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Bioactivity and phytochemical constituents of marine red seaweeds (*Jania rubens*, *Corallina mediterranea* and *Pterocladia capillacea*)

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Abstract

Seaweeds are potential renewable resources in the marine environment. The antibacterial activity of *Jania rubens*, *Corallina mediterranea* and *Pterocladia capillacea* were analyzed against human pathogenic bacteria. The present study was performed to investigate the phytochemical constituents of seaweeds, such as alkaloids, flavonoids, steroids, terpenoids and phlobatannins. In this study, we estimated phenols, flavonoids, tannins, pigments and mineral contents and determined the hydrogen peroxide scavenging activity, reducing power and total antioxidant activity of various extracts of selected seaweeds. Phytochemicals were extracted from the three seaweeds using various solvents, such as methanol, ethanol, acetone and chloroform. Among the various extracts, the methanolic extract was found to have the highest reducing power and total antioxidant capacity. We evaluated the seaweeds against *Vibrio fluvialis*, and *Pterocladia capillacea* was the most effective at controlling its growth. The highest zone of inhibition was recorded in the methanol extract. The chemical constituents of the seaweeds were characterized by GC–MS, which showed that they contain organic compounds, such as 1,2-benzenedicarboxylic acid.

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1. Introduction

Since ancient times, macroscopic marine algae has been closely associated with human life and has been exhaustively used in numerous ways as a source of food, feed, fertilizer and medicine, and chiefly used for economically important phycocolloids [1,2]. Marine algae

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contain more than 60 trace elements in a concentration much higher than in terrestrial plants. They also contain protein, iodine, bromine, vitamins and substances of stimulatory and antibiotic nature. The phytochemicals from marine algae are extensively used in various industries such as food, confectionary, textile, pharmaceutical, dairy and paper, mostly as gelling, stabilizing and thickening agents. Seaweeds or marine macro algae are renewable living resources that are also used as food, feed and fertilizer in many parts of the world.

In addition to vitamins and minerals, seaweeds are also potentially good sources of proteins, polysaccharides and fibres [3,4]. Recently, Hebsibah and Dhana Rajan [5] studied variations in the chemical constituents of the marine red alga *Hypnea valentiae* from the

Tuticorin and Mandapam Coasts. Dinesh et al. [6] studied the nutritive properties of 20 species of seaweeds from the Gulf of Mannar. Seenivasan et al. [7] screened the antibacterial activity of extracts of marine algae representing *Chlorophyta* and *Rhodophyta* collected from the Vishakapatnaam Coast against two pathogens and also tested their ability to inactivate the enzyme penicillinase in vitro. Extracts of marine algae were reported to exhibit antibacterial activity [8,9]. Vanitha et al. [10] reported the antibacterial action of nine seaweeds collected from the Kanyakumari Coast against human upper respiratory tract pathogens, which include both grampositive and gram-negative bacteria.

Kandhasamy and Arunachalam [11] determined the in vitro antibacterial properties of the seaweeds Caulerpa racemosa, Ulva lactuca, Gracilaria foliifera, Hypnea musciformis, Sargassum tenerrimum, S. myriocystem and Padina tetrastromatica collected from Koodankullam, and Tirunelveli against gram-negative and gram-positive pathogenic bacteria. Anitha et al. [12] determined the antibacterial activity of methanol, diethyl ether, acetone and dichloromethane extracts of Padina Boergesenii collected against 10 human pathogenic bacteria. Marine resources are an unmatched reservoir of biologically active natural products, many of which exhibit structural features that have not been found in terrestrial organisms [13]. There are numerous reports on compounds derived from macro algae with broad ranges of biological activities, such as the antimicrobial, antiviral, anti-tumour, anti-inflammatory, and neurotoxic [14]. The present study was performed with three marine seaweeds: Jania rubens, Corallina mediterranea and Pterocladia capillacea red algae. The study was performed with the following objectives: (1) To investigate the preliminary phytochemical constituents present in the three seaweeds. (2) To estimate the biochemical composition and photosynthetic pigments of the selected seaweeds. (3) To analyze the mineral composition of the three seaweeds. (4) To evaluate the antibacterial activity of the three seaweeds. (5) To reveal the chemical constituents in the three seaweeds using GC-MS analysis.

2. Materials and methods

2.1. Collection and identification of seaweeds

The studied algal species were collected from the coastal area of Abu-Qir Alexandria – North Egypt. Algal samples were cleaned of epiphytes, and necrotic parts were removed. Then, cleaned samples were rinsed with sterile water to remove any associated debris. The cleaned fresh materials were shade air-dried and ground

into fine powder, as described by Gonzalez del Val et al. [15]. The samples were identified as, *Jania rubens* (Linnaeus), *Corallina mediterranea* (J. Agardh) and *Pterocladia capillacea* (Gmelin).

2.2. Preparation of seaweed extracts

Ten grams of powdered samples were extracted with 50 ml of solvents, such as methanol, ethanol, acetone and chloroform. The samples were kept in the dark for 72 h with intermittent shaking. After incubation, the solution was filtered through filter paper, and the filtrate was collected (crude extracts) and stored in the refrigerator until further use.

2.3. Gas chromatography and mass spectrometry analysis

Gas chromatography–mass spectrometry (GC–MS) analysis was performed using an Agilent GC-MC-5975C with a Triple-Axis Detector equipped with an auto sampler. The GC column used was fused with silica capillary column (length 30 m × diameter 0.25 mm × film thickness 0.25 µm) with helium at 1.51 ml for 1 min as a carrier gas. The mass spectrometer was operated in the electron impact (El) mode at 70 eV in the scan range of 40-700 m/z. The split ratio was adjusted to 1:10, and the injected volume was 1 µl. The injector temperature was 250 °C, and the oven temperature was kept at 70 °C for 3 min, rose to 250 °C at 14 °C min⁻¹ (total run time 41 min). Peak identification of crude seaweed extracts were performed by comparison with retention times of standards, and the mass spectra obtained were compared with those available in NIST libraries (NIST 11 – Mass Spectral Library, 2011 version) with an acceptance criterion of a match above a critical factor of 80% according to Musharraf et al. [16].

2.4. Estimation of flavonoid content

Total flavonoid content was determined according to the method of Chang et al. [17]. A one-ml aliquot of each extract was mixed with 0.1 ml of 10% aluminium chloride and 0.1 ml of 1 M potassium acetate. Methanol (2.8 ml) was added and kept at room temperature for 30 min. The absorbance of the reaction mixture was measured at 415 nm. The flavonoid content was expressed in mg/g, and Quercetin was used as a standard compound.

2.5. Estimation of tannin content

Total tannin content was determined according to the method of Julkunen-Titto [18]. Briefly, 50 µl of seaweed

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