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### BIOACTIVITY AND PHYTOCHEMICAL CONSTITUENTS OF MARINE BROWN SEAWEED (*PADINA TETRASTROMATICA*) EXTRACT FROM VARIOUS ORGANIC SOLVENTS

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#### ABSTRACT

Nowadays marine algae are having a numerous commercial applications in various fields. Marine algae produce 60 trace elements in a concentration much higher than in terrestrial plants then the macroscopic marine algae also contain protein, iodine, bromine, vitamins and substances of stimulatory and antibiotic nature. This present study was summarized to the antimicrobial activity of the organic solvents of the Marine alage *Padina tetrastromatica* and phytochemical analysis of *Padina tetrastromatica* with various organic solvents such as Acetone, Ethanol, Ethyl acetate, Methanol and Toluene. The antibacterial activity against *Bacillus subtilis*, *Proteus sp* and *Streptococcus sp* in various solvents extracts of *Padina tetrastromatica*. The higher inhibitory activity was in Ethyl acetate extracts of *Padina tetrastromatica* and lesser inhibitory effect was in Acetone. The responsible functional groups in the solvent extract were identified by Fourier Transform Infrared (FTIR). Mostly marine algal products play a crucial role in various fields. May it's become even more diversified and economically competitive.

**Key Words:-** Marine algae, *Padina tetrastromatica*, Antimicrobial Activity, Brown Seaweed.

#### INTRODUCTION

In the ancient time the macroscopic marine algae are closely related with human life they are being used in numerous ways as the source of food, feed, fertilizers, medicine (Levering *et al.*, 1969; Chapman, 1970). Marine algae produce 60 trace elements in a concentration much higher than in terrestrial plants then the macroscopic marine algae also contain protein, iodine, bromine, vitamins and substances of stimulatory and antibiotic nature. The phytochemicals from the marine algae are largely used in various industries like food, textile,

pharmaceutical, dairy and paper industries mostly used in gelling, stabilizing and thickening agents. In addition the seaweeds are good source of proteins, vitamins, polysaccharides and fibers. Then the Indian seaweeds are great food value and certain seaweeds contain 16 to 30% protein on dry weight and have all essential amino acids these are not available in vegetable food materials (Lahaye, 1991 and Darcy-Vrillon, 1993). In Biodiversity the screening of microorganisms seeking therapeutic drugs from natural products, there is a high level interest in marine organisms especially algae. During ancient times the marine algae have been known rich source of pharmacologically active compounds. (Faulkner 2000, Tziveleka *et al.*, 2003). The screening of micro and macro algae in marine systems has contain high levels of molecules with antibiotic, antiviral and anticancer

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activities and many other pharmacological properties (Borowitzka, 1999). In the marine environment is abode of many numbers of microorganisms (Nair and Simidu, 1987). Marine organisms are containing rich source of materials such as structurally unique natural products with pharmacological and biological activities (Faulkner 2001). Marine macro algae are the renewable living resources which are also used as food feed fertilizers in many parts of the world then the sea weeds are contain high level of antioxidants such as carotenoids, pigments, polyphenols, enzymes and polysaccharides (Morgan *et al.*, 1989). Then the sea weeds are occupying an important place of a source of biomedical compounds (Manilal *et al.*, 2010). Several works have been reported that the seaweed extracts exhibit the inhibitory activity against a number of gram positive and gram negative bacterial strains. And the many number of seaweeds have been studied for their antibacterial activity both in India and abroad (Pratt *et al.*, 1951; Chester and Stott, 1956; Burkholder *et al.*, 1960; Allen and Dawson, 1960; Nunez and Sanabria, 1975). *Padina tetrastromatica* is the family of Dictyotaceae one of most pertinent marine brown seaweed and it will contains the major component of fucopyranosyl residues containing sulfated fucans called as fucoidans, Alginc acid and glucans were isolated from the seaweed and minor amount of glucronic acid and uronic acid are also found and they will play a important role in the field of Antibacterial and Antiviral activities (Paramita, 1999).

#### ***Padina tetrastromatica* Characteristic Features:**

Divison : Phaeophyta  
 Order : Dictyotales  
 Family : Dictyotaceae  
 Genus : *Padina*  
 Species : *Tetrastromatica*

*Padina tetrastromatica* (Figure 1) is a yellowish brown fan shaped algae these are olive green Become drying (Mica1966). It grows in shallow and sand covered rocky pools at the adjacent coastal areas (Shameel 1990). It proliferates in tropical waters these algae are mainly used as seasoning in dried flake form and as table salt replacement for high blood pressure patients (Novaczek & Athy, 2001). These are studies which showed that it contains alginic acid, a major polysaccharides which show high anticoagulant (Prasada Rao, Sastrey, & Venkata Rao, 1984) and antiviral properties (Chatterji *et al.*, 2004). In this study, we describe the antibacterial activity of Acetone, Ethanol, Ethyl acetate, Methanol and Toluene extracts of *Padina tetrastromatica* algae obtained from the Tutucorin costal area. The effective extract activation was measured by well diffusion method and this organic solvents of brown seaweed algae extract characterization

and functional groups are analyzed by Fourier transform infrared (FTIR).

## **MATERIALS AND METHODS**

### **Sample collection**

The brown algae *Padina tetrastromatica* was collected from Tuticorin coastal area in South Tamilnadu, India.

### **Preliminary Phytochemical Analysis**

The plant extracts were analyzed for the presence of Proteins and amino acids, Flavonoids, Tannins and Phenolic compounds according to the standard procedures.

### **Test for proteins and amino acids**

Equal volume of 5% of sodium hydroxide and 1% solution of copper sulphate were added. Appearance of pink color shows the presence of proteins and free Amino acid.

### **Test for Anthraquinone glycosides**

#### **Borntrager's test**

The small quantity of the test solution was boiled with diluted sulfuric acid and filtered. Ether was added to the filtrate and shaken well. The separated organic layer was added with ammonia. The layer became pink to red. It indicates the presence of Anthraquinone glycosides.

### **Test for Flavonoids**

To the small quantity of the test solution, add aqueous sodium hydroxide solution, appearance of blue to violet color indicates the presence of anthocyanins, yellow color indicates the presence of flavones, yellow to orange indicates the presence of Flavonoids.

### **Test for Tannin and Phenolic Compounds**

The small quantity of the test solution was taken separately in water and tested for the presence of Phenolic compounds and tannins with following reagents such as

Diluted Ferric Chloride solution (5%) –  
 Violet Colour  
 1% Solution Gelatin containing 10% NaCl –  
 White Precipitate  
 10% Lead Acetate solution  
 – White Precipitate

### **Bacterial strains used**

The bacterial strains *Basillus subtilis*, *Proteus sp*, *Streptococcus sp* used for present study. Antibacterial assay

Method using Muller-Hinton agar medium. The inoculation of all agar plates with bacterial strains as Nutrient broth; (containing bacterial cultures incubated in shaker for eight hours at 37°C) Excess inoculums were removed by pressing the swab against the inner wall of the culture tube. The entire Antibacterial activity of algal extracts was determined by agar well diffusion agar plates were swabbed horizontally, vertically and outer edge of the plate to ensured heavy growth over the entire surface. All the culture plates were allowed to dry for about five minutes. Then the prepared extracts are poured in to the well in the standard concentration. All the plates were incubated for 24 hours at 37°C. Then the presence of zone of inhibition could be measured on the plates.

## RESULTS AND DISCUSSION

### Antibacterial Activity of *Padina tetrastromatica*

Antibacterial activity against *Bacillus subtilis*, *Proteus sp* and *Streptococcus sp* in different solvents extracts of *Padina tetrastromatica* have shown below. The higher inhibitory activity was in Ethyl acetate extracts of *Padina tetrastromatica* and lesser inhibitory effect was in Acetone and Ethyl acetate of *Padina tetrastromatica* were tabulated and recorded in Table (1)

### Preliminary Phytochemical Screening

Phytochemical screening tests indicated the presence and absence of, Proteins and Amino acids, Flavonoids, Glycosides, and Phenolic compounds,

follows: Sterile cotton swab was dipped in to a well mixed

Saponins, in different plant extracts. The Preliminary Phytochemical Screening of *Padina tetrastromatica* on different extracts has shown the following results. Acetone extract showed the presence of Proteins and amino acids, Flavonoids but the absence of Glycosides and Phenolic compounds. Toluene extract showed the presence of Proteins and amino acids, Flavonoids Glycosides and Phenolic compounds. Methanol extract showed the presence of Proteins and amino acids, Flavonoids Glycosides but the absence of Phenolic Compounds. Ethyl acetate extract showed the presence of Proteins and amino acids, Flavonoids but the absence of Glycosides Phenolic Compounds. Ethanol extract showed the presence of Proteins and amino acids, Flavonoids but the absence of Glycosides Phenolic Compounds.

### FTIR Analysis

FTIR spectra for *Padina tetrastromatica* by various organic solvents results shown in (Table 2) the adsorption peaks are noted in 518 to 3419cm<sup>-1</sup> the 518 peaks shows the Ti-O stretching vibrations 679 peaks shows the OH group, N-H stretching Vibrations 1° & 2° amines 1064 peaks shows the C-N stretching vibrations, Aliphatic amines 1392 peaks shows the alkenes - C-H bending vibrations 1642 peaks shows the -C=C- stretching vibrations and the 3419 peaks shows the phenol groups - O-H stretching vibrations.

**Table 1. Antibacterial activity of *Padina tetrastromatica* against different pathogens**

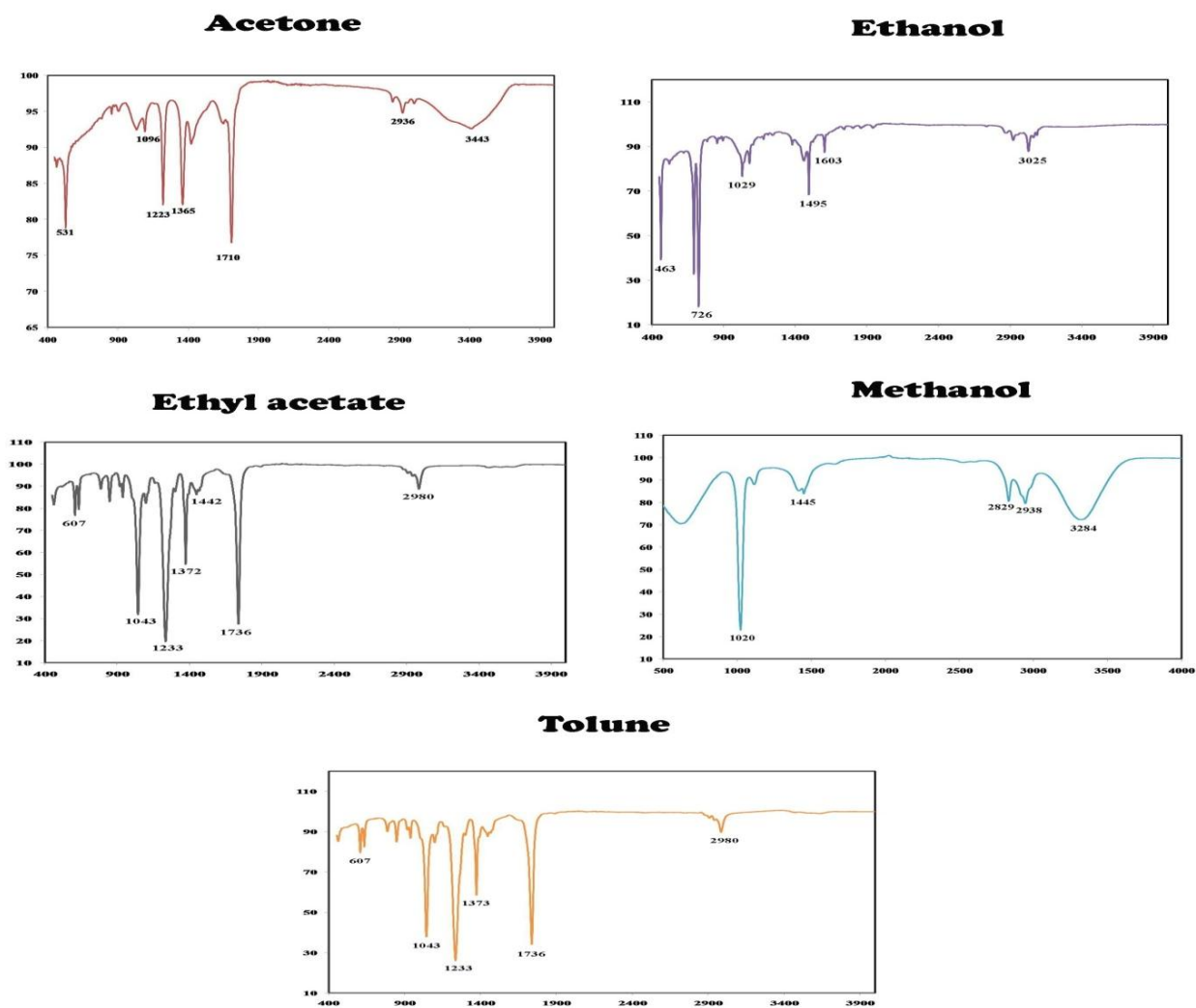
Bacterial culture	Zone of Inhibition (mm)				
	Acetone	Toluene	Methanol	Ethyl acetate	Ethanol
<i>B.subtilis</i>	17.33±0.667	19.33±0.667	18.83±0.667	17±0.289	21.16±0.601
<i>Proteus sp</i>	25.33±0.667	25.33±0.667	22.16±0.601	26.5±0.289	23.66±0.166
<i>Streptococcus sp</i>	22.83±0.166	22.16±0.166	23.83±0.166	20.16±0.441	20.16±0.441

**Table 2. Functional Groups present in *Padina tetrastromatica* by various organic solvents.**

S.No	Group frequency cm <sup>-1</sup> of the sample	Functional group assignment
1	518	Ti-O stretching vibrations
2	679	OH group, N-H stretching Vibrations 1° & 2° amines
3	1064	C-N stretching vibrations, Aliphatic amines
4	1392	alkenes - C-H bending vibrations
5	1642	-C=C- stretching vibrations
6	3419	phenol groups - O-H stretching vibrations

**Table 3. Preliminary Phytochemical Screening of extracts of *Padina tetrastromatica***

S.No	Tests	Acetone	Toluene	Methanol	Ethyl acetate	Ethanol
1	Proteins and Amino acids	+	+	+	+	+
2	Flavonoids	+	+	+	+	+
3	Glycosides	-	+	+	-	-
4	Phenolic compounds	-	+	-	-	-

Figure 1. *Padina tetrastratica*Figure 2. FTIR spectrum of *Padina tetrastratica* by various organic solvents Acetone, Ethanol, Ethyl acetate, Methanol and Toluene

## CONCLUSION

Seaweeds are marine macroscopic algae they are primitive non flowering plants without root, stem and leaves. They contain different vitamins, minerals. In the present study the Phytochemical screening of seaweeds showed the presence of proteins, amino acids and Flavonoids in the all the solvents and the glycosides are absence of acetone, ethyl acetate, and ethanol solvents. And the Phenolic compounds are absence of acetone, methanol, ethyl acetate and ethanol solvents. Among

the seaweeds the antibacterial screening of *Padina tetrastromatica* to controlling the growth of most of the pathogens the highest zone of inhibition (26mm) was recorded in Ethyl acetate extract of the *Padina tetrastromatica* against *Proteus sp.* FTIR spectra for the *Padina tetrastromatica* in the various solvents. The adsorption peaks are showed in the (Table 2). Thus the investigation brings out the phytochemical constituent and the antibacterial potential of the seaweeds.

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