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Screening of Phytochemical Analysis of Selected Green Seaweeds Isolated From Mandapam Coastal Region

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INTRODUCTION

Marine algae is the one of the most important large group of microorganism in marine environment. It is present in tidal and intertidal region of the coastal area. Seaweeds play an important role in marine environment for that 90% are algae and over 50% algae were globally photosynthetic contributed algae. Seaweeds are single celled, microscopic flagellates. They don't have seeds and flowers, it contains only asexual spores.

Marine algae is classified in to three different groups such as Red algae, Green algae, Brown algae. It can grow only salty, brackish water areas. Seaweeds contain highly nutritive value substance such as minerals, vitamins, and polysaccharides. They were also contains bioactive compounds like proteins ,lipids and polyphenols.

The bioactive compounds play a vital role in biological activites such as antibiotics, antifungal,and antibacterial, antiviral, antioxidant and anti-inflammatory process. For that last few years seaweeds had been consumed as a medicine,food and their extract have also been used as a bioactive substance which have a lots of medicinal values. The bioactive compounds having a broad range of biological activity. Seaweeds do not have a toxic absorbing characters for any element. It is a toxic free substance.

Halimeda is the one of the important genus of green algae. It contains calcified green segments. Calcium carbonate is deposited in its whole algal body.

Division: Chlorophyta
Class: Ulvophyceae
Order: Bryopsidales
Family: Halimedaceae
Genus: Halimeda

Halimeda is a tropical benthic habitat. It is a unicellular, heterogenous group, the thalli were composed of multi-nucleate filaments. Halimeda lives in coral reefs habitat. The plant body is composed of branching ,calcified green filaments. The filaments joined together by flexible, uncalcified nodes. It can grow up to 25cm hight. Halimeda species contain photosynthetic pigments of the chlorophyta, siphonoxanthin and siphonein.

The aim of the study to evaluate the phytochemical analysis of Halimeda species. Phytochemical substance are non-nutritive chemical substance. They can also protect humans from different kinds of diseases. It can also play a vital role in antioxidant activity, antibacterial antiviral effects.

MATERIALS AND METHODS

Collection of Seaweeds And Sample Preparation:
The green seaweeds was collected from intertidal zone of Mandapam coastal region of Ramanathapuram district in Tamilnadu. The sample were manually collected, after sample collection, it should be washed thoroughly with seawater to remove the sand particles, epiphytes, pebbles and shells, again washed with tap water after that washed with distilled water to remove the surface salt concentration and microbes. After complete the washing process shade dried the sample for one week at room temperature and make it as a fine powder with the heip of electric blender.

Preparation O Crude Extrct Seaweed:

Methanol Extraction:

50g of the dried green seaweeds were extracted in 500ml of methanol solution .The mixture was kept in a orbital shaker for 24hrs at room temperature. The solvent were filtered by using Whattman filter No.1filter paper. The solvent were allowed to evaporate to get a fine concentrated substance. The residues were stored in a refrigerator for further studies.

Acetone Extraction:

50g of powdered were extracted in 500ml of acetone solution. The mixture was kept in a homogrnizer for 3 hrs at 32ºC.After completion of 3 hrs the extracted sample were filtered by WhattmanNo.1 filter paper. The solvent were allowed to dry .The fine powdered residues were stored in refrigerator for future studies.

Aquesous Extraction:

The seaweeds washed thoroughly with distilled water and shade dry for one week, make a fine powder. The powdered sample was sterilized at 120ºC for 15 mimutes. 2g of powdered seaweeds was taken and mixed with 150ml of distilled water or Milli Q water. The solution mixture was boiling in a water bath at 90ºC for 20 minutes. The extract was further made upto 200ml by adding of distilled water. The extract was filtered by using muslin cloth, extracted sample were kept in a refrigerator for further use.

Phytochemical Analysis

Test for Alkaloids-Dragendroff's Test

Take four clean test tube add 1 ml of extract in each tubes. Add few drops of Dragendr off's reagent were added to each tubes, mix it well, the colour changed in to red precipitates it shows the presence of alkaloids.

Test for Carbohydrates-Benedict test:

1 ml of filterate was taken in to a clean test tube, add few few drops of Benedict's reagent in each tube. Orange red precipitate indicates the presence of Sugars.

Test for Glycosides-Borntrager's Test:

Take clean test tube add 2ml of filtrate ,and 3 ml of Chloroform from each tube shake well,10% ammonia solution was added to it. The appearance of pink colour indicates the presence of Glycosides.

Test for proteins-Biuret test:

1 ml of extract was treated with 5% Sodium hydroxide solution to that add 1% Copper sulphate solution. Formation of pink or purple colour indicates the presence of Proteins.

Ninhydrin Test:

Few drops of Ninhydrin solution were mixed with 2 ml of extract. Appearance of purple colour indicates the presence of Amino acids.

Lead acetate Test:

1 ml of filterate extract was treated with 10% lead acetate solution. Formation of white precipitate shows the presence of Phenolic compounds.

Test for Tannins:

1 ml of extract were taken in clean test tube add 2 drops of Ferric chloride solution in each tube.A green colour formation indicates the presence of Tannins.

Test for Flavonoids-Alkaline reagent Test:

1ml of extracted sample were treated with few drops of Sodium hydroxide solution. Yellow colour indicates the presence of Flavonoids.

Test for Terpenoids-Salkowsk's Test:

1 ml of extract were taken in clean test tube add 1 ml of Chloroform mixed well, add few drops of concentrated Sulphuric acid for each tube. Formation of reddish brown layer, it shows the presence of Terpenoids.

Detection of Steroids:

1 ml of Chloroform was added to 1 ml of filterate to that add few drops of acetic acid, mix it well. In this mixture add concentrated Sulphuric acid .Appearance of blue and green colour indicates the presence of Steroids.

RESULT AND DISCUSSION

Extraction of Seaweeds:
The seaweeds collected from the Mandam coastal region ,after collection of sample washed thoroughly with tap water followed by distilled water to remove Debris, dust, and and soil particles.

The sample were shade dried for one week and make if fine powder .The powdered seaweeds contain a bioactive compounds. The bioactive compounds were obtained by using adifferent solvent extraction such as Methanol, Acetone and Aqueous solution. The extract were filterate by Whattman No.1 filter paper. The filterate substance were stored in a refrigerator for further phytochemical analysis.

Phytochemical Analysis:
The phytochemical substance such as alkaloids, carbohydrates, glycosides, saponins, aminoacids, phenolic compounds, proteins, steroids, terpenoids, flavanoid and tannin were identified by using different solvent extraction.(acetone, methanol, and aqueous) in species of algae *Halimeda tunica, Halimeda macroloba, Halimeda opuntia.*

The phytochemical analysis of Methanol and Aqueous extract *Halimeda macroloba* having a major six primary photochemical compounds such as alkaloids, saponins, phenolic compounds, flavanoid,tannins,aminoacids. Where as other compounds carbohydrates, proteins, glycosides, aminoacids, terpenoids were absent.

The solvent extraction of of Methanol and Aqueous extract of *Halimeda tunica* contain alkaloids, saponins, phenolic compounds, terpenoids, tannin, steroids, tannins were as other phytochemical substance carbohydrates, proteins, glycosides, aminoacids, flavanoid absent.

The extraction of Aqueous *Halimeda opuntia* contain the presence of alkaloids, phenolic compounds, terpenoids, flavanoid, steroids, tannins.

More yield was obtained depending upon the type of solvent which dissolves more particular compounds *Halimeda opuntia* contains a lot of phytochemical substance in Acetone extraction compared to Aqueous solvent.

The phytochemical analysis results of different solvents revealed the presence of various Secondary metabolites were present in seaweeds like alkaloids, phenolic compounds, terpenoids, flavanoid, steroids, tannins.

Flavanoid contain most numerous biological and pharmacological activities. Phenolic compounds were widely used in antioxidant activity. Saponins has enormous amount of antimicrobial, anti inflammatory effects.

Hence the above result shoes that seaweeds contain a lot of secondary metabolites, it contains high medicinal values, it can also be used as antimicrobaial, antioxidant, anti-parasitic, and anti cancer activity. It play a significant role in Drug and Pharmaceutical industry.

Phytochemical analysis of the extract of *Halimeda macroloba, Halimeda tunica, Halimeda opuntia*

S.NO	NAME OF THE TEST	*Halimeda macroloba* Methanol	*Halimeda macroloba* Aqueous	*Halimeda tunica* Methanol	*Halimeda tunica* Aqueous	*Halimeda opuntia* Acetone	*Halimeda opuntia* Aqueous
1	Alkaloids	+	+	+	+	+	+
2	Carbohydrates	-	-	-	-	-	-
3	Glycosides	-	-	-	-	-	-
4	Saponins	+	+	+	+	-	-
5	Proteins	-	-	-	-	-	-
6	Aminoacids	-	+	-	-	-	-
7	Phenolic compounds	+	+	-	+	+	+
8	Trepenoids	-	-	+	-	+	+
9	Steroids	+	+	+	-	+	-
10	Flavanoids	+	+	-	+	+	+
11	Tannin	+	+	+	+	+	+

+

CONCLUSION

The present study revealed that *Halimeda macroloba, Halimeda tunica, Halimeda opuntia* contained noble amount of phytochemical constituents. This bioactive compounds having a lot of potential medicinal values. This contributes to future pharmaceutical industry to obtain lot of bioactive compounds present in the green seaweeds, which can also be used to treat many Diseases.