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Research Article

# ANTIOXIDANT ACTIVITY OF SOME GREEN AND RED SEAWEEDS FROM WEST COAST OF MAHARASHTRA, INDIA

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**ABSTRACT**

Antioxidant potential of methanolic, ethanolic and aqueous extracts of three green (*Chaetomorpha antennina,* Kuetz, *Enteromorpha intestinalis* (L.) Nees, *Ulva fasciata* ) and three red seaweeds (*Acanthophora spicifera,* (Vahl) Borgesen, *Jania rubens* (Lin) and *Porphyra vietnamensis* T. Tanaka and Pham-Hoang Ho) collected from Kunakeshwar in Sindhudurg district of Maharashtra. In present study antioxidant activities was determined with respect to DPPH, FICA, $H_2O_2$, reducing power and total antioxidant capacity. DPPH, FICA and TAC is good in both green and red seaweeds. DPPH activity was more than 70% in most the samples in three different solvents. In methanol TAC was even greater than 130mg/g and maximum was recorded in *Enteromorpha intestinalis* 193mg/g. In *Enteromorpha intestinalis* the FICA was more than 60% in all the three solvents. *Chaetomorpha* and *Porphyra* exhibited better $H_2O_2$ activity in all the three media used. Reducing power is better in red seaweeds. Organic solvent revealed better activity, however $H_2O_2$ is better in aqueous medium for both green and red seaweeds.

**Keywords**: Seaweeds, DPPH, FICA, $H_2O_2$, TAC

## INTRODUCTION

Seaweeds are a potential resource in marine habitat which are exposed to a varied combination of light intensity and temperature. Inspite of the critical environmental conditions they exhibit an ability to cope up with the destructive effects of free radicals and other oxidative agents. They possess certain antioxidant properties which can minimize the oxidative damage caused by ROS in human cells leading to cancer and other diseases.[1] Food materials rich in antioxidant properties are hence recommended in human diet.

In present study antioxidant activities of some red and green seaweeds along the west coast of Maharashtra are reported.

## MATERIALS AND METHODS

### Collection of Seaweeds And Preparation Of Algal Extracts

Fresh and mature thalli of green (*Chaetomorpha antennina,* Kuetz, *Enteromorpha intestinalis* (L.) Nees, *Ulva fasciata* Delile) and red seaweeds (*Acanthophora spicifera,* (Vahl) Borgesen, *Jania rubens* (Lin) and *Porphyra vietnamensis* T. Tanaka and Pham-Hoang Ho were collected during low tide from rocky seashore at kunakeshwar ($16^0$ 40' 120' N Latitude and $73^0$ 28'.120' E Longitude) located in Sindhudurg district of Maharashtra (India). Fresh samples were washed thoroughly with fresh water to remove salt, sand particles and other epiphytes, dried in shade for 7 days, and then powdered using a grinder. One gram of seaweed powder was mixed with 10 ml solvent (methanol/ethanol/distilled water) and extracted for 24h using a rotary shaker.[2] The extract was filtered through Whatman no. 1 filter paper and stored in a glass vials in refrigerator at $4^0$c for further use.

### DPPH Radical Scavenging Activity

DPPH (2, 2-diphenyl-1-picrylhydrazyl) radical scavenging activity was determined according to the method[3]. 0.5 ml extract was mixed with 3 ml of 25mM DPPH solution prepared in methanol and incubated for 30 min. in dark at room temperature. Absorbance was measured at 517 nm against a blank (methanol without DPPH). Methanol with DPPH solution was used as the control. Percent inhibition of DPPH was calculated using the following formula.

$$* \% \text{ inhibition} = A_C - A_S / A_C \times 100$$

Where $A_C$ – Absorbance of control, $A_S$ – Absorbance of sample

### Ferrous Ion Chelating Activity (FICA)

Ferrous ion chelating activity was estimated following the method[4]. The reaction mixture containing 0.5 ml of extract, 0.1 ml 2mM $FeCl_2$ and 0.2 ml of 5mM ferrozine solution was incubated at room temperature for 10 minutes. Absorbance was measured at 562 nm. Percentage of ferrous ion chelating activity was calculated using the above formula * and expressed as percentage.

### $H_2o_2$ Scavenging Activity

The ability of the algal extracts to scavenge hydrogen peroxide was determined according to the method[5]. 0.5 ml extract was mixed with 0.6 ml hydrogen peroxide solution (40mM prepared in 0.2M phosphate buffer pH 7.4). Absorbance was recorded after 10 min. at 230 nm on a UV spectrophotometer against a blank solution containing the phosphate buffer without $H_2O_2$. Hydrogen peroxide scavenging activity of samples and standard ascorbic

acid was calculated using the formula *and expressed as percentage.

**Reducing Power**

Reducing power of the extract was determined according to the method[6]. In this assay ability of reduction of Fe III to Fe II was determined (ferric to ferrous) One ml of extract was mixed with 0.2 ml phosphate buffer (0.2M, pH 6.6) and 1ml potassium ferricyanide (1%) and incubated at $50^0$C for 20 minutes. After cooling, 1 ml trichloroacetic acid (10%) was added and mixed well. 1.5 ml of this mixture was transferred to other test tube containing distilled water (1.5 ml) and 0.1% $FeCl_3.6H_2O$ (0.5 ml). The contents were centrifuged and kept at room temperature for 10 minutes. Absorbance was read at 700 nm on a spectrophotometer. The antioxidant activity was expressed in terms of ascorbic acid equivalent and expressed as mg/g.

**Total Antioxidant Capacity (TAC)**

Total antioxidant capacity (TAC) of extracts was determined as per method[7]. The extract (0.5 ml) was diluted to 1 ml and mixed with 3 ml of phosphomolybdate reagent (0.6M $H_2SO_4$, 28mM sodium phosphate and 4mM ammonium molybdate were mixed together in 250 ml distilled water). This reaction mixture was kept in water bath at $95^0$C for 90 minutes, cooled and absorbance was recorded at 695 nm on a spectrophotometer. Ascorbic acid (0.1mg/ml in distilled water) was used as a standard and values were expressed as mg/g.

**RESULTS**

**DPPH Radical Scavenging Activity**

DPPH assay is used to test the ability of the antioxidant compounds present in the seaweed extract to function as proton radical scavengers or hydrogen donars[8]. In the present study DPPH activity was more than 70% in most the samples using three different solvents (Figure.1). It was maximum in methanol extracted samples and slightly lower in ethanol and water extracted algal material.

**Ferrous Ion Chelating Ability (FICA)**

Metal chelating ability of seaweed extract was tested at a concentration of 100mg/ml. FICA varied from 60 to 70% in methanol extracted seaweeds. In *Enteromorpha intestinalis* the FICA was more than 60% in all the three solvents. In aqueous medium FICA was reduced in all seaweeds except *Enteromorpha intestinalis*. In ethanol FICA varied much (45 to 77%) in different algal samples. (Figure. 2). In red seaweed the activity was more than green in all the three solvents used in present study.

**$H_2O_2$ Scavenging Activity**

The scavenging ability of methanolic, ethanolic and aqueous extracts of green and red seaweeds for hydrogen peroxide varied much and ranged from 16 to 46% in different seaweeds. *Chaetomorpha* and *Porphyra* exhibited better $H_2O_2$ activity in all the three media used.(Figure. 3). In green seaweeds this activity was better than red samples in all the solvents.

**Reducing Power**

Reducing power was high in red seaweeds analysed as compared to the green samples in all solvents and varied from 0.254 to 0.578 mg/g. (Figure. 4). In aqueous extracted samples it was slightly higher than in organic solvents. In red seaweeds this activity was more than the green ones.

**Total Antioxidant Activity (TAC)**

Total antioxidant capacity was more than 100mg/g in the algal samples extracted in ethanol. In methanol this activity was even greater than 130mg/g and maximum was recorded in *Enteromorpha intestinalis* 193mg/g (Figure. 5). In aqueous medium TAC was slightly at a lower level and less than 100mg/g in few samples. The activity was greater in green and red samples extracted in methanol than in ethanol.

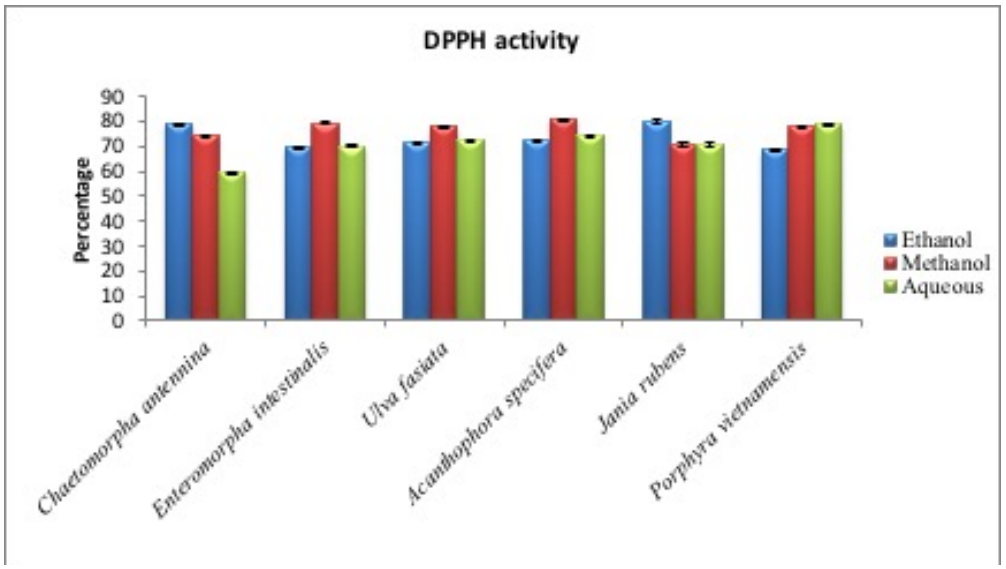


**Figure. 1. DPPH radical scavenging activity in green and red seaweeds from Sindhudurg coast of Maharashtra**

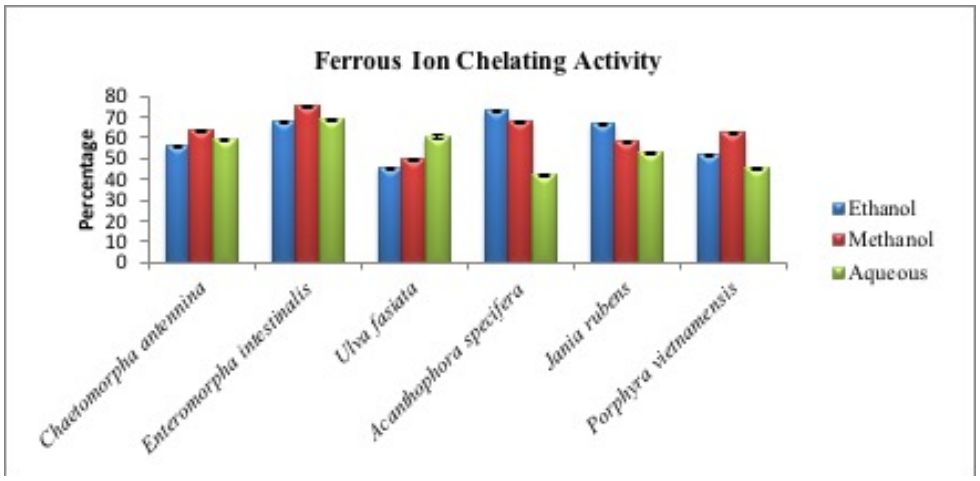


**Figure. 2. Ferrous ion chelating activity in green and red seaweeds from Sindhudurg coast of Maharashtra**

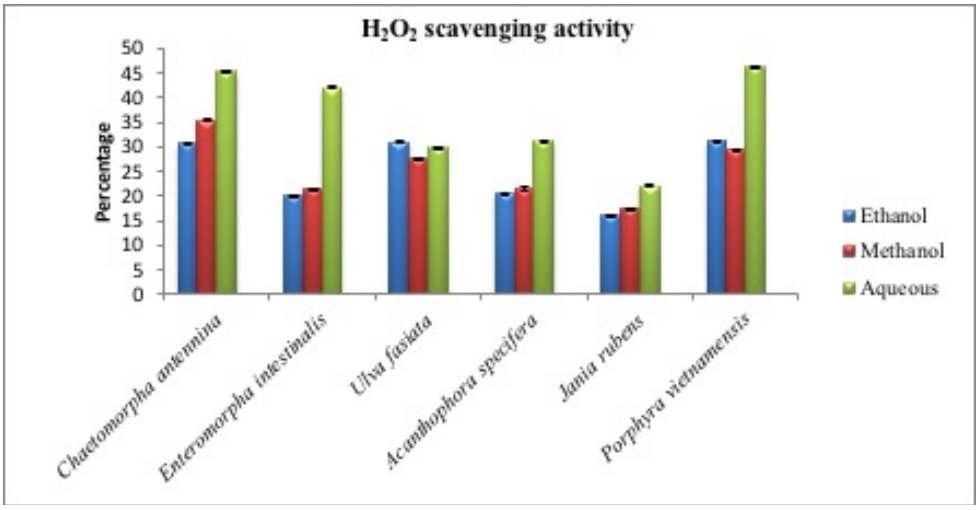


**Figure. 3. Hydrogen peroxide scavenging activity in green and red seaweeds from Sindhudurg coast of Maharashtra**

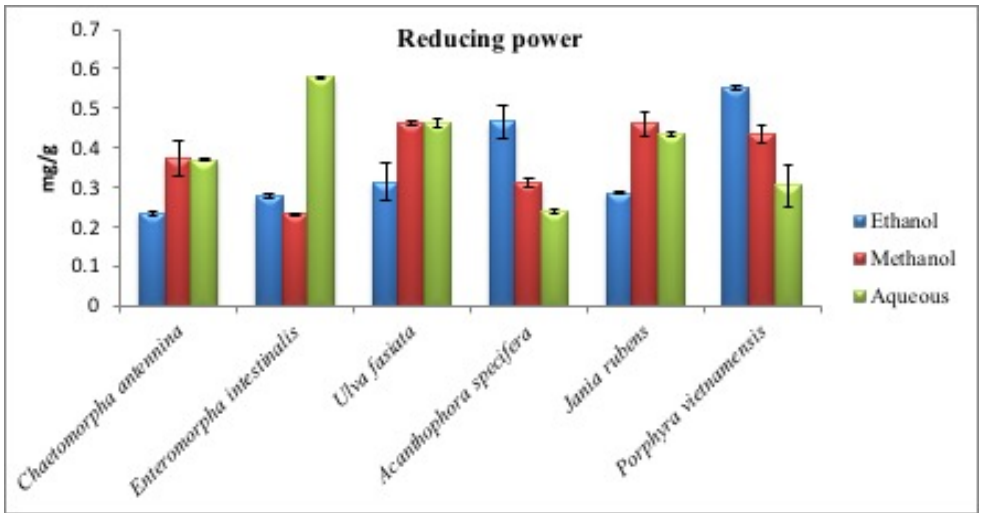


**Figure. 4. Reducing power in green and red seaweeds from Sindhudurg coast of Maharashtra**

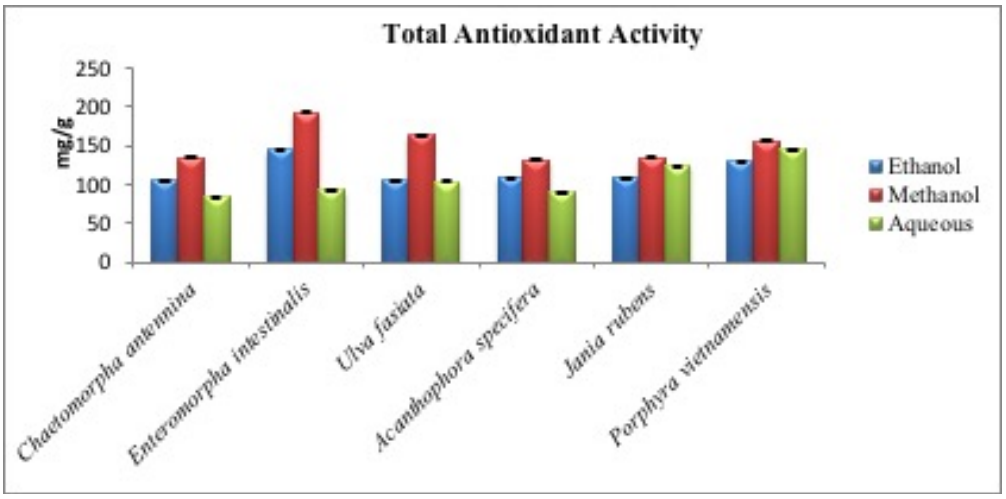


**Figure. 5. Total antioxidant capacity in green and red seaweeds from Sindhudurg coast of Maharashtra**

## DISCUSSION

The highest DPPH activity in methanolic extract of red seaweed *Porphyra* species (91.85%).[9] In *Gracilaria biradae* it was 60% in ethanol extract[10]. In *Gracilaria edulis* red seaweed a high activity in aqueous and methanolic extracts (95% and 82%) respectively was observed[11]. In methanolic extract of *Caulerpa scalpeliformis, Halimenia durvilae and Halimenia microloba* a low to moderate DPPH reaction has been reported [12]. Excellent DPPH radical scavenging in methanolic extract of *Enteromopha compressa*[13].

Higher $H_2O_2$ scavenging effect in aqueous and organic fractions of *Hijikia fusciformis* (red seaweeds)[14]. Hydrogen peroxide radical scavenging activity of *Gratilopia lithophila* (54%) and *Hypnea valentiae* (54%) respectively. Green seaweed samples $H_2O_2$ activity was about 30% [15]. Maximum activity in methanolic extracted *Corollina mediterrane, Pterocladia capillacea* and *Jania rubens*[16]. Highest reducing power activity in aqueous extract of *Scytosiphon lomentaria* than in ethanol extract[17].

Total antioxidant activity in methanolic extract of *Gracilaria edulis* (0.31 mg/g) which was very low when compared to the values obtained in the efficient solvent for the extraction of compounds with antioxidant activity[18]. In present study methanolic extraction of seaweeds was found to favour the antioxidant activity.

## CONCLUSION

DPPH, FICA and TAC was good in both green and red seaweeds. DPPH activity was more than 70% in most of the seaweed samples and was maximum in methanol extracted samples as compared to those in ethanol and water. $H_2O_2$ activity is less in both green and red seaweeds. Reducing power is better in red seaweeds. Organic solvent revealed better activity, however $H_2O_2$ is better in aqueous medium for both green and red seaweeds.

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