

## Antimicrobial potential of seaweed epiphytic bacteria against nosocomial pathogens

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### Abstract

Seaweed Epiphytic microorganisms have transformed into a critical goal for the biotechnology business because of the tremendous number of bioactive blends actually found from them. In this current review, the antimicrobial strains were isolated from Seaweed gathered from Bay of Mannar, South east shore of India. Totally 101 organisms were isolated from five particular Seaweed. Ethyl acidic corrosive concentrates of bacterial supernatant were assessed for antibacterial action. In this manner, ten bacterial species have the wide reach antibacterial activity against human organisms. Generally, the hostile strains were having a spot with the genera *Pseudomonas*, *Alteromonas*, *Pseudo alteromonas* and *Bacillus*. In this study contemplated that the Seaweed related microorganisms can conveyed bioactive combinations may be significant in biomedical applications.

Keywords:Antibacterial; bacteria; epiphytes; seaweed; pathogens.

### Introduction

Seaweeds are one of the colossal and different natural frameworks; it expects a major part in marine climate. It is on a very basic level drew in with overall fundamental creation and giving food and haven to collection of natural elements. Ocean growth supplies protected and supplement rich conditions for the bacterial turn of events. (Armstrong *et al.*, 2000), so diverge from other multicellular animals, Seaweed protects a rich assortment of related microorganisms, these microorganisms maybe supportive or destructive to the Ocean growth. In particular, epiphytic bacterial organizations have been represented as major for morphological improvement of ocean growth, and microorganisms with antibacterial properties are made sure to defend the Seaweed from organisms and surface colonization of other contention animals. Since the last piece of the 1980s, more than 50,000 bioactive ordinary things have been found from marine microorganisms. Among these blends more than 8,000 had bactericidal development (Betina 1983, Berdy, 1989). Marine microscopic organisms as often as possible produce bactericidal combinations for staying aware of the

associations between epiphytic bacterial organizations and curbing microbial microorganisms (Avendaño-Herrera *et al.*, 2005). A couple of bacterial creature classifications show have distinction and bactericidal activity against express microorganisms; this unequivocality attract complex biochemical associations among Ocean growth and microscopic organisms (Strobel, 2003). In this momentum concentrate on zeroed in on the groundwork screening of the anti-microbial conveying microorganisms related with different Seaweed of Bay of Mannar, South east coastline of India.

## Materials and methods

Five seaweed samples such as *Gracillaria corticata*, *Hypnea musiformis*, *Ulva fasciata*, *Padina tetrastromatica* and *Valoniopsis pachynema* are collected *in situ* at a depth of 0.5 - 2 m during November 2018 from different locations in the coastal area of Mandapam, Southeast Coast of India (latitudes 9 ° 17 ° 17 ° No Nom, Longitude 79 ° 22 e). Seaweed samples are picked by hand and immediately washed with sea water to remove foreign particles, sand particles, and macro epiphytes. The sample was transported to the laboratory on the ice. Seaweed samples seem healthy at the time of collection. Sterile swab are used to rub the surface of seaweed; Swab is dipped in sterile sea water, and diluted serially with sea water. The serially diluted samples were spread on the Zobell Marine plates. The plates were incubated for 24-48 hours at 28 ° C. After incubation of bacterial colonies is calculated using a colony counter.

Bacteria are separated based on the morphology of colonies, cell morphology and coloring. Pure bacterial colonies are identified using morphological, biochemical and physiological tests.

## Preparation of raw extract

Bacterial culture overnight (100 ml) with O.D 0.6-0.9 is centrifuged at 4°C at 7000rpm for 20 minutes. Supernatants are collected and extracted with ethyl acetate. The organic layer is concentrated with vacuum and concentrated powder dissolved in water deionization and used for bioassay for several human pathogens such as *E.coli*, *Staphylococcus sp.*, *Staphylococcus sp.*, *Klebshillapneumonia*, *Pseudomonas aeruginosa*, *Micrococcus sp.*, *Salmonells sp.*, *Vibrio cholera*, *Shigella dysenteriae* and *Serratia sp.*

## Disk diffusion method

Mueller-Hinton is prepared and poured in the Petriplate in aseptic conditions. Concentrated powder is diluted with deionization of water and a concentration of 50 µg/ml is prepared. Using sterile discs (Himedia) prepared and soaked in a bottle and stand for 30 minutes. Pathogenic culture is highlighted in petriplates. Then the disc is placed on the mueller-hinton using forceps and petriplates incubated overnight without turning at optimal temperatures of 37°C for 24 hours. Experiments are repeated three times with duplicate.

## Results and Discussion

Seaweed and their surface related microbial organizations structure complex and significantly strong natural frameworks (Holmstrom *et al.*, 2002). A couple of new bacterial creature assortments and genera have been portrayed from ocean growth recommending that Seaweed address an interesting biotic environment for disclosure of new bacterial taxa, whether or not the start basically show a specific connection (Goeckee *et al.*, 2010; Seyedsayamdost *et al.*, 2011; Hollant *et al.*, 2012). A couple of phylogenetic examinations have given pieces of information into the complex epiphytic bacterial organizations related with ocean growth (Penesyane *et al.*, 2009). Yet exhaustive assessments of whole bacterial organizations on Seaweed surfaces are still commonly insufficient (Burke *et al.*, 2011), the open data suggest that bacterial organizations related with ocean growth are good for the Seaweed have.

Seaweed are normal living space to an alternate get-together of organisms with densities moving from  $10^2$  to  $10^7$  cells  $\text{cm}^2$  depending upon the host species, and season (Armstrong *et al.*, 2000; Bengtsson *et al.*, 2010). In this flow study, the amount of commonsense microorganisms on the ocean growth surface was explored; the rundown of culturable not entirely settled as CFUs. The bacterial depends on the external layer of the Seaweed tests were  $2.8 \times 10^3$  CFU per  $\text{cm}^2$  (*Gracillariacorticata*),  $3.7 \times 10^3$  for each  $\text{cm}^2$  (*Padina gymnosphora*),  $6.2 \times 10^3$  for each  $\text{cm}^2$  (*Valoniopsis pachynema*),  $5.6 \times 10^3$  for each  $\text{cm}^2$  (*Ulva fasciata*) and  $3.2 \times 10^3$  for each  $\text{cm}^2$  (*Hypneamusiformis*) (Figure 1). The typical taxa have been perceived on ocean growth surfaces yet for the most part at the phylum level. The typical epiphytic microorganisms consolidate people from Alphaproteo bacteria,

Gammaproteo microbes, Bacteroidetes, and Cyanobacteria was represented in different red, green, and earthy colored ocean growth (Hollant *et al.*, 2012). In like manner in this momentum concentrate totally 101 marine microbes were isolated from the Seaweed surface. In this, over portion of microbes are going under gamma proteobacteria. The alpha proteobacteria and firmicutes are moreover affluent in these epiphytic organizations in light of the fact that proteobacteria is ordinary and winning in maritime circumstances, as a rule (Cottrell and Kirchman 2000a, b; Biegala *et al.*, 2002; Simonato *et al.*, 2010). Due to Gram s gathering >60% of the epiphytes are gram negative organisms. Nakanishi *et al.*, (1996) have uncovered that couple of bacterial genera are stressed in morphogenesis of *U. pertusa* including, *Flavobacterium*, *Vibrio*, *Cytophaga*, *Pseudomonas*, *Escherichia*, and Gram-positive cocci.

Different examinations have as of late given insights about antimicrobial combinations of Seaweed starting (Paul and Ritson-Williams, 2008; Goeckee *et al.*, 2010). In this study the basic screening of hostile to microbial conveying microorganisms was analyzed against the human organisms. The microbes lined up with five Seaweed surfaces were isolated using Zobell marine agar that better mirror the enhancements open to Ocean growth epiphytes comfortable. These microorganisms were then refined in Zobell marine stock and the supernatant was isolated with ethyl acidic corrosive determination (Boyd *et al.*, 1999) and using a plate scattering procedure against human organisms, for instance, *E.coli*, *Staphylococcus sp.* *Staphylococcus sp.* *Klebshilla pneumonia*, *Pseudomonas aeruginosa*, *Micrococcus sp.* *Salmonells sp.* *Vibrio cholera*, *Shigella dysenteriae* and *Serratia sp.* (Table

1). Among the anti-toxin conveying microorganisms *Pseudomonas*, *Pseudoalteromonas* and *Alteromonas* sp. have serious solid areas for the development against most of the microorganisms. These ethyl acidic corrosive inference eliminated antibacterial combinations are maybe Quinolinol/pyrrole/normal blends or a polysaccharide (Jayanth *et al.*, 2002). In past assessments, Ismail-Ben Ali *et al.*, (2012) confined the bacterial genera

*Pseudomonas*, *Pseudoalteromonas*, *Paracoccus* and *Bacillus* from earthy colored green growth *Padina tetrastromatica* having the antibacterial activity. The ongoing result showed that the microbes Seyedsayamdost *et al.*, 2011; Hollant *et al.*, 2012). A couple of phylogenetic assessments have given pieces of information into the complex epiphytic bacterial organizations related with Seaweed (Penesyane *et al.*, 2009). Though exhaustive evaluations of whole bacterial organizations on Ocean growth surfaces are still commonly meager (Burke *et al.*, 2011), the open data suggest that bacterial organizations related with Seaweed are in a degree obvious for the Seaweeds epiphytes.

Burgess *et al.*, (2003) isolated a couple of microorganisms with high antibacterial activity against fouling life forms and saw that as by far most of these organisms had a spot with *Bacillus* sp. likewise, the earthy colored Seaweed *Sargassum* have more than 100 particular antimicrobial strains covering the phyla Proteobacteria, Bacteroidetes, Firmicutes, and actinobacteria (Wiese *et al.*, 2009). Jensen *et al.*, (2005) and Macherla *et al.*, (2005) reported that, 35 bacterial strains isolates from six Seaweed, it was plausible to phylogenetically recognize 33, arranged inside the phyla Firmicutes, Proteobacteria and Actinobacteria and these are seen as astonishing producers of bioactive discretionary metabolites. Finally this study agreed Zheng *et al.*, (2005) report, totally 341 bacterial withdraws were refined from marine sources. In these 42 separates are having antimicrobial development, among these, 11% of antibacterial strains are segregated from ocean growth surface, were has a spot with the genera *Alteromonas*, *Pseudomonas*, *Bacillus* and *Flavobacterium*. This study contemplated that the Seaweed related microscopic organisms are found to have a bactericidal activity against a couple of human microorganisms and further assessments are normal for different bioactive mixtures from ocean growth related organisms.

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Table.1 Zone of inhibition produced by seaweed epiphytic bacteria against human pathogens

Seaweed	Epiphytes	Human pathogens									
		<i>E.coli</i>	<i>Staphylococcus sp.</i>	<i>Streptococcus sp.</i>	<i>Klebsiella pneumonia</i>	<i>Pseudomonas aeruginosa</i>	<i>Micrococcus sp.</i>	<i>Salmonella asp.</i>	<i>Vibrio cholera</i>	<i>Serratia sp.</i>	<i>Shigella dysenteriae</i>
<i>Gracillaria corticata</i>	<i>Pseudoalteromonas sp.</i>	12.2mm	8.6mm	8.3mm	3.2mm	-	-	9.6mm	12.6mm	-	7.6mm
	<i>Pseudomonas sp.</i>	13mm	9.8mm	7.3mm	-	-	6.8mm	6.5mm	-	7.8mm	7.3mm
	<i>Pseudomonas sp.</i>	6.0mm	7.1mm	11.7mm	12.6mm	3.2mm	-	-	15mm	-	-
<i>Padina tetrastrum</i>	<i>Alteromonas sp.1</i>	4.8mm	16mm	14mm	11.8mm	13.2mm	11.2mm	-	-	-	12.7mm
	<i>Alteromonas sp.2</i>	6.8mm	-	-	8.38mm	13.3mm	-	12.6mm	15.2mm	9.3mm	11.2mm
<i>Ulva fasciata</i>	<i>Alteromonas sp.3</i>	3.1mm	8.9mm	9mm	2.6mm	3mm	11.6mm	-	-	-	-
	<i>Oceanobacillus sp.</i>	13.6mm	-	-	12.6mm	11.6mm	-	12mm	8.4mm	12.6mm	11.8mm
	<i>Bacillus sp.1</i>	13mm	7.6mm	12.1mm	-	-	11.8mm	-	4.6mm	8.9mm	10.4mm
	<i>Bacillus sp. 2</i>	15mm	-	-	12.7mm	11.8mm	-	13.2mm	16mm	12.8mm	14.4mm
<i>Hypnea musiformis</i>	<i>Rhodobacter sp.</i>	10.2mm	-	-	9.6mm	8.5mm	10mm	8.6mm	11.6mm	8mm	5.2mm