Botany
Explore–Journal of Research
Peer Reviewed Journal
ISSN 2278 – 0297 **(Print)**
ISSN 2278 – 6414 **(Online)**

https://www.patnawomenscollege.in/journal

Phytochemical screening and antimicrobial study of *Euphorbia hirta* Linn extracts

• **Muskan Sinha** • **Sujata Kumari** • **Jagriti Priya**
• **Shilpi Kiran**

Received : December, 2021
Accepted : January, 2022
Corresponding Author : Shilpi Kiran

Abstract: *In India, folklore medicinal herbs possess long-established applications that can be utilized for the treatment of various bacterial infections. Medicinal plants possess therapeutic properties due to the presence of many chemical substances, which are present as secondary plant metabolites in various parts of these plants. Owing to the availability of these secondary constituents, medicinal plants exhibit antibacterial and anti-inflammatory activities. The present study was aimed to analyse the phytochemical screening and investigate antimicrobial activities of methanolic, ethanolic and aqueous extract of leaf, stem and whole plant of Euphorbia hirta. Phytochemical screening showed the presence of alkaloids, flavonoids, saponins, carbohydrates, cardiac glycosides in the extracts of different parts (stem, leaves, and whole plant) of Euphorbia hirta. Antimicrobial study of various extracts was done by disc diffusion method against Staphylococcus aureus, Proteus vulgaris and Escherichia coli. The anti-microbial study revealed that methanolic extract of E. hirta showed highest anti- bacterial activity against S. aureus and E. coli, while ethanolic extract against P. vulgaris. This reveals the importance of choice of solvent for proper extract preparation. The present work suggests promoting utilization of weeds like E. hirta for the benefit of mankind so as to prevent the over-exploitation of endangered medicinal plants.*

Keywords : *Euphorbia hirta, phytochemicals, antibacterial activity, S. aureus, E. coli, P. vulgaris.*

Muskan Sinha
B.Sc. III year, Botany (Hons.), Session: 2019-2022,
Patna Women's College (Autonomous),
Patna University, Patna, Bihar, India

Sujata Kumari
B.Sc. III year, Botany (Hons.), Session: 2019-2022,
Patna Women's College (Autonomous),
Patna University, Patna, Bihar, India

Jagriti Priya
B.Sc. III year, Botany (Hons.), Session: 2019-2022,
Patna Women's College (Autonomous),
Patna University, Patna, Bihar, India

Shilpi Kiran
Assistant Professor, Department of Botany,
Patna Women's College (Autonomous),
Bailey Road, Patna–800 001, Bihar, India
E-mail : shilpikpwc@gmail.com

Introduction:

Microorganisms are responsible for innumerable infectious diseases that leads to death worldwide. In the last few decades the antibiotic resistant infection causing microorganisms has become a global threat to public health. Emergence of multidrug resistant pathogens has threatened clinical efficacy of many existing antibiotics (Westh et al., 2004; Bandow et al., 2003). This has led researchers to discover new compounds of plant origin with antimicrobial activity. In this regard phytomedicines have gained much attention and have become popular and their utility is widespread.

India is tremendously rich in aromatic and medicinal plants. Since time immemorial, plants have

been considered for their biological properties to cure illness and prevent diseases and several drugs have been formulated by the application of bioactive compounds found in these medicinal plants (Rahmati et al., 2015). These phytomedicines are beneficial to mankind, secure by all means and are environment friendly (Bansod and Rai, 2008). Plants manufacture a wide range of bioactive molecules known as phytochemicals (Suresh et al., 2012; Ramproshad et al., 2012). They are incorporated naturally in all plant parts; bark, leaves, stem, root, flower, fruits, seeds, etc. Usually, these phytochemicals are secondary metabolites such as flavonoids, steroids, alkaloids, resins, fatty acids, tannins, etc. Regarding the vast potentiality of plants as genesis for antimicrobial drugs with respect to antibacterial and antifungal agents, a systematic examination was carried out to screen the phytochemical constituents and antimicrobial activity of Euphorbia hirta (Ahmad et al., 2017).

The plant Euphorbia hirta is a small annual herb, often found in open waste spaces, roadsides, grasslands, pathways, rice fields. It is native to Central America. It is said to be a plant of medicinal importance inspite of being a weed and is regarded as an outstanding medication to treat respiratory system disorders including asthma, bronchitis, hay fever, coughs and colds (Asha et al., 2014). By the phytochemical screening and study of antimicrobial assay of E. hirta, the properties of prime importance possessed by the plant can be made apparent which will help to prevent the over-exploitation of some of the endangered medicinal plants. The present study aims with study of phytochemical screening and antimicrobial activity extract of leaf, stem, whole stem of *Euphorbia hirta*. This study will create awareness among researchers about a basic idea as to what phytochemicals are present in *E. hirta* that can be used to produce medicines for curing various ailments.

Methodology :

Sampling: Fresh plants of E. hirta were collected from the area outside Patna Women's College. The leaves, stems and roots were thoroughly washed under tap water and dried under shade for 3-5 days. The dried plant parts were ground to fine powder and stored in containers for further use.

Preparation of extracts : Shade dried plant leaves, stems, roots were chopped into small pieces and then grinded to powdered form. 2 g of dried powder was kept in 3 separate round bottomed flasks and sample extraction was done for different solvents (distilled water, methanol and ethanol). Extraction was done with 20 ml of each solvent for 24 h. Crude extracts were then stored in refrigerator for further use.

Phytochemical analysis : Various phytochemical tests were conducted to check the presence of different phytochemicals such as alkaloids, saponins, quinones, amino acids and proteins, carbohydrates, coumarins, phenols, flavonoids and cardiac glycosides (Arsule and Sable, 2017).

Test for alkaloids (Wagner's test): 3-5 drops of Wagner's reagent was added to 1ml of plant extract. Appearance of reddish brown precipitate or coloration confirms the presence of alkaloid.

Test for carbohydrates: Both Fehling A and Fehling B solution were mixed in equal volume and this reagent was added to crude extract and boiled smoothly. Appearance of brick red precipitate at the bottom of the test tube indicates the presence of carbohydrates.

Test for Cardiac glycosides (Keller Kelliani's Test): 1ml of plant extract was treated with 1ml glacial acetic acid and 2-3 drops of 5% ferric chloride solution. To this mixture, 0.5 ml of conc. sulphuric acid was added. Appearance of a brown ring at the interface confirms the presence of deoxysugar characteristics of cardenolides.

Test for flavonoids (Alkaline reagent test): 1ml of plant extract was treated with 3-5 drops of 20% NaOH. Intense yellow color was observed and it became colorless on addition of 0.5ml dil HCl.

Test for phenols (Ferric chloride test): 5-6 drops of aqueous ferric chloride solution was added to 1ml extract. Appearance of a deep blue or black coloration confirms positive test.

Test for amino acids and proteins: 2-5 drops of aqueous ninhydrin solution was added to 1ml plant extract and the test tube was kept in a boiling water bath for 1-2 minutes. Appearance of purple colour confirms positive test for same.

Test for saponins (Foam test): 5 ml of distilled water was added to 1ml of extract and it was agitated vigorously. Formation of consistent foam for 10-15 minutes confirms the presence of saponin.

Test for quinones: 5ml of distilled water was added to 1ml of extract. Appearance of turbidity confirms the presence of quinones.

Test for coumarins: 1.5 ml of 10% NaOH was added to 1ml plant extract. Appearance of yellow colouration confirms the presence of coumarins.

Antimicrobial study : Antimicrobial activity can be defined as a collective term for all the active principles (agents) that inhibit the growth of microbes such as bacteria and fungi and prevent the formation of microbial colonies, and may destroy microorganisms. Three bacterial strains viz., *Staphylococcus aureus* and *Proteus vulgaris* were obtained from SRL Diagnostics, Fraser Road, Patna and *E. coli* was procured from CRL, Patna Women's College. All these bacterial strains were sub-cultured in NA media and maintained at 4°C for further experiment studies.

Preparation of bacterial culture : One loopful of each bacterial strain were inoculated in 50 ml of sterile nutrient broth and incubated for 24 h in shaker incubator.

Assay of anti-bacterial activity :

Assay of anti-bacterial activity of extract of leaf, stem and whole plant of Euphorbia hirta was done by Disc Diffusion method. In this method 20ml of sterilized Nutrient Agar was poured into sterile petri plates and after solidification 120µl of bacterial culture was poured on the plates and the culture was spread on plates using spreader. Then, the Whatmans filter paper discs (6mm in diameter) were kept over the agar plates using sterile forceps at various concentrations of plant extracts. Concentrated solvent was used as negative control. The anti-bacterial assay plates were kept in the incubator, where all the plates were incubated at 37ºC for 24h. The diameter of inhibition zone was noted down (Ahmad et al., 2017).

Results and Discussion:

Phytochemical analysis : Phytochemicals play a key role in the growth and development of plants. Upon phytochemical screening, the crude extract exhibited the presence of carbohydrates, alkaloids, phenols and cardiac glycosides in aqueous, methanol and ethanol extracts whereas quinones were absent in all the extracts (Arsule and Sable, 2017). Amino acids and proteins were present only in aqueous extract and the remaining showed negative result. Saponins were present in all the extracts. Flavonoids were present only in ethanol and methanol extracts and were absent in the aqueous extract. Coumarins were present in methanol and ethanol extracts and absent in the aqueous. Alkaloids are basically involved in the defence mechanisms of plant against herbivores and pathogens and plays a key role in plant metabolism and plant physiology. Carbohydrates in plants are a great source of energy. The phenolic compounds, flavinoids and saponins present in plants also help in the defence mechanism of the plant and amino acids help in protein synthesis.

Table 1. Result of phytochemical screening

S. No.	Phytoconstituents	Leaf			Stem			Whole plant		
		Aq	E	M	Aq	E	M	Aq	E	M
1.	Alkaloids	+	+	+	+	+	+	+	+	+
2.	Carbohydrates	+	+	+	+	+	+	+	+	+
3.	Phenols	+	+	+	+	+	+	+	+	+
4.	Cardiac glycosides	+	+	+	+	+	+	+	+	+
5.	Flavonoids	-	+	+	-	+	+	-	+	+
6.	Coumarins	-	+	+	-	+	+	-	+	+
7.	Saponins	+	+	+	+	+	+	+	+	+
8.	Quinones	-	-	-	-	-	-	-	-	-
9.	Amino acids and proteins	+	-	-	+	-	-	+	-	-

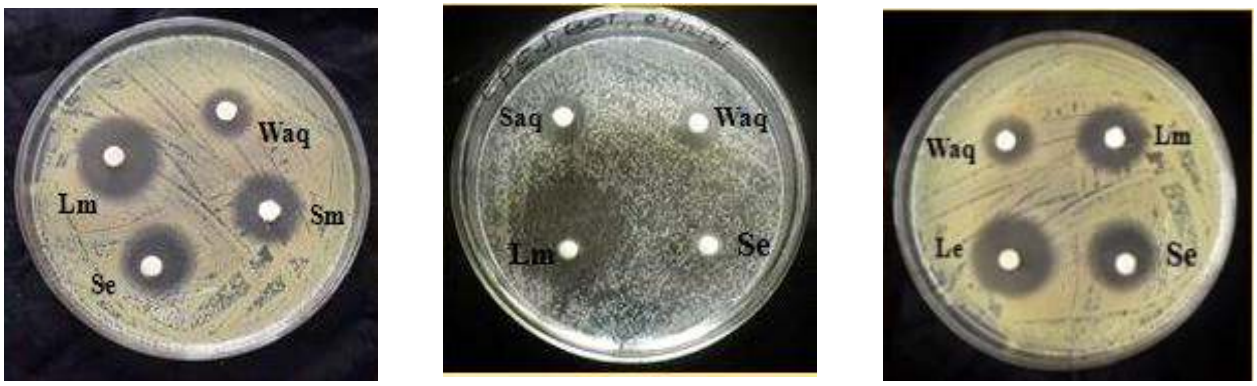
(Aq= aqueous, E= ethanol, M=methanol, + = positive result, - = negative result)

Anti- bacterial activity

Antibacterial activity is anything that destroys or kills bacteria or suppresses their growth or their ability to reproduce. Heat, chemicals such as chlorine and antibiotic drugs all have antibacterial properties. Result of the antibacterial activity of the isolated extract by using different solvents (ethanol, methanol and aqueous) is represented in table 2. The antibacterial activity of ethanol, methanol and aqueous extract of leaf, stem and whole plant of *Euphorbia hirta* were inspected against the selected experiment pathogens such as *Proteus vulgaris, Staphylococcus aureus* and *Escherichia coli* by disc diffusion method (Fig. 1). The various zones of inhibition shown by different extracts are represented in the table below. A significant inhibition of 15 mm was obtained in ethanol extracts and 13 mm from aqueous extract of E. hirta against S. aureus (Kumari and Pandey, 2017). In minimum inhibitory concentration (MIC), the methanolic extract exhibited maximum inhibition in comparison to ethanolic and aqueous extracts (Gupta and Gupta, 2019). Antibacterial properties of a plant are used in the treatment of bacterial infections.

Table 2. Result of anti-bacterial activity against various test organisms

Test organism	*Different types of extracts*	*Zone of inhibition (in mm)*		
		Aqueous extract	*Ethanolic extract*	*Methanolic extract*
Proteus vulgaris	Leaf extract	8	15	14
	Stem extract	6	15	11
	Whole plant extract	*7*	10	11
Staphylococcusaureus	Leaf extract	13	15	17
	Stem extract	10	12	15
	Whole plant extract	6	8	10
Escherichia coli	Leaf extract	12	13	14
	Stem extract	9	10	12
	Whole plant extract	6	7	9



(A) **(B)** **(C)**

(Lm = Methanolic extract of leaf, Se = Ethanolic extract of stem, Waq = Aqueous extract of whole plant, Sm = Methanolic extract of stem)

Fig. 1. Results of anti-bacterial activity against (A) *S. aureus* (B) *E. coli* and (C) *P. vulgaris*

Conclusion:

The results obtained in the present investigation reveals that E. hirta is a rich repository of various phytochemicals viz., alkaloids, carbohydrates, flavonoids, phenols, saponins, coumarins, cardiac glycosides, etc. The anti-microbial study revealed that methanolic extract of *E. hirta* showed highest anti-bacterial activity against S. aureus and *E. coli* while ethanolic extract against *P. vulgaris.* This reveals the importance of choice of solvent for proper extract preparation. Leaves of *E. hirta* exhibit the highest antimicrobial activity against all 3 test strains. The present work suggests promoting utilization of weeds of medicinal importance like *E. hirta* for the benefit of mankind so as to prevent the over-exploitation of endangered medicinal plants and which may reduce the antibiotic dependency.

Acknowledgement:

The authors are thankful to the Principal, Patna Women's College and Head, Department of Botany for providing laboratory facility for carrying out the research work. We are also grateful to UGC, Government of India for funding this research project.

References :

Ahmad W, Singh S and Kumar S. (2017). Phytochemical screening and antimicrobial study of *Euphorbia hirta* extracts. Journal of Medicinal Plants Studies; 5(2): 183-186.

Arsule CS and Sable KV. (2017). Preliminary phytochemical analysis of *Euphorbia hirta* Linn. leaves. International Journal of Life Sciences, Vol. 5 (4): 746-748.

Asha S, Deevika B, Mohamad Sadiq A. (2014). *Euphorbia hirta* Linn - a review on traditional uses, phytochemistry and pharmacology. World Journal of Pharmaceutical Research, Vol 3, Issue 4.

Bandow JE, Brotz H, Leichert LI, Labischinski H, Hecker M. (2003). Proteomic approach to understanding antibiotic action. Antimicrob Agents Chemother, 47(3):948- 55.

Bansod S, Rai M. (2008). Antifungal activity of essential oils from Indian medicinal plants against human pathogenic *Aspergillus fumigatus* and A. niger. World Journal of Medical Sciences, 3:81-88.

Gupta R and Gupta J. (2019). Investigation of antimicrobial activity of *Euphorbia hirta* leaves. International Journal of Life Science and Pharma Research, 9(3): (32- 37).

Kumari I, Pandey RK. (2017). Antibacterial Activity of *Euphorbia hirta* L., Applications of biotechnology for sustainable development, 1-6.

Rahmati H, Salehi S, Malekpour A, Farhangi F. (2015). Antimicrobial activity of castor oil plant *(Riccinus communis)* seeds extracts against gram positive bacteria, gram negative bacteria and yeast. International Journal of Molecular Medicine and Advance Science, 11:9-12.

Ramproshad S, Mondal B, Khan R, Afroz T, Ahmed S. (2012). Screening of phytochemical and pharmacological activities of leaves of medicinal plant *Plumeria rubra.* International Journal of Research in Pharmacy and Chemistry,2:1001.

Suresh SN, Rathshkumar S, Rajeshwari V, Sagadevan P, Gayathri S, Vithya D. (2012). Phytochemical analysis and antibacterial potential of *Cardiospermum halicacabum* Linn. (Sapindaceae). International Journal of Pharmacy and Life Science, 3:2209.

Westh H, Zinn CS, Rosdahl VT. (2004). An international multicenter study of antimicrobial consumption and resistance in *Staphylococcus aureus* isolates from 15 hospitals in 14 countries. Microbial Drug Resistance,10:169–76.