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ORIGINAL ARTICLE

Antibacterial substances from marine algae isolated from Jeddah coast of Red sea, Saudi Arabia

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Abstract Marine algae are known to produce a wide variety of bioactive secondary metabolites and several compounds have been derived from them for prospective development of novel drugs by the pharmaceutical industries. However algae of the Red sea have not been adequately explored for their potential as a source of bioactive substances. In this context *Ulva reticulata, Caulerpa occidentalis, Cladophora socialis, Dictyota ciliolata,* and *Gracilaria dendroides* isolated from Red sea coastal waters of Jeddah, Saudi Arabia, were evaluated for their potential for bioactivity. Extracts of the algae selected for the study were prepared using ethanol, chloroform, petroleum ether and water, and assayed for antibacterial activity against *Escherichia coli* ATCC 25322, *Pseudomonas aeruginosa* ATCC 27853, *Stapylococcus aureus* ATCC 29213, and *Enterococcus faecalis* ATCC 29212. It was found that chloroform was most effective followed by ethanol, petroleum ether and water for the preparation of algal extract with significant antibacterial activities, respectively. Results also indicated that the extracts of red alga *G*. dendroides were more efficient against the tested bacterial strains followed by green alga *U*. *reticulata*, and brown algae *D*. *ciliolata*. Chemical analyses showed that *G*. dendroides recorded the highest percentages of the total fats and total proteins, followed by *U*. *reticulata*, and *D*. *ciliolate*. Among the bioflavonoids determined Rutin, Quercetin and Kaempherol were present in high percentages in *G*. *dendroides*, *U*. *reticulata*, and *D*. *ciliolate*. Estimation of saturated and unsaturated fatty acids revealed that palmitic acid was

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present in highest percentage in all the algal species analyzed. Amino acid analyses indicated the presence of free amino acids in moderate contents in all the species of algae. The results indicated scope for utilizing these algae as a source of antibacterial substances.

1. Introduction

Marine organisms are potential sources of bioactive secondary metabolites with potential for use in the development of new pharmaceutical agents (Abedin and Taha, 2008; EL-Gamal, 2010) and many of these substances have been demonstrated to possess interesting biological activities (Faulkner, 2002; Abdel-Raouf et al., 2008). Marine algae were reported to produce a wide variety of bioactive secondary metabolites as antimicrobial, antifeedant, antihelmintic and cytotoxic agents and the bioactive substances included alkaloids, polyketides, cyclic peptide, polysaccharide, phlorotannins, diterpenoids, sterols, quinones, lipids and glycerols (Cabrita et al., 2010) and marine macro-algae are considered as the actual producers of some bioactive compounds with high activity (Shimizu, 1996). Hence they have drawn great attention recently (Abdel-Raouf et al., 2008; Ibraheem et al., 2008; Al-Haj et al., 2009; Bazes et al., 2009; Vallinayagam et al., 2009; Cabrita et al., 2010).

Bromophenol compounds have been frequently encountered in various marine algae including red and brown algae. Especially, the red algae of family Rhodomaceae are known as a rich source of bromophenols (Oh et al., 2008). Some of these compounds previously isolated from the family exhibited a wide spectrum of pharmacological activities such as enzyme inhibition, cytotoxic, antioxidant, feeding deterrent, antiflammatory and antimicrobial activities (Tapiero et al., 2002; Matanjun et al., 2008; Jaganathan and Mandal, 2009; Williamson and Carughi, 2010).

The green algae *Ulva lactuca* commonly known as sea lettuce has long been used as food and as a traditional medical agent to treat helminthic infections, fever, urinary diseases and dropsy (Kim et al., 2007). The antimicrobial activity of *Ulva lactuca* was reported to be caused by the acrylic acid commonly found in the algae (El-Yamany, 2008). Extract of green algae *Caulepra prolifera* was reported to exhibit significant activity against strains of marine bacteria (Rajasulochana et al., 2009). Adiphenyl ether isolated from the extract of the green algae *Cladophora fascicularis* was found to inhibit the growth of *Escherichia coli*, *Bacillus subtilis* and *Staphylococcus aureus* (Kuniyoshi et al., 1985). Diterpene isolated from unidentified species of *Dictyota* exhibited significant cyotoxicity (Awad, 2004). Extracts from algae from Indian waters *Dictyota dichotoma*, and *Padina gymnosora* were reported to be effective against *Bacillus megatherium* and *S. aureus* (Rao et al., 1977). Ethanolic extracts of *Zandariania prototypus*, *Cystoseria sricata* and *Cymbula compressa* were reported to inhibit the growth of different bacteria and fungi (Pesando and Caram, 1984). Extracts of Egyptian marine algae *Dictyota dichotoma*, *Dilophus fasciola* and *Cystoseria barbata* were found to show antibacterial activities (El-Naggar, 1987). Antibacterial effects of hexane and methanol extracts of the macro-algae *Mastocarpus stellatus*, *Laminaria digitata* and *Ceramium rubrum* on 12 marine and 7 prominent fish pathogenic bacteria were also reported (Dubber and Harder, 2007). Methanolic extracts from 32 macro-algae from the Atlantic and Mediterranean coasts of Morocco were evaluated for the production of antibacterial compounds against *E. coli, S. aureus*, *Enterococcus faecalis* and *Klebsiella pneumonia* (Ibtissam et al., 2009). The antibacterial activities of four important seaweeds namely *Ulva lactuca*, *Padina gymnospora*, *Sargassum wightii* and *Gracilaria edulis* were screened against human bacterial pathogen (Vallinayagam et al., 2009).

In fact the potential biological resources of marine environments of the Kingdom of Saudi Arabia represented by the Red sea and the Arabian Gulf have not been adequately explored and harnessed for biotechnological applications and deriving biopharmaceuticals. In this context a study was undertaken to isolate and evaluate prospective bioactive substances from marine algae of the Red sea coast. Herein we report the isolation of antibacterial substances from selected species of algae and the phytochemical composition of the extracts of algae that showed antibacterial activities.

2. Materials and methods

2.1. Algae

Algal materials were collected from the littoral zone of the Obhor region between (0.2–2.5 m depth) along the Red sea coast of Jeddah, Saudi Arabia. The collected algal samples were stored in plastic bags and transported to the laboratory under iced conditions. The samples were initially washed thoroughly with sea water to remove sand and any adhering substance and then washed thoroughly with fresh water to remove salts, and stored at −20 °C until compound extraction. The algal species were identified based on the schemes reported in the literature (Nasr and Aleem, 1949; Smith, 1944; Levring, 1946; Bouck, 1965; Scagel, 1966; Bold, 1978; Aleem, 1993; Coppejans et al., 2009) and saved in the Lab of Phycology, Botany and Microbiology Department, Faculty of Science, King Saud University (El-Malaz Center), Riyadh, Saudi Arabia.

2.2. Extraction of selected algal species

After washing with distilled water for several times, the algal samples were again washed with 5% ethanol to remove any epiphytes or any salts. One portion of the samples was kept under frozen condition and another small portion was kept in media containing Sea water-Formalin-Glycerol-Cupper sulfate. The remaining parts of the samples were subjected to air drying under the shade. After drying they were ground either mechanically or by an electrical mixer untill they became a powder. Then the powdered samples were stored in a dark place, and subjected to different extraction methods.

Extraction of powdered algal samples was done using ethanol, chloroform, petroleum ether and water. Aliquots of