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Research Article

Preliminary phytochemical analysis of the ethanolic extract of brown Seaweed *Sargassum wightii*

Balachandran P, Anson S Maroky, Ajay Kumar T.V, Parthasarathy V*

Immunology Laboratory, Department of Pharmacy, Annamalai University, Annamalai Nagar-608002, Tamil Nadu, India

ABSTRACT

The big stores for the favorable algae are the Seas and Oceans and the study of seaweeds is called as phycology or ecology. The identification and isolation of new substances are growing from the source of marine organisms. The seaweeds live in salty water and are eukaryotic organisms considered as a good source of bioactive natural products. The present study investigated to explore the phytochemical constituents of the seaweed *Sargassum wightii* (Brown algae). The brown marine algae "*Sargassum wightii*" belongs to the family "Phaeophyceae". Ethanol was used as a solvent system for the preparation of the extract of *Sargassum wightii*. The ethanolic extracts of Sargassum Wightii were undergone to the qualitatively phytochemical test by means of typical measures. Phytochemical analysis shows the presence of alkaloids, tannins, steroids, flavonoids, and carbohydrates, whereas proteins, free amino acids and saponins were found to be absent. The results of the study may lead a foundation for the further studies on this marine algae *Sargassum wightii*.

Keywords: *Sargassum wightii*; seaweed; ethanol; alkaloids; glycosides; tannins and flavonoids.

INTRODUCTION

Nature has turned into the cause for the medicinal agents for about yard of years and exciting amounts of recent drugs have been isolated from natural sources; and most of the isolated bioactive compounds were obtained from the support of the agents found in traditional medicine (Abraham J et al, 2012). Seaweeds are macrophytic marine algae and are particularly wealthy and at various sources of structurally unique natural products. They grow in salty water and are the source of wild and as well as cultivated (Tong-Jen Fu et al, 1999). They are present as a huge amount in shallow coastal waters (Antonisamy, JM et al, 2012). Also, they offer an extensive choice of curative possibilities in both internal and as well as external. Intake of raw and dried seaweeds can give way to many healing benefits. Most of the human illness in humans can be res olved by regularly with the simple addition of seaweeds to their respective diets. Based on epidemiological and natural data, the usage of seaweeds is considered as a significant aspect showing a relatively low breast cancer rates statement in Japan (R. Zakir, 2006). The marine algae and the isolated seaweed have possible benefit to both healthy and improve the food acceptability, also offer exciting potential as a constituent in the expansion of latest foodstuff products (Dhargalkar VK et al, 2005). The seaweeds like *Ulva lactuca, Ulva reticulata, Enteromorpha intestinalis, Acanthophora spicefera, Gracilaria edulis, Padina tetrastomatica and Sargassum wightii* are reported and are highly concentrated in the coastal belt of the Gulf of Mannar, Rameshwaram to Kanyakumari in Tamil Nadu. They are found throughout the year and can be stored for long duration in dry form. Also Seaweeds do not absorb toxic amounts of any element. It is a toxin free and also provides hundreds of organic compounds (Dos Santos SC et al, 2014). Seaweeds are whispered as a spring of bioactive compounds and they have the capacity to produce a huge diversity of derived metabolites characterized by a wide range of biological activities (P. Rajasulochana et al, 2009). The present study was ca r-ried out to provide essential phytochemical screening of the ethanolic extract of *Sargassum wightii*.

MATERIALS AND METHODS

Collection of samples

The brown marine algae "*Sargassum wightii*" was collected from the Bay of Bengal during the month of August, 2014 by hand picking. It belongs to the family "Phaeophyceae" was identified from Mandabam, Rameswaram region of Tamil Nadu, India. The sample was identified and authenticated by Dr. K. Sivakumar, Associate Professor, Department of Botany, Annamalai University, Tamil Nadu, India and the voucher specimen (No. SW/PB/15) was prepared and preserved in the Department of Botany, Annamalai University for future studies and reference.

* Corresponding Author
Email: vapartha@yahoo.com
Contact: +91-9443512724
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Processing of Algae sample

The algae sample which was collected were thoroughly cleaned manually in the sea water to remove the sand particles, epiphytes, pebbles, shells, loosely attached microorganisms and animal waste. The seaweeds were packed in plastic bags to prevent evaporation and taken to the laboratory. The sample of algal was further washed with plenty of tap water and followed by distilled water twice to remove the attached salts. The water was drained off and dried by spread over the blotting paper followed by further drying under the shade for two days and then washing with sterile distilled water to remove the remaining salt on the surface of the algae. This is process avoids pumping of the solvent during the extraction process. The algal samples were placed in the shadow and dried at normal temperature for a period of one week and powered using an electric mixture grinder to get the particles of 40 meshes. The powder sample was packed in airtight plastic bags and laid in a refrigerator.

Preparation of Extract

The ethanol extract of *Sargassum wightii* was extracted using 300gm of the powder sample with 1000ml of ethanol using soxhlet apparatus for about 24 hours at 60^0C. By the distillation process the extract was concentrated to a semisolid consistency. To evaporate the solvent the residue of the extract was kept in a wide mouth beaker at room temperature for 8 hrs. The crude extract consisted of two layers, namely, the upper oil like layer and the lower water soluble layer. Using the separating funnel the upper layer was separated from the lower layer. The upper layer was used for the present study and lower water soluble layer was discarded (Trease G E et al, 1989).

Phytochemical analysis

The various qualitative chemical tests can be performed in finding a profile of a given extract for its bioactive compounds. The prepared extracts using ethanol were analyzed for the occurrence of alkaloids, saponins, tannins, steroids, flavonoids, glycosides, proteins, amino acids and reducing sugars by using the protocols offered in the literature (Sofowora A, 1982).

Test for alkaloids

The ethanolic extract of the *Sargassum wightii* was made to vanish to dehydration by using a boiling water bath. The obtained filtrates were dissolved in 2N Hydrochloric acid. The mixtures were filtered and the scum was separated into three equivalent portions. One part was added with a few drops of Mayer's reagent, one part was mixed with an equal amount of Dragondroff's reagent and the last part was mixed with an equal quantity of Wagner's reagent correspondingly. The emergence of creamish precipitate, the orange precipitate and brown precipitate indicates the presence of individual alkaloids (Trease G E, 1983).

Test for saponins

Mix about 0.5 g of the ethanolic extract of the *Sargassum wightii* extract and add water and forcefully shaken in a test tube and then heated in a boiling water bath to get boil. The Effervescent was observed, which is considered as a preliminary support for the existence of the saponins (Kokate C K et al, 1997).

Test for tannins

Take 0.5 g of *Sargassum wightii* extract and added to a 10 ml of water kept in the test tube and filtered. Added a few drops of 0.1% ferric chloride and observed for brownish green or blue-black coloration. A brownish green color was formed which indicate the presence of tannins.

Test for steroids

To about 2ml of acetic anhydride was added to a 2 ml of ethanolic extract of *Sargassum Wightii* along with 2 ml of sulphuric acid. The change of color from violet to blue or green sample showed the presence of steroids.

Test for flavonoids

With 2 ml of extract was added to 1.5 ml of 50% methanol solution. The solution was warmed and added magnesium metal. In continuation added a few drops of conc. Hydrochloric acid and the red color were formed which showed the presence of flavonoids and the orange color indicated the presence of flavones (Hegde Karunkar et al, 2012).

Test for glycosides

About 0.2 g of the ethanolic extract was added to 1 ml of glacial acetic acid, which containing 1 drops of ferric chloride solution was dissolved. They were kept under layered by adding 1ml of conc. sulphuric acid. At the interface a brown ring was formed indicated the presence of a deoxy sugar characteristic of glycosides.

Test for Proteins

To a 2ml of ethanolic extract, 1ml of 40% NaOH solution was added and the added 2 drops of 1% $CuSO_4$ solution. The presence of a peptide linkage of the molecule was indicated by the violet color which shows the presence of protein.

Test for Amino Acids

To 2 ml of ethanolic extract, 2 ml of Ninhydrin reagent was added and laid in a water bath for about 20 minutes. The visual aspect of purple color formed indicated the presence of amino acids.

Test for Reducing Sugars

To a 2 ml of ethanolic extract, 2 drops of Molisch's reagent were added and shaken well. About 2ml of conc. Sulphuric acid was added drop wise along the sides of the test tube. A reddish violet ring formed at the connection of two layers which indicated the presence of

Table 1: The phytochemical constituents of ethanolic extract of Sargassum wightii.

S.No	Phytoconstituents	Ethanolic Extract
1	Alkaloids	++
2	Saponins	--
3	Tannins	++
4	Steroids	++
5	Flavonoids	++
6	Glycosides	++
7	Proteins	--
8	Amino acids	--
9	Carbohydrates	++

reducing sugar or carbohydrates (Siddiqui A A et al, 1997).

RESULTS AND DISCUSSION

The preliminary phytochemical analyses make known that alkaloids, tannins, steroids, flavonoids, glycosides and carbohydrates are present in the ethanolic extract of *Sargassum wightii* as shown in the table. 1. From the ethno-medicinal data it is clear that this seaweed *Sargasssum wightii* may have a lot of potential chemical constituents. The obtained results can be used as an initial step for further identification of bioactive compounds from the ethanolic extract of seaweed *Sargassum wightii*.

CONCLUSION

The present work is under further process in obtaining the fractions using column chromatography. The fractions are further to be analyzed using GCMS and LCMS studies to identify and isolate the novel compounds. This is expected that these bioactive compounds may show the way to the preparation of new and more powerful remedies and that may prove useful in the management of a variety of illness and diseases.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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