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Research Article

PRELIMINARY PHYTOCHEMICAL SCREENING AND INVITRO ANGIOTENSION ACTIVITY OF BIOACTIVE COMPOUND - STEROID ISOLATED FROM SARGASSUM ILICIFOLIUM

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ABSTRACT

Objective: Seaweeds are one of the important constituents of the primary producers and contribute substantially to the carbon budget of the coastal ecosystem which is rich in antioxidant such as Carotenoid, pigments, polyphenols, enzymes and diverse functional polysaccharide. Hypertension is a major risk factor for stroke, myocardial infarction (heart attacks), heart failure, aneurysms of the arteries (e.g. aortic aneurysm), peripheral arterial disease.

Materials and Methods: The steroid compound from *Sargassum ilicifolium* was isolated from the seaweed and to evaluate the ACE inhibitory activity by Spectrophotometeric method.

Results: The seaweed contains four bio active compounds such as alkaloids, Carbohydrate, Sterol and tannin were present in the aqueous extract of *Sargassum ilicifolium*. Steroid is auto lysate the compound (50 µg) was able to inhibit near 16.8 % of ACE activity, whereas the control has 2.5 % of ACE activity.

Conclusion: During this study the Fatty compound steroid was isolated from the seaweed *Sargassum ilicifolium* is used for the antihypertensive activity which is used to treat for Hypertension.

Keywords: Seaweed, *Sargassum ilicifolium,* Hypertension, ACE inhibition and anti-hypertensive.

INTRODUCTION

Hypertension (HTN) or high blood pressure, sometimes called arterial hypertension, is a chronic medical condition in which the blood pressure in the arteries is elevated. This requires the heart to work harder than normal to circulate blood through the blood vessels. The application of seaweed antioxidants in foods, food supplements, nutraceutical supplements and medicine is considered from the perspective of benefits to human health. Economic utilization of seaweeds for their ACE inhibitory properties remains in its infancy.

Seaweeds are one of the important constituents of the primary producers and contribute substantially to the carbon budget of the coastal ecosystem. The phycocolloid align in all brown algae, carrageenan and agar in many red algae aggressively trap metallic ions.

The seaweeds containing such compound are able to remove heavy metals from food and body, excrete metals out in the stool. Seaweeds have been used by human as medicine and food for at least 13,000 years. Seaweeds are rich in antioxidant such as Carotenoid, pigments, polyphenols, enzymes and diverse functional polysaccharide [1].

Description of the experimental plant

Scientific classification of *Sargassum ilicifolium* (Agardh 1820)

Kingdom: Chromista
Phylum: Ochrophyta
Class: Phaeophyceae
Order: Fucales
Family: Sargassaceae
Genus: *Sargassum*
Species: *ilicifolium*

Fronds 40-65 cm high, brown. Holdfast a plate like disc, 1-1.4cm wide. Main axis terete, 2-4 cm long, 1.5 mm in diameter, bearing several primary branches. Primary branches sub cylindrical, slightly compressed, with smooth surface, 1 mm in diameter. Secondary branches similar to primary branches, racemose arranged, generally 5-15 cm long. Branchlet giving rise from secondary branches, shorter and more slender than secondary branches, 1.5-2.5 cm long.

Uses

Sargassum seaweeds are eaten by people, and used fish bait in basket traps, animal feed, fertilizer and insect repellent. Various species are used as medicine for ailments ranging from children's fever, cholesterol problems, cleansing the blood, skin ailments. In the tropics, sargassum seaweeds are a significant source of alginates. They are also used as a component in animal feed and liquid plant food or plant biostimulants [2-3].

MATERIALS AND METHODS

Collection of Samples

The dried algae of *Sargassum ilicifolium* was collected in Central Marine Research Institute of Rameshwaram at Ramanathapuram District in Tamil Nadu.

Extraction

Take 10 gram of *Sargassum ilicifolium* powder was placed in 100ml of distilled water and allowed for boiling at 100°C for 30 minutes. The extract was filter through a sterile funnel containing sterile Whatmann No.1 filter paper then filtered and stored in brown bottle at 4°C.

Preliminary Phytochemical Screening

The phytochemicals in the seaweed extract of *Sargassum ilicifolium* was allowed for qualitative analysis of phytochemical compounds. Chemical test were carried out on the aqueous extract using standard procedures to identify the phytoconstituents such as, Alkaloid, Carbohydrate and Glycosides, Flavonoids, Saponins, Coumarins, Steroids,

Tannins, Chlorogenic acid and Anthocyanin as described by Harbone method[4].

Isolation of Steroid by Column Chromatography

The extract was subjected to column chromatography for the isolation of steroid compound over silica gel 100-200 mesh. The column was eluted with solvents of increasing order of polarity. The fractions were collected in 25ml each and allowed to evaporate to get the residue.

Determination of ACE Inhibitory Activity

ACE inhibitory activity was determined according to the methods of Cushman and Cheung[2] with slight modifications. For each assay, 50 µl of sample solution with 50 µl of ACE solution (25 mu/ml) were pre-incubated at 37^{0}C for 10 min,

After which the mixture was subsequently incubated with 100 µl of substrate (25 mM Hipuril-His-Leu in 50 mM sodium borate buffer containing 500 mM NaCl at pH 8.3) at the same temperature for 60 min.

The reaction was terminated by adding 250 µl of 1 M HCl. After that, the resulting hippuric acid was extracted with 500 µl of ethyl acetate. After centrifugation at 4,000 rpm for 10 min, 200 µl of the supernatant was transferred into a glass tube and dried in a dry oven at 80^{0}C for one hour.

The residue was dissolved in 1 ml of distilled water, and the absorbance was measured at 228nm using a spectrophotometer. The extent of inhibition was calculated [5].

RESULTS AND DISCUSSION

In this present investigation *Sargassum ilicifolium* was used for the Screening of Phytochemical analysis and ACE inhibitory activity. The results are tabulated and discussed here.

Phytochemical screening and Isolation of steroid

The present study carried out the phytochemical compound was screened in aqueous extract of ***Sargassum ilicifolium*** by qualitative method. In the phytochemical analysis, four bio active compounds such as alkaloids, Carbohydrate, Sterol and tannin which are presents in the ***Sargassum ilicifolium*** aqueous extract. The phytochemical active compounds of selected seaweed were qualitatively analyzed and the results are recorded in (Table-1).

ACE Inhibitory Activity

The steroid compound is allowed to purification process which is carried out the following study such as Anti-hypertensive activity by ACE Method. The result are tabulated in table 2

Table 1: Preliminary phytochemicals analysis of *Sargassum ilicifolium*

S. No.	Phytochemical constituents	Name of the Test	Result
1.		a)Mayer test	**+**
	Alkaloids	b)Dragondroff test	+
		c) Wagner test	+
2.	Carbohydrates and Glycosides	a) Molish's test	**+**
		b) Fehling's test	+
		c) Benedict's test	-
3.	Phytosteroids	a)Libermann's Burchard test	**-**
		b) salkowaski test	+
		c) saponin	-
		Glycosides	
4.	Tannins	Lead Acetate	**+**
5.	Pseudo tannins	Ferric chloride	**+**
6.	Cholorogenic	Ammonia	**-**

+ Present: - Absent

Table 2: *Invitro* Angiotension inhibitory activity of Seaweed *Sargassum ilicifolium*

S. No.	Name of Test	Values in %
1	Control	2.5
2	Steriod	16.8

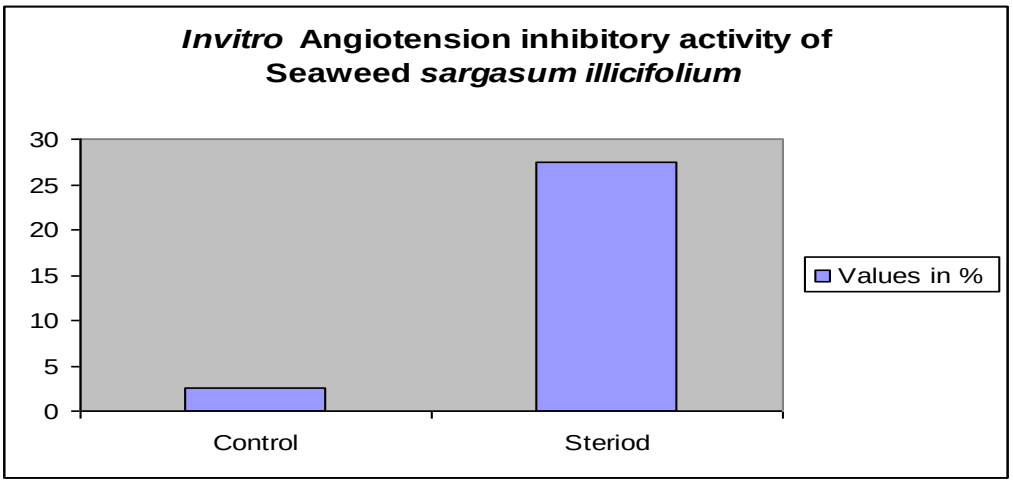

Fig. 1: *Invitro* Angiotension inhibitory activity of Seaweed *Sargassum ilicifolium*

The present investigation has been undertaken to find out the presence of steroid compound in the selected seaweed was confirmed by Column Chromatography. Using column chromatography Steroid was purified from seaweed and it was used for ACE inhibitory Activity. The purified steroid was once again confirmed by Preliminary analysis.

Alkaloids are important defence of the plant against pathogenic organism and herbivores. It also toxin for insects which further modify the alkaloids and incorporate them into their own defence secretion. The tannins are an important ingredient in the process of tanning leather. Oak bark, mimosa and quebracho tree have traditionally been the primary source of tannery tannin, though inorganic tanning agents are also in use today and account for 90% of the world's leather production.

The evaluation of anti-hypertensive activities of whole hydro lysate and fractions were also tested, monitoring the ability of the fraction to inhibit the activity of angiotension compound involved in hypertension regulation. Steroid is auto lysate the compound (50 µg) was able to inhibit near 16.8 % of ACE activity, whereas the control is 2.5 % of ACE activity. In this present investigation the isolated compound having the maximum inhibitory activity which is used to anti hypertensive compound.

In this present investigation the Hypertension activity was observed in maximum level of isolated compound when compared with normal and the same activity was observed [6].

The hypertensive activity of five beta-adrenoceptor antagonists with different ancillary pharmacological properties was compared in a randomized double-blind factorial trial in 25 untreated patients with stable, uncomplicated essential hypertension. A novel ACE-inhibitory active peptide, Tyr-Ile may have potential for use in antihypertensive therapy [7].

CONCLUSION

In this present Investigation Phytochemical and ACE inhibitory activity was analyzed in selected sea weed *Sargassum ilicifolium*.. During this study the Fatty compound steroid was isolated from the seaweed *Sargassum ilicifolium* is used for the antihypertensive activity which is used to treat for Hypertension.

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