ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Isolation of Phenolic Compounds from Marine Algal Extracts

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ABSTRACT

Marine algae are considered as a source of bioactive compounds as they are able to produce a great variety of secondary metabolites characterized by a broad spectrum of biological activities. In the present study using three marine algae namely, Gracilaria cortica, Enteromorpha flexuosa and Enteromorpha clathrata, the compound imparting antibacterial properties was identified as a phenolic compound.

Keywords: phenolic compounds, marine algae, antibacterial activity, solvent extraction

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ISSN: 0975-8585

INTRODUCTION

Most of the bioactive substances isolated from marine algae are chemically classified as brominated, aromatics, sterols, proteins, peptides and sulphated polysaccharides. [1-3]. Phenolic compounds have an activating or inhibiting effect on microbial growth according to their constitution and concentration [4]. The extracts of marine algae were reported to exhibit antibacterial activity. The antimicrobial activity of seaweeds on bacteria and fungi were reported by various researches [5, 6]. There were variations between the activity of brown, green and red algae [7,8].

Trace elements such as iron, zinc, manganese, copper, cadmium, cobalt, nickel and lead were examined using SEM/EDX analysis. The red algae Gracilaria sp contained high concentration of iron than Ulva sp and was very low in Sargassum sp [9]. Seasonal variation in protein content and metal content was observed [10, 11].

Phenolic compounds can act as antioxidants by chelating metal ions, preventing radical formation and improving the anti oxidant endogenous system [12]. The environment in which seaweeds grow is harsh as they are exposed to a combination of light and high oxygen concentrations. These factors can lead to the formation of free radicals and other strong oxidizing agents but seaweeds seldom suffer any serious photodynamic damage during metabolism. This fact implies that seaweed cells have some protective mechanisms and compounds [13].

The presence of phenolic compounds was confirmed by thin layer chromatography on a silica gel plate [14]. The appearance of blue spot in TLC chromatogram indicated the presence of phenolic compounds [15]. In the present study an attempt was made to isolate the active principle from Enteromorpha flexuosa and Gracilaria cortica which imparts the antibacterial activity using thin layer chromatography.

MATERIALS AND METHODS

Collection and preparation of samples

The marine algal samples namely Enteromorpha flexuosa and Gracilaria cortica were collected from Covelong beach, Chennai, Tamil Nadu. They were identified at the Dr. Krishnamurthy Institute of Algology. The algae after drying were weighed and finely powdered using a mixer grinder. The solvent extraction was done using ethanol in a Soxhlet apparatus.

Identification of phenolic compound by TLC

The phenolic compounds present in extracts were detected by TLC. Thin layer chromatography was performed on silica gel plates. An aliquot of each sample was spotted on the silica gel plates. The solvent system adapted was chloroform: methanol (10:1: v/v). The

spots were visualized using a UV detector after spraying with potassium ferric cyanide (1%) and ferric chloride (1%) in water.

Test organism

The test organisms used were Klebsiella sp, and Escherichia coli.

Evaluation of antibacterial activity

The antibacterial activity imparted by the phenolic compound was measured using the well diffusion method [16]. An aliquot containing 150µl of the algal extract was used to study the antibacterial activity. The plates were incubated at 37ºC for 24 hr. The antibacterial activity was assessed by measuring the diameter of the zone of inhibition.

RESULTS AND DISCUSSION

The algal extract was separated using TLC to identify the active principle present in the two seaweeds used in the study. The appearance of blue spot in TLC chromatogram confirmed the presence of phenolic compounds (Figure 1). Gracilaria cortica and Enteromorpha flexuosa extracts were active against both the pathogens namely, Klebsiella pneumonia and Escherichia coli. Maximum activity was reported in Enteromorpha flexuosa against Escherichia coli (3.7cm) and minimum activity was reported in Gracilaria cortica (1.5 cm) against Klebsiella sp. as shown in Table 1 and Figure 2. These results indicate that the extracts contained different antibacterial substances and reflect the variety of secondary metabolites. In recent years, there are numerous reports of macroalgae derived compound that have a broad range of biological activities such as antibacterial, antifungal, antiviral, antineoplastic, antifouling and anti inflammatory and antimitotic activites [17, 18, 19]. As a consequence of an increasing demand in screening for new therapeutic drugs from natural products, there is a greater interest towards marine organisms. Several marine organisms produce bioactive metabolites in response to ecological pressures [20].

Table 1: Antibacterial activity of the phenolic compounds isolated from algal extracts against pathogenic organisms

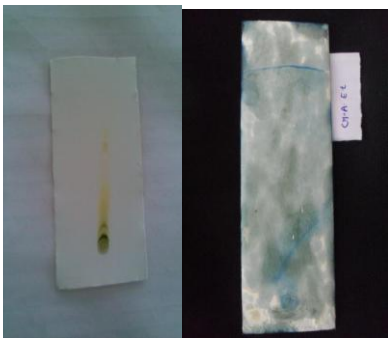
S.No	Species name	Test Organism	Zone of inhibition (mm)
1	*Gracilaria cortica*	*Klebsiella sp*	1.5
		E.coli	1.9
2	*Enteromorpha flexuosa*	*Klebsiella sp*	2.5
		E.coli	3.7

CONCLUSIONS

Several compounds isolated from marine organisms are of pharmacological importance. There have been reports on the isolation of bioactive compounds for deadly diseases like cancer, arthritis, AIDS etc. The present work was done to isolate and identify the compound

ISSN: 0975-8585

from the two species of marine algae. Elucidating the structure of the phenolic compound thus isolated is of prime importance before it can be targeted for application.



a b

Fig-1: a) Separation of ethanolic algal extract using TLC; b) TLC plate after spraying

Gracilaria cortica *Enteromorpha flexuosa*

Fig-2: Zone of inhibition shown by the phenolic compound isolated from *Gracilaria cortica* and *Enteromorpha flexuosa* using TLC against *Escherichia coli*

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