had the highest yield (12%), followed by Maesa lanceolata (11.12%) and the lowest extraction yield was obtained with Morus mesozygia 1.85% (Elisha *et al*.,2017).

3.3.3 Preliminary phytochemical screening

In the present study observed that preliminary phytochemical investigation of petroleum ether extracts of the leaves of *Cardiospermum halicacabum* (L.), *Ervatamia divaricata* (L.) Burkill, and *Clerodendrum inerme* (L.) Gaertn, as well as the rhizome of *Curcuma zedoaria* and the leaves of Sida rhombifolia L., revealed that these plant tested for positive results were alkaloids, flavonoids, and terpenoids while active compounds of Tannin and saponins were absent in the present study (Table-2). Previous studies, Wasihun *et al*., (2023) identified the active compounds of alkaloids, saponins, flavonoids, and tannins present in the ethanolic leaf extract of the plant *Calpurnia aurea*. The phytochemical characteristics of various species were examined, such as *Artemisia afra* (Ariti), Aloe Vera (Erret), *Syzygium guineense* (Dokuma), *Ruta chalepensis* (Tenadam), *Ocimum gratissimum* (Damakese), *Nigella sativa* (Tikur Azmud), *Lepidium sativum* (Feto), *Hagenia abyssinica* (Kosso), *Croton macrostachyus* (Bisana), and *Rhamnus prinoides* (Gesho) (Agidew,2022). This finding results, suggests the plant has potential therapeutic applications for treating skin and systemic infections. Further research is warranted to conduct a comprehensive comparative study, including quantitative phytochemical analysis of the most antibacterial fractional extracts of the plant's tannins, alkaloids, flavonoids, saponins, and other compounds. The variation in the presence of various chemicals can be attributed to factors such as the type of solvent used, the extraction method employed, the characteristics of the soil, and the age of the plant, as noted by Hu et al., (2017).

Table-1: Yield of the extracts in selected medicinal plants

Sl.No.	Medicinal plants	% of the extracts yield
1	*Cardiospermum halicacabum* (L.)	5.8
2	*Ervatamia divaricata* (L.) Burkill	3.7
3	*Curcuma zedoaria* Rosc	2.2
4	*Clerodendrum inerme* (L.) Gaertn	6.4
5	*Sida rhombifolia* L.	5.2

3.3.3 Preliminary phytochemical screening

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Table-2: Preliminary phytochemical screening

Sl. No.	Medicinal plants	Plant Parts	Bio active compounds				
			Alkaloids	Flavonoids	Tannin	Saponin	Terpenoids
1	*Cardiospermum halicacabum* (L.)	Leaves	+++	+	-	-	+
2	*Ervatamia divaricata* (L.) Burkill	Leaves	+	+	-	-	+
3	*Curcuma zedoaria* Rosc	Rhizome	+	+	-	-	++++
4	*Clerodendrum inerme* (L.) Gaertn	Leaves	+	+	-	-	+
5	*Sida rhombifolia* L.	Leaves	+	+	-	-	+

Table-3: Antibacterial activity of some medicinal plants

Plants	Zone of Inhibition in bacterial strains (mm)			
	Bacillus subtilis	*E. coli*	*S. aureus*	*Psudomonas aurigi-nosa*
Cardiospermum halicacabum (L.)	12	15	13	15
Ervatamia divaricata (L.) Burkill	16	14	11	12
Curcuma zedoaria Rosc.	17	16	18	17
Clerodendrum inerme (L.) Gaertn	11	13	12	11
Sida rhombifolia L.	11	13	15	12
Pencilline	13	-	12	-
Strptomycene	-	14	-	15

Conclusions
The conclusion of the present study was a preliminary phytochemical analysis of five chosen five plant extracts involves a variety of tests to identify the presence of active components like terpenoid or steroids, flavonoids, and alkaloids while tannins and saponins are absent. Petroleum ether extracts of these plants can inhibit the good antibacterial activity.

Ethics approval and consent to participate
Ethics approval and consent to participate are not relevant in this case, as our research did not involve animals or human subjects.

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Conflicts of interest
There are no existing conflicts of interest.

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