RESEARCH ARTICLE

Screening and Antimicrobial Action of Compounds from Marine Polysiphonia sp.

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Abstract: *Macroalgae are a biochemically diverse assemblage of organisms, including Polysiphonia, which are able to produce a wide variety of biological active compounds. These include antibiotics, toxins, pharmaceutically, active compounds and plant growth regulators. Recently Polysiphonia has become targets for screening programmes and search of novel compounds of potential valuable medicine. The present investigation were made in the Red seaweed of Polysiphonia sp The antibacterial properties of acetone extracts of Polysiphonia sp .were used against 4 types of ram positive and Gram negative bacterial strains and 3 types of fungal strains. The fresh and dried algal sample were used for obtaining extracts with the help of suitable Solvent.(Acetone, petroleum ether, and benzene) The solvent extract of (Acetone, petroleum ether, benzene) fresh sample of Polysiphonia sp. in disc diffusion method shows a prominent inhibitory zone ranging from 17–18mm in diameter against Staphylococcus aureus and Salmonella typhi. The solvent extracts of dry sample of Polysiphonia sp by disc diffusion method, of the anti-fungal activity was found to be maximum for A. fumigatus and A. sulphreus. In agar well method, the dry ethanol extract of Polysiphonia sp shows the prominent anti-fungal activity against for Aspergillus sulphureus.*

Keywords: *Antimicrobial Action, Screening, Marine polysiphonia.*

1. INTRODUCTION

Algae are a major source of food in the aquatic environment. It is commonly used as a food supplements to the humans and feed materials to the domestic animals. Further, it has been used as a source of biogas production and raw materials for the salad preparation. They are commonly distributed on shallow and sheltered areas of coastal regions. About 40% of marine environment covered by algal population. It includes green, red and brown algae

More than 1, 50000 species of macro algae are found in the oceans of the globe but only few of them were isolated from fresh water ecosystem. Some members of algae produced secondary [or] primary metabolites and this organism have to potentially enrich the bioactive compounds of interest to the pharmaceutical industry. Now days special attention have been made by many phychologist in the seaweeds for getting antiviral, antibacterial and antifungal compounds related to controlling of pathogenic microbes.

The members of Rhodophyceae have a large number of economic values, but a few species of red algae are also serve as food and feeds. Usually they are processed in large quantities, after harvesting. One of the red algae were used to preparing Agar-Agar, it is solidified agent for bacteriological and mycological media. Apart from these activities the Genus *Porphyra* is used as important raw materials for producing soaps and also

used for cooked flavouring agents in meet industry of China.

Gelidium and *Chondrus crispus* is a common red algal members for using edible purpose in human. It is used as stabilizing agent for chocolate production of food items and basically used as a chocolate milk pudding production. Jelly agents and similar other products. Were obtained from *Rhodymenia palmata* is the known red algae for the production of pharmaceutically valuable drugs.

Numerous synthetic drugs are used to cure various microbial diseases of man. Allopathic medicinal system offer immediate cure against disease due to the presence of active principle in it. But at the same time it has adverse actions. Homeopathic medicine did not give immediate remedies but gives permanent solution. Over dosage of drugs can produce side effects but algae did not give any side effects. Algae are promising strains for the production of feed, fine chemicals and pharmaceutical compounds.

2. AIM AND OBJECTIVES OF THE STUDY

The present study was initiated to find out the biological active compounds and their antimicrobial activity with the following objectives:

- Culturing of marine red algae P.dichomata
- Analysis of biologically active compounds such as Protein, Carbohydrate, Vitamin B2 (riboflavin) and Amino acid (Lysine) and,
- To study the antibacterial activity of solvents extract of algae using Disc diffusion and agar well methods at various concentrations.

3. MATERIALS AND METHODS

3.1. Collection of marine red algae

The marine red algae *Polysiphonia Sp.* was collected from the mangrove area of Muthupet at Thiruvarur district. During the time of sample collection, the temperature of water and pH were recorded. The fresh algae were attached on the submerged rock area. The *Polysiphonia Sp.* were carefully collected from the substratum by using the sterile scalpel and placed in sterile polythene bags and brought into the laboratory. The algal sample was thoroughly washed several times with distilled water and stored in refrigerator for further study.

3.2.Identification marine red algae

The identified algal samples were comes under the group of Alginophytes, namely Polysiphonia Sp.

3.3. Morphological features of Polysiphonia sp

- The Polysiphonia Sp. is a member of red algae.
- They differ from their size and colour to one species to another.
- They usually appear red to purple colour. This colour is because of the over masking pigment- phycoerthythrin.

3.4. Bio- chemical features of Polysiphonia Sp.

Polysiphonia Sp. contains a different type of cell wall components, growth regulators and bio-chemical contents. *The* cells are connected to each other by cytoplasmic connections (Pit connections) i.e. Plasmodesmata. The outer layer of cell wall consist of pectic substances and the next to cellulose and protoplast. Each cell contain discoid chromatophore. They stored food materias.

3.5. Analysis of biological compounds of polysiphonia sp.

The blue colour developed by the reduction of the phosphomolybdic and phosphotunstic components in the Folin- Ciocalteau reagent by the amino acids tyrosine and tryptophan present in the protein plus the colour developed by the biuret reaction of the protein

with the alkaline cupric tzrtrate are measured in the Lowry's method.

3.6. Antimicrobial activity study

Bacterial cultures used

In our study strains Four bacterial members were used among these bacteria culture out of two in Gram-positive bacteria and another two are Gram negative bacteria.

Fungal cultures used

It is eukaryotic and non-photosynthetic achlorophillus group of organisms. They are commonly produced colonies on the growth medium or substratum. Majority of the fungi are grow on the other host (Parasitc). Few of them grow on non-living matter (Saprophytes).

Fungal members are typical type of eukaryotic cell. It consist of pectin (or) in their cell wall with distinct protoplasm.

- Preparation of algal extract
- Preparation of dry algal extract
- Preparation of antibiotic disc
- Assay of antimicrbial activity

3.7. Antibacterial activity of extract by Disc diffusion method

Sterile Muller Hinton agar medium was prepared in sterile petriplates. After solidification, the test bacterial cultures were inoculated by means of agar swab method using sterile cotton swab. The prepared extract discs were placed on the surface of the plates of seeded bacterial cultures. Control (Acetone) and standard (Ampicillin) were also maintained. The plates were incubated at 37°C for 24 hours. After incubation the zone of inhibition was measured and expressed as mm in diameter.

3.8. Antifungal activity of solvents extracts by agar well method

Sterile MHA plates were prepared. After solidification, the test bacterial cultures were inoculated by means of swab method using sterile cotton swab. Using sterile cork borer, the agar well (5mm size) was prepared at respective place in Muller Hinton Agar plates. The prepared extract of different microlitre (50, 100,150 and 200µl) were loaded in well containing Muller Hinton Agar plates by using micropipette. Control was also maintained with the solvent acetone. The plates were incubated at 37°C for 24 hours. After incubation the zone of inhibition was measured and expressed in mm in diameter.

4. RESULTS

The present study was revealed that the red algae Polysiphonia Sp. (Plate. 1) were used for the morphological, phytochemical and antimicrobial activity.

4.1. Morphological features of Polysiphonia Sp.

Polysiphonia morphologically shows various in colour from red to purple. They have two type of thallus system namely (1) creeping basel system (2)vertical (or) erect system.

The Polysiphonia is a member of marine red algae. They differ from their size and colour to one species to another. It contains discoid chromatophores. They stored food material is namely floridean starch. After the photosynthetic activity. Polysiphonia reproduce either vegetatively or sexually.

4.2. Bio- chemical features of Polysiphonia Sp.

The member of Polysiphonia contain different type of cell wall components, growth regulators and bio-chemical contents. The algal cells have a distinct cell wall, which made up of pectin substances.

The inner region of the cell wall consist of alginic acid, fucodian granules and laminarin etc., The component and fucodian fractions of Polysiphonia Sp. having antitumor activity. The present study was carried out to analyze the biological compounds of Polysiphonia Sp.

and its antibacterial activity against bacterial pathogens.

4.3. Analyses of biological compounds

Analysis of Protein, Carbohydrate, Crude Fiber, Vitamin B2 and Lysine were determined from Polysiphonia Sp. and tabulated in Table.1.

4.4. Antimicrobial activity in Polysiphonia Sp.

The antagonistic effects of the extracts in Polysiphonia Sp. against different pathogenic group of bacteria and fungi. It includes both Gram positive and Gram negative bacterial cultures and *Aspergillus* and *Penicillium* cultures were used. Comparative studies on the bacterial and fungal activity were made by using crude fresh and dried algal extracts of Polysiphonia. The antimicrobial activity were observed from algal samples by using disc diffusion and agar well method (Plate 2 & 3).

Antibacterial activity of acetone extract of Polysiphonia Sp.(fresh samples)

The crude extract of Polysiphonia Sp. in disc diffusion method shows a prominent inhibitory zone ranging from 16–18mm in diameter against *Staphylococcus aureus* and *Salmonella typhi*. The same algal crude extracts were showed lowest inhibitory activity for *Staphylococcus aureus* (17mm) and *Escherichia coli* (18mm). This was represented in Table 2.

Antibacterial activity were performed by fresh petroleum ether extract of Polysiphonia Sp.

The crude extract of Polysiphonia Sp. assayed in disc diffusion method. It shows a prominent inhibitory zone ranging from (9-14)mm in diameter against *Bacillus subtilis* and *Salmonella typhi* and algal crude extract were showed lowest inhibitory activity for *Staphylococcus aureus* (13mm)and *Escherichia coli* (11mm). This was represented in Table 3.

Antibacterial activity were observed from fresh benzene extract of Polysiphonia Sp.

The extract of Polysiphonia Sp. in disc diffusion method shows a prominent inhibitory zone ranging from (12-18)mm in diameter against *Salmonella typhi* and *Staphylococcus aureus* the same algal crude extract were showed lowest inhibitory activity for *Bacillus subtilis* (11mm) and *Escherichia coli* (12mm). This was represented in Table 4.

Anti-fungal activity in acetone extract of dry sample of Polysiphonia Sp. (Plate 4&5).

The acetone extract in dried samples of Polysiphonia Sp. the anti-fungal activity was found to be maximum zone of inhibition against *A. fumigatus* (23mm). The lowest activity was found (21mm) in diameter for *A.sulphreus* respectively (Table.5).

Anti-fungal activity in petroleum ether extract of dry sample on Polysiphonia Sp.

The petroleum ether extract of dried of *Polysiphonia dichotoma* shows the anti – fungal activity was found to be maximum zone of inhibition against *A. sulphreus* (20mm). The lowest was showed (18mm) in diameter for *A. fumigatus* respectively (Table 6).

Anti-fungal activity in benzene extract of dry sample on Polysiphonia Sp.

The benzene extract of dried sample for Polysiphonia Sp. shows anti-fungal activity was showed maximum zone of inhibition against *A. sulphreus* (20mm) diameter. The lowest anti-fungal activity

was found in (18mm) diameter for *A. fumigatus* (Table 7)

5. DISCUSSION

Screening of the antibacterial activity of phlorotannins extracted from brown algae and tested against food borne illness-causing bacteria. The phlorotannins which are oligomers of phloroglucinol compounds extracted from thalli of brown algae *Ecklonia kurome* was reported by Kokinagayama *et al*., (2002)

Sargassum pilgiophylum and *S.tenerrimum* have best antimicrobial activities against several test bacteriaexperimentally proved by Naqui *et al*., (1981). Suresh Gunasekaran and Muthukumaran Gunasekaran(1997) was studied antifungal activity of *Aspergillus fumigatus* to a series of Alfa,beta-unsaturated styryl ketones known to be thiol-alkylaters was examined, and the result were compared with those obtained for *Candida albicans*.

In our study various degrees of activity were present in 18 out of the 24 algae extracts. The highest activity was rhodophyta diameter of the inhibited zone ranged from 10- 22mm the lowest was found in chlorophyta 8-12mm. Mahasneh.l *et al*., (1995). In our study activity the present in east and fungi.. MIC of the antibiosis varied from 20-100 mg/ ml.(Hagul *et al.*, 1996).

In the study of in vitro antibacterial activity of the complex on *E.coli* and *Staphylococces aureus* was better than that of plain drug as evidenced from the reduction in MIC value. (Aithal *et al*.,1996)

Antimicrobial action of penicillin and some of its derivatives including fosfomycin in our study used gram positive and gram negative bacteria (Sujatha G Dasdiar *et al.,*1997).

In our study used for antifungal activity of *Aspergillus fumigatus* to a series of series of result were compared with those obtained for *Candida albicans* . Suresh Gunasekaran (2002).

6. SUMMARY

The present investigation were made in the Red seaweed of Polysiphonia Sp. is commonly found in East Coast region of Muthupet mangrove. These algae normally distributed in the marine water, especially tide and intertidal region. They are grown on the rocky substratum. It has several centimeters in height. Polysiphonia Sp. morphologically differ from other species of Polysiphonia to their size ,shape and its colour varies from red to purple. This colour is because of over masking pigment of r-phycoerythrin. Bio- chemical features of cell wall of Polysiphonia Sp. made up of pectin substances and the layer next to protoplast of cellulose. The antibacterial properties of acetone extracts of Polysiphonia Sp. were used against 4 types of ram positive and Gram negative bacterial strains and 3 types of fungal strains.

The fresh and dried algal sample were used for obtaining extracts with the help of suitable Solvent.(Acetone, petroleum ether, and benzene) In this study solvent extract of algae were used in disc diffusion, agar well method against the pathogenic bacteria and fungi. The solvent extract of (Acetone, petroleum ether, benzene) fresh sample of Polysiphonia Sp. in disc diffusion method shows a prominent inhibitory zone ranging from 17 –18mm in diameter against *Staphylococcus aureus* and *Salmonella typhi*. The solvent extracts of dry sample of Polysiphonia Sp. by disc diffusion method, of the anti-fungal activity was found to be maximum for *A. fumigatus* and *A. sulphreus*. In agar well method, the dry ethanol extract of *Polysiphonia dichotoma* shows the prominent anti-fungal activity against for *Aspergillus sulphureus.* Macroalgae are a biochemically diverse assemblage of organisms, including Polysiphonia, which are able to produce a wide variety of biological active compounds. These include antibiotics, toxins, pharmaceutically, active compounds and plant growth regulators. Recently Polysiphonia has become targets for screening programmes and search of novel compounds of potential valuable medicine.

7. CONCLUSION

The present investigation was reveled that the phonological, phytochemical and antibacterial activity in red algae of Polysiphonia. Among the whole group of algae, the Red algae shows the better inhibitory activity against bacteria and fungal pathogens. Among the large group of algae the Red algae shows a promising medicinal and economic value in the human society.

Table 1: Antibacteral activity in benzene extract of fresh polysiphonia dichotoma by disc diffusion technique.

S.No	Name of Bacteria	Control	Zone of inhibition (Represented by mm in diameter)			
			50µl	100µl	150µl	200µl
1	*Bacillus subtilis*	--	8	11	10	10
2	*Staphylococcus aureus*	--	9	11	12	10
3	*Escherichia coli*	--	7	8	8	12
4	*Salmonella typhi*	--	8	14	15	18

Table 2: Anti-fungal activity in acetone extract of dry polysiphonia dichotoma by disc.

Diffusion method. S.No	Name of the fungi	Control	Zone of Inhibition (Represented by mm in diameter)			
			50µl	100µl	150µl	200µl
1	*Aspergillus fumigatus*	--	18	21	20	23
2	*Aspergillus sulphreus*	--	16	20	18	21

Table 3: Anti-fungal activity in petroleum ether extract of dry polysiphonia dichotoma by disc diffusion technique**.**

S.No	Name of fungai	Control	Zone of inhibition (Represented by mm in diameter)			
			50µ	100µ	150µ	200µ
1	*Aspergillus fumigatus*	--	12	18	17	18
2	*Aspergillus sulphreus*	--	16	20	18	17

Table.4: Anti-fungalbacterial activity in benzene extract of dry *polysiphonia dichotoma* by disc diffusion method**.**

S.No	Name of the fungi	Control	Zone of inhibition (Represented by mm in diameter)			
			50µL	100µL	150µL	200µL
1	*Aspergillus fumigatus*	-	12	18	17	18
2	*Aspergillus sulphreus*	-	16	20	18	17

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