

STUDY OF PHYTOCHEMICAL AND ANTIOXIDANT ACTIVITIES OF GREEN ALGA Chaetomorpha antennina COLLECTED FROM RAIGAD COAST OF KONKAN

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ABSTRACT

The marine alga species *Cheatomorpha* was used to find out its phytochemical analysis and antioxidant activities. In this investigation, algal samples were collected along the coast of Raigad district of Konkan region, during low tides. Phytochemical screening of the algal extracts was carried out with using standard methods. The different extracts of *Cheatomorpha* showed the presence of alkaloids, flavonoids, glycosides, phenolic compounds, saponins, steroids, tannins etc. Antioxidant potential of *Cheatomorpha* carried out by using DPPH radical scavenging activity (%) and determination of total phenolic contents. The result of present study confirmed that *Cheatomorpha* is rich in secondary metabolites and it may be used as active antioxidant potential.

Keywords: *Cheatomorpha*, phytoconstituents, DPPH radical scavenging(%), total phenolic content.



INTRODUCTION

Marine algae are one of the most primitive types of plants in aquatic environment. They play ecologically & biologically important part in marine ecosystems Taylor, W.R. (1960). They grow on rocks in the intertidal zone as a giant underwater forest, which spreads in unlimited area. They also grow abundantly in shallow water of sea, backwater and estuaries, littoral and sub littoral rocky areas along the coast line all over world (Venkataraman, K. and Wafar, M. (2005).

The marine algae resources are abundant around coastlines of Mumbai, Palghar, Raigad, Ratnagiri & Sindhudurg in Maharashtra (Deodhar, H.D. (1989). Algae have been used since ancient times as food, fodder, medicine, fertilizer etc, in coastal area (Chapman AP. (1998).

Algae are potentially active sources of bioactive secondary compounds (Mittler R., 2002). They are able to produce a great variety of secondary metabolites characterized by a broad spectrum of biological activities (Patra J K, Rath S K, JenaK, Rathod V K and Thatoi. (2008). Algal compounds with antibacterial, antiviral, antifungal, antioxidant and antimicrobial activities have been detected in many species of green, brown & red algae(Yuan et al., 2005).

*Cheatomorpha*is marine macro an alga belonging to class Chlorophyceae & its family isGlomerataceae. It is a large in size and economically important. This green alga is ecologically dominant in much of the tropics, due to large appearance in aquatic ecosystem. It is well known algae obtained in various coasts of Konkan region. The present investigation was aimed to explore the phytochemical constituents of different extracts and antioxidant screening by using DPPH and total Phenolic contents of extracts of *Cheatomorpha*.

MATERIALS AND METHODS

Collection of marine algae:

In the present investigation, samples of macro marine alga i.e. *Cheatomorpha*were collected by hand picking, during low tide along the coast of Raigad district, Maharashtra, India (17°53' and 19°08' N Latitude , 72°51' and 73°42' E Longitude). The macro marine algae



were washed in sea water and fresh water thoroughly to remove the epiphytes and other contaminations. Then sample was transferred into a polythene bag with a small hole to leak out water drop wise and then shade dried.

Collection of algae was done in labeled polythene bags and brought to laboratory, and algal samples were analyzed macroscopically for their morphological characters like colour, shape, size, texture etc. Then collected species of algae were preserved in 4% formalin solution. Herbarium specimens of algal species were prepared for identification and confirmation of their taxonomic position. Identification of species was done by referring Taylor (1960), Deodhar (1987) and Dinabandhu sahoo (2001) and other previous publications.

Preparation of sample for qualitative phytochemicals analysis:

For the phytochemical screening, fresh samples were used. Five grams of fresh sample weighed and homogenized with 50 ml of water, ethanol, methanol, benzene, petroleum ether solution separately. The extract was boiled for one hour, cooled and filtered. The filtrate was used for screening phytochemicals by using standard procedure (Harborne, 1973).

Preparation of organic extracts of sample for Antioxidant activities:

The dried sample of seaweeds ground to coarse powder, weighed and wrapped in Whatman No.1 filter paper and successively extracted with 200 ml of different solvents such as benzene, chloroform, ethanol, ethyl acetate, methanol and petroleum ether with their increasing order of polarity by soxhlation for 12-24 hrs. The extract analyzed for the presence of antioxidant activities by referring standard procedure (Thoudam et al, 2011).

DPPH radical scavenging activity:

DPPH is a stable inorganic radical. To determine the radical scavenging effect of marine algal extracts, DPPH (1, 1-diphenyl-2-picryl hydrazyl) method is used. This method is based on estimating the reduction of alcoholic DPPH solution in the presence of a hydrogen donor. A solution of 0.135 mM DPPH in methanol was prepared and 1.0 ml of this solution was mixed with 1.0 ml of marine algal extracts in methanol with different concentrations (0.5-2.5 mg/ml). Then the reaction mixture mixed thoroughly and kept in the dark at room



temperature for 30 minutes and absorbance of the reaction mixture measured spectrophotometrically at 517 nm. BHT and BHA were used as references (Liyana-Pathiranan et al. 2005).

Determination of total phenolic content:

Total phenolic content in marine algal extracts was determined by using modified Folin-Ciocalteau reagent method. According to (Zahin et al 2009), gallic acid is a standard phenolic compound. The reaction mixture contained single concentration of the extracts i.e. 10 mg/ml and Folin- Ciocalteau reagent. To 500 μ L (10 mg/ml) of marine algal extracts in methanol, 2.5 ml of 1:10 dilution of Folin-Ciocalteau's reagent and 2 ml of Na₂CO₃ (7.5% w/v) were added and mixed thoroughly and incubated at 45^oC for 15 minutes. Same procedure is followed for other marine algal extracts with benzene, chloroform, ethanol, ethyl acetate and petroleum ether respectively. The absorbance measured at 765 nm. The concentration of total phenolic content in the marine algal extracts was determined as milligrams of Gallic acid equivalent per gram of dry weight (mg GAE/g dw) (Zahin et al 2009).

RESULTS

Sr.No	Name of the Algal Species	Solvent used	a	b	c	d	e	f	g	h	i	j	k
1	<i>Cheatomorphaantennina</i> (Bory) Kutzing.	water	I	-	+	+	+	+	+	I	+	I	+
		Ethanol	+	+	+	+	+	+	+	1	+	-	+
		Methanol	+	+	+	+	-	+	-	-	+	-	+
		Benzene	+	+	-	-	+	+	-	-	-	-	-
		Petroleum ether	+	-	-	-	+	-	-	-	-	-	-

 Table No. 1: Preliminary phytochemical analysis of Cheatomorpha.

Where, a: Alkaloids, b: Flavonoids, c: Glycosides, d: Phenolic compounds, e: Saponins,f: Steroids, g: Tannins, h: Carbohydrates, i: Proteins, j: Fats, k: Sugar and +: Present, -: Absent.



Antioxidant activities of *Cheatomorphaantennina*:

1) Table No.2: DPPH-radical scavenging activity (%) of *Cheatomorpha* at 517 nmis as follows,

Sr. No	Conc ⁿ (mg/ml)	DPPH-Radical Scavenging Activity (%) of Cheatomorphaat 517 nm.							
		Renzene	chloroform	Fthanol	Ethyl	Methanol	Petroleum		
		Denzene		Linunoi	Acetate	Witthanoi	Ether		
1	0.5	1.65	4.22	1.85	2.74	16.18	2.61		
2	1.0	5.40	8.73	5.07	4.93	33.07	4.14		
3	1.5	8.86	15.33	8.06	7.11	49.28	6.30		
4	2.0	11.78	22.46	10.78	9.17	64.07	9.41		
5	2.5	15.11	29.10	14.14	11.74	79.56	11.20		

2.Total phenolic contents of *Cheatomorphaantennina*at 765 nm in mg of gallic acid equivalent per gram is given below, (Concentration of extract used= 10mg/ml).

Table No.3: Total phenolic content of *Cheatomorpha* at 765 nm.

Sr.No	Solvent used	Absorbance at 765nm	Total phenolic content at 765 nm
1	Benzene	0.383 <u>+</u> 0.030	11.42 <u>+</u> 0.028
2	Chloroform	2.001 <u>+</u> 0.024	59.75 <u>+</u> 0.026
3	Ethanol	1.022 <u>+</u> 0.042	30.60 <u>+</u> 0.020
4	Ethyl acetate	2.074 <u>+</u> 0.052	62.01 <u>+</u> 0.018
5	Methanol	1.904 <u>+</u> 0.020	54.42 <u>+</u> 0.036
6	Petroleum ether	0.191 <u>+</u> 0.006	5.83 <u>+</u> 0.030



DISCUSSIONS

The present investigation brings out adequate data on phytochemical constitute and antioxidant potential of *Cheatomorpha*, collected from Raigad coastline.*Cheatomorpha*showed positive results for glycosides, phenolic compounds, saponins, steroids, tannins, proteins and sugar in water extracts. In ethanolic extract, alkaloid, flavonoides, glycosides, phenolic compounds, saponins, steroids, tannins, proteins and sugar are present. Methanolic extract, showed presence of alkaloids, flavonoides, glycosides, phenolic compounds, steroids, proteins and sugar. In benzene extracts, alkaloid, flavonoides, saponins and steroids are present. Petroleum ether extracts, showed presence of only two constituents i.e. alkaloids and saponins.

In antioxidant screening of the *Cheatomorpha*, by using DPPH-Radical scavenging activity, methanolic extracts showed highest radical scavenging activity (16.18%); followed by chloroform extracts (4.22%), ethyl acetate extracts (2.74%), petroleum ether extracts (2.61%), ethanol extracts (1.85%) and extracts of benzene showed lowest radical scavenging activity (1.65%).

In determination of total phenolic content of *Cheatomorpha*ethyl acetate extracts showed highest phenolic content (62.01); followed by chloroform extracts (59.75), methanolic extracts (54.42), ethanolic extracts (30.60), benzene extracts (11.42) and petroleum ether extracts showed lowest phenolic contents (5.83).

The results of *Cheatomorpha*were indicating that it may be useful against some potential human pathogens, which will be highly beneficial for human health. These results showed that *Cheatomorpha* have a less antioxidant potential, but due to its thallus structure it may be used as feed for grazing and domestic animals.

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