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## **Original Article : Open Access**

# Molecular identification and therapeutic potential of green seaweed *Chaetomorpha antennina* (Bory.) Kützing. from coastal area

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Article Info	Abstract
Article history Received 2 October 2024 Revised 16 November 2024 Accepted 17 November 2024 Published Online 30 December 2024 Keywords Antibacterial Antioxidant <i>Chaetomorpha antennina</i> (Bory.) Kützing. Phytocompounds Seaweed Sequencing	Seaweeds have created a wide impact the pharmaceutical and food industries with its various bioactive substances. <i>Chaetomorpha antennina</i> (Bory.) Kützing, one of the fast growing marine algae which is categorized under green algae, was up to 10 cm tall, with cylindrical or barrel-shaped, unbranched filaments, clinging to rocks and calcareous stones. The primary focus of this research explores the molecular taxonomy, as well as the anti-inflammatory, antibacterial, and antioxidant properties of <i>C. antennina</i> . Seaweed extraction was performed using the ethyl acetate, the phytochemical and biochemical compositions were analyzed along with its total phenolic content. The antibacterial and antioxidant potentials were evaluated by ferric ion reduction power, DPPH radical scavenging and hydrogen peroxide scavenging assays. Moreover, the anti-inflammatory activities were evaluated by denaturing albumin and detection of hemolysis by inducing heat. The biochemical composition such as carbohydrate (39.05%), protein (18.47%), lipid (6.62%) as well as the total phenol content (6.9 µg/ml), DPPH (54.5 µg/ml), FRAP (59.08 µg/ml) and H <sub>2</sub> O <sub>2</sub> (67.47 µg/ml). Albumin denaturation exhibits 74.36 µg/ml, whereas heat induced hemolysis was reported as 54.89 µg/ml and the significant antimicrobial activity was observed. The taxonomic range and molecular characterization of <i>C. antennina</i> using primer and 18S rRNA primer was identified. The study concludes that the crude ethyl acetate extract of <i>C. antennina</i> possesses strong anti-inflammatory antioxidant properties. The bioactive chemicals found in this green seaweed extracts have the potential to serve as a valuable source for pharmaceutical applications.

## 1. Introduction

The marine environment's primary source of biomass is marine algae. Seaweeds are a flexible and adaptable biomass that is used extensively in many different industries, including pharmaceuticals, skincare, renewable energy, animal husbandry, human nutrition, and dietary supplements. In particularly, seaweed extracts are essential for promoting sustainable farming methods and increasing plant resilience (Mahalakshmi *et al.*, 2024). For thousands of years, traditional medicines have been utilized to address everyday health concerns and maintain overall well-being (Mohd. Kashif Husain, 2021), As most "super foods" have benefits, along with their palliative or therapeutic values, the idea has recently gained popularity among people worldwide (Divya Pingili *et al.*, 2023).

Seaweeds are receiving the highest priority in research, as they are extensively explored for their significant potentials in synthesizing bioactive compounds such as proteins, dietary fiber, lipids, essential amino acids, vitamins (A, C, D,  $B_1$ ,  $B_{12}$  and E), minerals, polysaccharides and secondary metabolites (Plaza *et al.*, 2008).

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Copyright © 2024Ukaaz Publications. All rights reserved. Email: ukaaz@yahoo.com; Website: www.ukaazpublications.com Natural resources such as soils, plants, and marine life have gained a lot of interest for centuries because of their various pharmacological characteristics and the presence of wide variety of novel biomolecules (Malarvizhi and Muhammad Musthafa, 2024). Macroalgae produce a diverse range of bioactive compounds including polysaccharides, phlorotannins, sterols, lipids, alkaloids, glycerols, diterphenoids, quinines, polyketides, and cyclic peptides that exhibits a variety of biological functions (Al-Saif *et al.*, 2014).

The phytochemical analysis of seaweed algal species will be a good preliminary approach to reveal its secondary metabolite constituents and ensure their medicinal values that possess numerous pharmacological and biochemical properties (Lee et al., 2013). The pharmaceutical industry has shown a great deal of interest in a number of various compounds derived from algae. Numerous seaweed species in the coastal region of south India remain undiscovered, despite the fact that antioxidants have demonstrated their value in the prevention of free radical-related diseases in recent decades (Hybertson et al., 2011). The structural components of the species were protected from environmental oxidative damage by antioxidants present in macroalgae (De Alencar et al., 2016). Recent research on multidrug resistance among pathogenic microorganisms are primarily driven by the overuse and improper application of commercial antimicrobial drugs, the scientists worldwide are striving to develop alternative solutions that are more environment friendly, have fewer side effects, and offer long-lasting effectiveness (Tankeswar Nath et al., 2024).

# 2. Materials and Methods

## 2.1 Collection of samples

Algal seaweed samples were collected that include *C. antennina* along in Thiruvottiyur (Latitude 13° 9' 28 N and Longitude 80° 18' 15 E), Chennai, Tamil Nadu, India. The collected algal samples were authenticated by R.K.Algal Centre, Mandapam, Ramanathapuram District, Tamil Nadu, supported by CSIR, Ramanathapuram District Ref. No. -Algae/Athen/2020/115). To get rid of any adhered sediments and contaminants, live, healthy samples were gathered and cleaned at the sampling location with seawater and also with tap water. The cleaned seaweed was shade-dried and algal material were grained manually and stored for further use.

# 2.2 Algal extraction

The dried fine powder of C. antennina (100 g) was taken for soxhlet extraction using ethyl acetate and the extraction was repeated and after the computation, the collected extracts can be used for subsequent analysis, with the positive results being underlined for its potential applications.

## 2.3 Pharmacognostic study

A pharmacognostic study was done by organoleptic evaluation. In this method the algal sample was examined under a magnifying glass and the morphological characteristics of the various algal parts were taken for the further identification using the standard methods (Khandelwal, 2008).

#### 2.4 Qualitative phytochemical analysis of algal extracts

The phytochemical analysis *C. antennina* ethyl acetate extract was done by using the standard method (Baxter *et al.*, 1998). The naturally available secondary metabolites such as alkaloids, terpenoids, triterphenoids, steroids, tannins, phenols, flavonoids, coumarins, quinines, and glycoside compounds were tested for the presence or absence in the above mentioned algal extract.

## 2.5 Biochemical analysis

## 2.5.1 Carbohydrate analysis

The presence of carbohydrates was estimated using dubois method (Dubois, 1956) in which 20 mg of the seaweed sample was mixed with 1 ml of 4% phenol solution and 5 ml of concentrated sulfuric acid and mixture was then stored for 30 min in a dark room. The color intensity developed was measured at a wavelength of 490 nm using the spectrophotometer. The amount of carbohydrates was determined using the D-glucose, as standard, and the results were reported in milligrams per gram of sugar.

#### 2.5.2 Protein analysis

The protein content present in the ethyl acetate extract was estimated with the biuret method (Raymond *et al.*, 1964) in which 5 mg of the sample was mixed in 1 ml of distilled water along with 4 ml of biuret reagent. It was incubated at room temperature for 30 min and then centrifuged at 4000 rpm for 10 min, after centrifugation the supernatant was calculated using a spectrophotometer set at 540 nm and bovine serum albumin (BSA) served as a standard. The result was expressed in g/g protein.

## 2.5.3 Lipid analysis

Using a methanol-chloroform mixture, the lipid content was calculated in accordance with Folch *et al.* (1957) description. The sample (400 mg) was mixed with 5 ml of methanol-chloroform (1:2), and it was incubated for 24 h at room temperature, after incubation the mixture was filtered using a filter paper. The filtrate was collected in 10 ml beaker (previously weighed) and was placed on a hot plate for evaporation till residue was left at bottom of the beaker. The weight of reside was calculated by removing the weight of the beaker.

#### 2.6 Assessment of total phenol content

Initially, algal extract (100  $\mu$ l) and Folin-Ciocalteu's reagent (500  $\mu$ l) were mixed in a 1:1 ratio with distilled water and was kept at room temperature in the dark for 5 min. To the mixture 1 ml of 20% Na<sub>2</sub>CO<sub>3</sub> (sodium carbonate) was added and incubated at room temperature for 40 min again in the dark condition. The phenol content present in the algal samples were measured using spectrophotometer, at 725 nm and the result was expressed in gallic acid equivalent per gram (GAE/g) (Siddhuraju and Becker, 2003).

## 2.7 Antibacterial activities

The antibacterial assay was performed using well diffusion method with six distinct bacterial pathogens such as *Klebsiella pneumoniae*, *Escherichia coli*, *Enterococcus faecalis*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus* (Zain *et al.*, 2012). Bacterial inoculums were seeded onto mueller hinton medium, wells with six mm in diameter were bored on agar, and various concentrations of algal extracts (0.125 g, 0.25 g, 0.5 g, and 1.0 g) were added to separate wells and were incubated for 24-48 h at 37°C, the zone of inhibition was measured and recorded in mm.

## 2.8 Determination of MICs (minimal inhibitory concentrations)

To calculate the MICs of *C. antennina* ethyl acetate extract (Wayne, 2010), it was serially diluted using in mueller hinton broth in sterile 96-well plates. The bacterial cells at the concentrations of 50  $\mu$ l of 10<sup>5</sup> CFU ml<sup>-1</sup> were incubated into each well and were incubated overnight at 37°C. After incubation OD value was taken to measure the growth of the test organism and among them the lowest value indicates the minimum growth of the tested organism and was recorded as minimum inhibitory concentrations of the ethyl acetate extract.

#### 2.9 Determination antioxidant activity

## 2.9.1 Total antioxidant activities

The total antioxidant activity of algal extract was investigated as described by Prieto *et al.* (1999), the reagent consists of 0.9942 g of  $Na_2SO_4$  (28 mM), 1.2359 g of ammonium molybdate (4 mM solution) and 7.45 ml of  $H_2SO_4$  (0.6 mM) in 250 ml of distilled water. For dissolving 300 µl of the seaweed extract and 3 ml of the above said total antioxidant reagent was used. The antioxidant activity of the seaweed extract was measured using spectrophotometer at 695 nm and the distilled water was used in the test as blank instead total antioxidant reagent. The quantity of ascorbic acid equivalents is used to represent total antioxidant activity in the present study.

## 2.9.2 DPPH radical scavenging assay

For the DPPH assay, the protocol outlined by Burits and Bucar, (2000) was adopted, where a mixture of methanolic DPPH (2, 2diphenyl-1-picrylhydrazyl) reagent (1 ml, 0.002%, w/v) and crude sample 1 ml was kept in the dark at room temperature for half an hour. Absorbance was read spectrophotometrically at 517 nm, using ascorbic acid as the control. The inhibition percentage was found out by: Absorbance of control–Absorbance of sample/Absorbance of control × 100.

#### 2.9.3 Assessment of ferric ion reducing antioxidant power (FRAP)

The FRAP of the extract was found by the following procedure Benzie and Strain, (1996) protocol. 10 mM 2, 4, 6-tris (2-pyridyl)s-triazine (TPTZ) solution, 20 mM ferric chloride solution and 30 mM sodium acetate buffer, in a 10:1:1 ratio makes up the FRAP solution. After combining 900  $\mu$ l of FRAP solution with 30  $\mu$ l of algal extract was allowed to remain in the dark at room temperature for half an hour and the results were calculated using a spectrophotometer at 593 nm and were recorded.

#### 2.9.4 Hydrogen peroxide scavenging activities

To find out hydrogen peroxide scavenging activity of the sample, the standard protocols of Govindarajan *et al.* (2003) were employed. 2 ml of 10 mM hydrogen peroxide solution was prepared in 0.1 M buffered with phosphate (pH 7.4) and were mixed with 1 ml of the algal extract (0.25 mg) and the whole set up was kept at  $37^{\circ}$ C. After 10 min the absorbance of samples was measured using a spectrophotometer at 230 nm and were quantified with the equation: Absorbance of control–Absorbance of sample/Absorbance of control × 100 to determine the inhibitory percentage.

#### 2.10 Anti-inflammatory activities

#### 2.10.1 Albumin denaturation assay

The sample concentrations varied from 25 to  $1000 \ \mu g/ml$  were mixed with 1% aqueous albumin solution and the pH was adjusted using 1N hydrochloric acid. After 20 min of incubation at room temperature, it was heated at 51°C for 20 min and then allowed to cool, the results were read spectrophotometrically at 660 nm and the result was calculated as Inhibition (%) = Absorbance of control–Absorbance of sample/Absorbance of sample × 100 (Mizushima and Kobayashi, 1968).

#### 2.10.2 Heat induced hemolysis assay

One ml of 10% RBC suspension and 1 ml of crude extract in different concentrations make up the reaction mixture's composition. Then, in a water bath set up with the temperature of 56°C, reaction mixtures were incubated for half an hour, and then the tubes were cooled in running water. Then the test tubes were subjected to centrifugation for 5 min at 3000 rpm, the supernatant was collected and the OD value was read using spectrophotometrically at 560 nm and the result was calculated by Inhibition (%) = Absorbance of control–Absorbance of sample/Absorbance of sample × 100 (Sakat *et al.*, 2010).

# 2.11 Molecular identification of C. antennina

#### 2.11.1 DNA extraction

The DNA was extracted from the sample by centrifuging it for 10 min at 10,000 rpm and pooled 4-5 times to obtain biomass, the pellet was homogenized in a mortar and pestle whilst adding 2 ml of extraction buffer and 40  $\mu$ l of 2%  $\beta$ -mercaptoethanol for lysis of cell wall/membrane. In a water bath set at 60-65°C, the homogenized mixture was taken as duplicates and incubated for digestion 1 to 1.5 h with gentle inversion of the mixture every 15 min. After adding equivocates of chloroform: isoamyl alcohol (24:1 ml) to the digested sample, for room temperature centrifugation at 10,000 rpm for 10 min. The obtained supernatant was then mix with 10 µl of 3 M sodium acetate and 1 ml of isopropanol. It was incubated at -20°C for overnight and then the supernatant was disposed and subjected for centrifugation at 4°C at 10,000 rpm. After dissolving the pellets in 50 µl of MilliQ water, 1 ml of absolute alcohol ethanol was used to dissolve the pellets and was incubated at -20°C. At 4°C and again centrifuged at 10,000 rpm for 10 min, and the pellet was dissolved in 100 µl MilliQ water and precipitated using 70% ethanol. The supernatant was disposed and the reaction mixture was again centrifuged for 10 min at 4°C with 10,000 rpm. After being air-dried, the pellet was kept in 50 µl of TE buffer (Doyle, 1991).

# 2.11.2 PCR amplification and sequencing reactions

The veriti 96 well thermocycler of Applied Biosystems was used for the polymerization chain reaction and the program for 18S rRNA amplification was entered and saved. The PCR cycles were as follows: initial denaturation temperature was 95°C for 5 min, the temperature was set at 95°C for 30 sec of denaturation, 61°C for 30 sec of annealing, and 30 cycles were complete at 72°C for 1 min and 30 sec and the final extension temperature was set to 72°C for 10 min. Applied biosystems were used to carry out the sequencing reactions, for single-pass sequencing on every template; 18S rRNA universal primers were utilized for DNA sequencing. The forward primer 5'-ACCTGGTTGATCCTGCCAGT-3' and reverse primer 5'-TCAGCCTTGCGACCATAC-3'. The fluorescently labeled fragments were separated from the unincorporated terminators using an ethanol precipitation process (Applied biosystems).

## 3. Results

#### 3.1 Phytochemical analysis

The green seaweed, *C. antennina* survives in a marine habitat with a fishy odor, smooth texture which is filamentous shape measuring 7 cm. The phytochemical screening was performed to investigate the major secondary metabolites present in the extracts. In the present study, the *C. antennina* in the ethyl acetate crude extract such as the carbohydrates, flavonoids, tannins, alkaloids, terpenoids, triterpenoids, phenols, steroids, phlobatannins, quinines, glycosides and cardiac glycosides.

#### 3.2 Biochemical composition

The ethyl acetate of crude extract had important secondary metabolites, showed in Figure 1. The biochemical results showed the maximum carbohydrate content was observed in *C. antennina* (carbohydrate content-39.05%, protein-18.47% and lipid-6.62%).

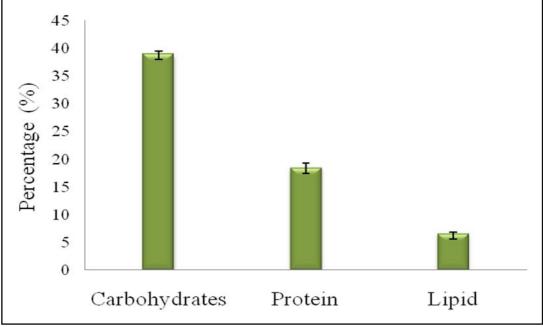


Figure 1: Biochemical composition of C. antennina.

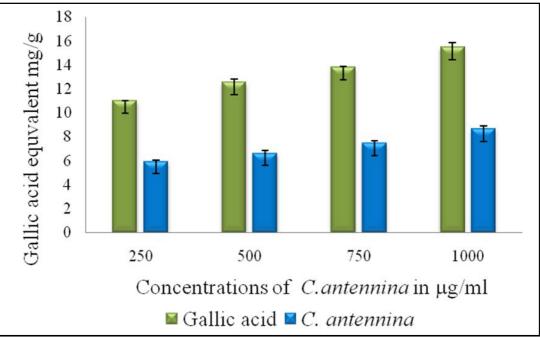


Figure 2: Total phenolic content of C. antennina.

## 3.3 Total phenolic content

Figure 2. showed the total phenolic content of tested seaweed crude extract. The GAE (Gallic acid equivalents) were used to express the results for *C. antennina* (8.61  $\mu$ g GAE/g) in 1000  $\mu$ g/ml a concentration.

## 3.4 Antibacterial activity

At all investigated concentrations, the ethyl acetate crude extract of *C. antennina* displayed antibacterial potential against gram negative

and positive bacteria using well diffusion and minimum inhibitory concentration (MIC) techniques are shown in Table 1. By measuring the inhibitory zone and MIC values on each tested bacterial strain, *C. antennina* extract was evaluated. The highest inhibitory zone was observed for *Pseudomonas aeruginosa* at 1 mg/ml concentrations which was 21 mm, while, *Escherichia coli* was 19 mm. *Acinetobacter baumannii* (10 mm) and *Staphylococcus aureus* (18 mm) were observed the minimum zone of inhibition. At levels between the concentrations of 0.125 and 0.25 mg/ml, *Enterococcus faecalis* and

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*Klebsiella pneumonia* showed no inhibitory effect. The results demonstrated that the bacteria with the highest MIC of *C. antennina* crude extract were *Acinetobacter baumannii* (05.73  $\pm$  03.11 mg/ml), *Pseudomonas aeruginosa* (6.13  $\pm$  00.02 mg/ml), *Staphylococcus* 

*aureus* (07.12  $\pm$  02.00 mg/ml), *Escherichia coli* (08.43  $\pm$  01.00 mg/ml), *Enterococcus faecalis* (90.00  $\pm$  00.00 mg/ml) and *Klebsiella pneumonia* (40.00  $\pm$  00.00 mg/ml) proved to be the most sensitive strain of bacteria among those tested.

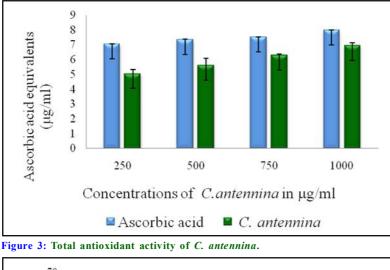
Table 1: The antibacterial properties and minimal inhibitory concentration of C. antennina

Bacterial isolates	Zone of inhibition concentration (mm)				Minimum inhibitory
	0.125 mg/ml	0.25 mg/ml	0.5 mg/ml	1 mg/ml	concentration (mg/ml)
Staphylococcus aureus	6	9	12	18	$07.12 \pm 02.00$
Enterococcus faecalis	-	-	-	09	$90.00 \pm 00.00$
Acinetobacter baumannii	-	9	13	18	$05.73 \pm 03.11$
Escherichia coli	9	12	16	19	$08.43 \pm 01.00$
Pseudomonas aeruginosa	10	14	19	21	$06.13 \pm 00.02$
Klebsiella pneumonia	-	-	-	10	$40.00 \pm 00.00$

## 3.5 Determination of antioxidant activity

The ethyl acetate of crude extract from *C. antennina* demonstrated the highest antioxidant activity, measuring  $6.9 \pm 0.19$  mg AAE/g, as shown in Figure 3. This seaweed extract exhibited significant DPPH

radical scavenging activity at 54.5%. Additionally, *C. antennina* recorded higher ferric reducing antioxidant power values at 59.08% and hydrogen peroxide radical scavenging activity at 67.47% at 1000  $\mu$ g/ml a concentration as shown in Figure 4. These findings highlight *C. antennina* as a potent source of natural antioxidants.



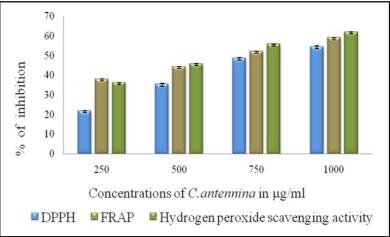
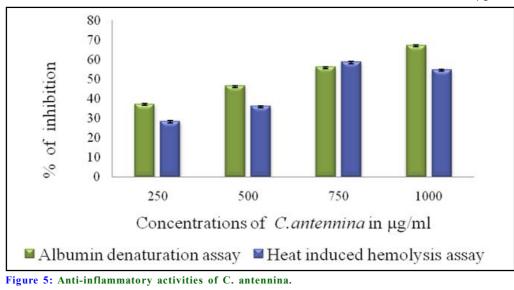


Figure 4: In vitro antioxidant analysis of C. antennina.

# 3.6 Anti-inflammatory assays

Anti-inflammatory assay for seaweed extract was observed as concentration-dependent are shown in Figure 5. The findings revealed that the greatest inhibition of protein denaturation occurred at a concentration of 1000  $\mu$ g/ml of *C. antennina* (74.36%). Heat-induced hemolysis of human erythrocyte membrane stabilization at different concentrations was used to test the ethyl acetate extract. The highest percentage of inhibition observed in *C. antennina* was 54.89% with the crude extract concentrations were 1000  $\mu$ g/ml.



#### 3.7 DNA sequencing

The DNA sequencing was presented in the DNA with a high optical density was observed and for further PCR analysis. The 1Kb ladder to lane one ladder was moved. Lane 1 is mentioned as a standard. Lane 2 approximately 1100 bps of 18S rRNA of *C. antennina*, and

lane three were showed 700 bps of rcbL gene. Phylogenetic tree sequences were elucidated with the obtained data (Figure 6). Apart from the rbcL gene, 18S rRNA gene sequencing were well corresponded with the rcbL BLAST analysis. Using rbcL and 18S rRNA as marker genes, the green algae *C. antennina* was molecularly identified.

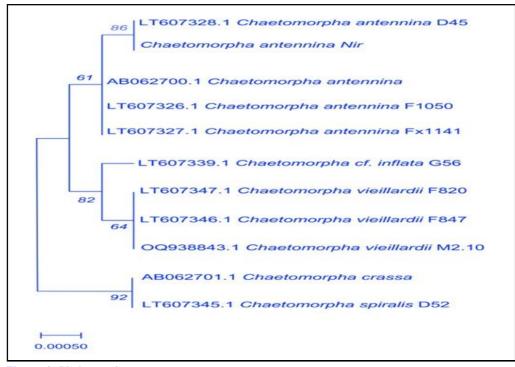


Figure 6: Phylogenetic tree.

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# 4. Discussion

Seaweeds are a rich source of biologically active substances, which contain compounds with a relatively high antioxidant activity. These studies revealed the presence of carbohydrates, protein, and lipids of higher concentration in these macro algae, and hence they are said to be the richest source for these bioactive principles. A similar concentration was recorded from green algae like in *C. antennina* extracts 34% of carbohydrates, 0.3% of lipids and 13% of protein were reported (Saranya *et al.*, 2014). Premalatha *et al.* (2011) reported that the ethanolic extracts of the green algae *C. antennina* contained several phytochemicals, including steroids, alkaloids, flavonoids, tannins, phenols, terpenoids, and glycosides and the author also reported that *C. antennina* and in DPPH radical scavenging activity of *C. antennina* exhibited 63.77%.

Phenolic compounds have been reported for their biological activities, including antioxidant properties which are widely distributed in the kingdom of plants. The methanolic extract of C. antennina revealed a phenol content of  $9.39 \pm 0.04$  mg GAE/g (Jimenez-Escrig and Sánchez-Muniz, 2000). A similar study result was shown by Saranya et al. (2014), where C. antennina had (0.28 mg GAEg<sup>-1</sup>) phenolic content. The antioxidant activity in naturally occurring compounds has been explored for recent decades, a variety of seaweed species have been identified as a source of oxygen species that are reactive inhibitors that can be added to food and protect against tissue oxidation. As per Nagai and Yukimoto (2003), the antioxidant activities of C. antennina were  $0.64 \pm 0.03$  mg ascorbic acid equivalent/g, with phlorotannins and fucoxanthin being the main active compounds. Similarly, C. antenna was found to have 9.32% hydrogen peroxide radical scavenging activity, respectively (Saranya et al., 2014). These findings are significant because radical scavengers can help protect cellular tissues from damage caused by free radicals, possibly averting illnesses like cancer.

Haq *et al.* (2019) found that the ethanol extract of *C. antennina* has a total antioxidant content of 189.14  $\pm$  0.99 mg QE/g and has 21.92  $\pm$  0.43 mg GAE/g, 21.81  $\pm$  0.04 mg GAE/g of phenolic and tannin content, respectively. Thanigaivel *et al.* (2014) reported that the ethanol extract of *C. antennina* showed greater antioxidant activity, indicated by a lower IC<sub>50</sub> value of 9.41  $\pm$  0.54 mg/ml, in comparison to the aqueous extract, which had an IC<sub>50</sub> of 15.44  $\pm$  0.98 mg/ml. According to Prasanthi *et al.* (2024), *C. antennina* had a smaller inhibition zone, ranging from 0.2 to 0.3 nm, against the same bacterial strains, indicating comparatively lower antibacterial potential. Raju *et al.* (2015) observed significant anti-inflammatory effects from methanol extracts *of C. antennina* in a carrageenan-induced paw oedema model. These effects were compared with the standard antiinflammatory treatment (diclofenac) which resulted in a reduction of 72.41  $\pm$  1.49 mg/ml at a dose of 50 mg/kg.

Sivakumar *et al.* (2013), collected and isolated *C. antennina* from the Kovalam seashore near Chennai, and analyzed the antibacterial activity of petroleum ether extract using the agar well diffusion method and even at low concentrations (50 µg/ml), the results demonstrated the significant inhibition zones for *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* was the most susceptible, exhibiting inhibition zones ranging from  $7.3 \pm 0.8$  mm to  $18 \pm 2.4$  mm at concentrations ranging from 50 to 500  $\mu$ g/ml, these results emphasized the need for sampling at Chennai coastal area for the collection of algal samples .

Chanthini *et al.* (2021), reported crude methanol extract of *C. antennina* was highly toxic to *Spodoptera litura* larvae, suggesting that it could be used as a natural larvicidal agent. 11 bioactive compounds were found by GC-MS analysis of methanol extract, with methyl esters of derivatives of chloroacetic acid, including dichloroacetaldehyde, methyl chloroacetaldehyde and methylene chloride, being particularly abundant. Furthermore, a chlorinated compound such as cis-2-chlorovinylacetate, trichloromethane, and chloromethane sulfonyl chloride, supports the fraction's bioactivity properties. This research could be taken as a reference; the *C. antennina* isolated in this study exhibited higher antibacterial, anti-inflammatory and antioxidant activity. The research further will focus on the separation and characterization of responsible for bioactive compound for therapeutic use.

Green algae's 18S rRNA gene has been extensively studied by amplification and sequencing with universal eukaryotic primers. Complete 18S rRNA gene sequences and partial rbcL gene sequences from genus were investigated for their molecular characteristics. Sequence comparisons were carried out using related *C. antennina* taxa that were redistributed in Genbank (Mahendran *et al.*, 2017). The phylogenetic tree was constructed using 18S rRNA sequence of green algae *Ulva pertusa* and *Ulva prolifera*. The study's barcoded *C. antennina* was well-known for its meticulous growth patterns caused by its quick uptake of nutrients (Imchen and Ezaz, 2020). In the near future, it may be possible to identify the ideal species for the specified ecosystems by comparing DNA barcodes with surrounding seawater conditions.

## 5. Conclusion

The *C. antennina* molecular identification of 18S rRNA data was useful in understanding the taxonomic and genetic level relationships of seaweeds. The conclusion of current research may provide a logical foundation for using marine algal extracts in the potential treatment of illnesses related to oxidative stress. They also provide more evidence that the antioxidant-rich extracts can be taken as a dietary supplement to support overall health.

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## **Conflict of interest**

The author declares no conflicts of interest relevant to this article.

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