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Original Research Article

Physicochemical screening and antibacterial activity of fresh water macroalgae, *Cladophora glomerata* (L.) Kutz

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ABSTRACT

The plant kingdom is rich in bioactive natural compounds, and extracts from various plants, as well as red, green, and brown macro and micro algae, can be used. *Cladophora* is one of the largest filamentous green-alga genus. The majority of *Cladophora* species can be found all over the world, in both temperate and tropical climates. They are rich in secondary metabolites. The dried algal mass was crushed into a fine powder and acetone extract was used for phytochemical screening. Phytochemical analysis showed the presence of various secondary metabolites like alkaloids, steroids, flavonoids, carbohydrates, and proteins. Antibacterial activity of *C. glomerata* against *Staphylococcus aureus* and *E.coli* showed positive results.

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1. Introduction

C. glomerata is common macroalgae bloom that produces a lot of biomass in a short amount of time. Macroalgae are the most important primary producer in the aquatic ecosystem¹ Algae formed the foundation of the aquatic food chain, and they were vital in maintaining CO₂ in the carbon cycle via photosynthesis, as well as playing a significant part in biogeochemical cycles.²⁻⁴ *C. glomerata* consists of saturated and unsaturated fatty acids, sterols, terpenoids, and phenolic compounds also contain various bioactive compounds including pigments (carotenoids, chlorophylls, and tocopherols), sulphated polysaccharides (fucoidan), amino acids, and mono- and polyphenols. Algal phenolic compounds were reported to have antioxidant, anticancer, antibacterial, antiviral, and anti-inflammatory

activities.⁵⁻⁷ Humans have used land and aquatic plant extracts and essential oils as a natural cure against various illnesses for many decades in human history. It is widely used in food preservation and pharmaceuticals alternative medicine, antimicrobial, antifungal properties, and Antioxidant Properties.^{8,9}

The aim of the present study was to carry out the phytochemical screening and antibacterial activities of *C. glomerata* extract.

2. Materials and Methods

2.1. Plant collection

Samplings were carried out from Sawarde, Chiplun, Maharashtra during autumn in 2021. Samples of *C. glomerata* were collected manually from the riverside and rock. Then the sample was thoroughly rinsed with fresh distilled water to remove other materials. The algae biomass

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was sundried. Then the dried algae were powdered and weighed and store in a clean container.

2.2. Extract preparation

8gm of powdered material mixed with 150ml acetone and Soxhlet extraction process was carried out at temp 51⁰C for 4-5hrs.



Fig. 1: Extraction of Algae.

2.3. Phytochemical screening

Using general and particular chemical reagents, and subjected to several chemical tests to detect the presence of various phytoconstituents.^{10,11}

3. Evaluation of Antibacterial Activity

3.1. Micro biological assay

A Petri dish is used to culture microorganisms (agar plates). Then petri dish were sterilized. This helps prevent the contamination of the new culture. Agar well diffusion method was used to determine the antimicrobial activity of plant extract in vitro. Agar was used to culture different micro-organisms examined in this study. Against the wall of the tube above the liquid to remove excess inoculum. The entire surface of agar plate wash then swabs bed 3 times with the cotton swab, transferring the inoculum, while the plates were rotated by approximately 60°between streak stone sure even distribution. The overall procedure of inoculums

preparation and inoculation of culture media remained the same for all bacteria. Each bacterium was inoculated on 2 agar plates.¹²

4. Preparation of Agar Plates

Before starting, ensure that the Petri dish (dishes) is closed with its lid on until to pour the agar into them.

4.1. Sterilization of equipment's and the chemicals

Nutrient agar medium and normal saline solution were sterilized in an autoclave at 15 lbs pressure (121°C) for 150 mins. Petri plates Whatman filter paper, cotton swab were sterilized in the oven at 160°C for 2 hrs.¹³

4.2. Preparation of nutrient agar medium slant

Nutrient agar powder 5gm was dissolved in 200ml distilled water, boiled and then poured in the test tubes then plugged with cotton and sterilized in autoclave at 15lbs for 15 min. After sterilization the tubes containing the nutrient medium were kept in an inclined position for 30 min. Then on the surface of theslant pure culture of *Staphylococcus aureus* and *E.Coli* were streaked in aseptic condition and incubated at 37 ° c for 24 hrs.

Table 1: Phytochemical screening of acetone extract of *C. glomerata*.

Chemical constituents	Test	Acetone extract
Alkaloids	Dragendroff's test	+
	Wagner's test	+
	Hager's test	+
	Mayer's test	+
	Molisch's test	+
Carbohydrates	Barfoed's test	+
	Benedict's test	+
Flavonoids	Lead acetate test	+
	Salkowaski test	+
Steroids	Liebermann Burchard test	+
	5% Ferric chloride test	–
	10% lead acetate	–
Tannins	Acetic acid	–
	Pot. Permagnate	–
	Dil. Iodine	–
Proteins	Millon's test	+
	Xanthoproteic test	+

4.3. Preparation of suspension of test Bacteria

Using the 24 hrs old growth of test bacteria from the slant, suspension of the bacteria was made separately in sterile normal saline solution (0.85%Nacl in distilled water) in aseptic condition to get moderate turbidity.

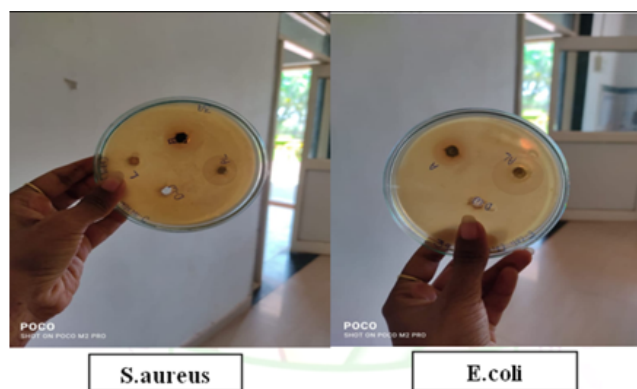


Fig. 2: Antibacterial activity of *C. glomerata* extract

Table 2: Antibacterial activity of extracts of *C. glomerata*

Sample	Test organism	Zone of inhibition(mm)
Acetone extract	E. coli	14.04
	S. aureus	20.7
Aqueous extract	E. coli	–
	S. aureus	–

5. Result and Discussion

Acetone extract were screened qualitatively for detection of phytoconstituents using general and specific chemical reagents as per the following.

6. Conclusion

In the present study, we have found that most of the biologically active phytochemicals were present in the standardized extract of *C. glomerata*. The antibacterial activities of *C. glomerata* may be due to the presence of phytochemicals.

7. Source of Funding

None.

8. Conflict of Interest

None.

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