Antioxidant Potential from Padina Gymnospora Seaweed Methanolic Extracts

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ABSTRACT

Introduction: Seaweeds are associated with antibacterial, antinociceptive effects. The interest in marine organic entities is a potential and promising source in marine life in the last few years. Seaweeds have been used as food in Asian countries for centuries. Seaweeds possess valuable medicinal properties that exhibit anti adhesive activities. The dietary profile of Padina gymnospora incorporates a lot of carbohydrates, lipids, proteins and other minerals alongside it includes cell reinforcements like nutrients and other vitamins (C and E).

Aim: The main objective of the study is to evaluate antioxidant activity of Padina gymnospora seaweed methanolic extracts.

Materials And Methods: Sample collection of Padina gymnospora is collected and washed with distilled water, dried and powdered. Methanol ice extract preparation is done from crude oil extract and again converted into powder then it’s been dried for a few hours. Further antioxidant assay (DPPH, H2O2, total antioxidant assay) is done to find the bio active compounds from crude extract.

Result: The antioxidant property of seaweed Padina gymnospora was evaluated and it was found to increase with increasing concentration of extract. Similar results were observed in the present study with antioxidant activity increasing in the lower concentrations. In the present investigation methanolic extracts showed antioxidant activity for all the samples analysed.

Conclusion: This study indicated that the methanol extract of seaweed had better antioxidant effects in comparison with other previous studies.

INTRODUCTION

Seaweeds are associated with antibacterial, antinociceptive effects. The interest in marine organic entities are a potential and promising source in marine life in the last few years. Seaweeds possess various valuable medicinal values that exhibit anti adhesive and pharmacological activities (Corsetto et al., 2020; Liu and Sun, 2020; Wang et al., 2020). In recent years, the interest in marine organisms has increased due to their potential as natural antioxidants and cytotoxic properties that helps in preventing various cardiovascular diseases such as myocardial infarction, high blood pressure and coronary heart disease. Some important Sea weeds like Eckhnia cava and Hizikia fusiforms also exhibit potent antioxidant activity. High amounts of polyphenols are found in marine plants like seaweeds. Although seaweeds have wide application in the pharmaceutical industry, many of its uses are unknown. Seaweeds are basically classified as Rhodophyta, chlorophyta and phaeophyta. Cell reinforcement substances which ramage the resolutions assure a significant part in the counter action of free extremist incited infections. By denoting hydrogen radicals the essential revolutionaries are decreased to non-

KEYWORDS:
Padina gymnospora, antioxidant activity, methanolic extracts, DPPH assay

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Padina gymnospora is portrayed by the thick thallus, the circulation of sporangia basically on the prevalent surface of the thallus and transient indusium. The dietary profile of Padina gymnospora incorporates a lot of carboxydrates, lipids proteins and other minerals alongside it includes cell reinforcements like nutrients and other vitamins( C and E). Cancer prevention agent exercises of different Dissolvable concentrates of Padina gymnospora where evaluated by the DPPH assay. Previous studies showed that the Padina gymnospora has got a maximum protein content of 17.08%(N et al., no date; Singh et al., 2015; Shanmuganathan et al., 2019; Vasantharaja et al., 2019). Many research studies estimated that Padina gymnospora has high levels of amino acid. Free radicals are the cause of ageing and a variety of human diseases(N et al., no date; Karez and Pereira, 1995; Filho et al., 1996; Chapman, 2012; ‘Influence of Seaweed Extracts (Sargassum dentifolium or Padina gymnospora) on the Growth and Physiological Activities of Faba Bean and Wheat Plants Under Salt Stress’, 2013; JASAndrade et al., 2015). In accordance with previous studies, Antioxidant compounds that scavenge free radicals play a significant role in the prevention of free radical-induced diseases. The primary radicals are reduced to nonradical chemical compounds and then converted to oxidize antioxidant radicals by donating hydrogen radicals to maintain stability in the body. This helps in protecting our health from various degenerative diseases(Rajendran et al., 2019)(Ashok, Ajith and Sivanesan, 2017)(Malli et al., 2019)(Mohan and Jagannathan, 2014)(Menon et al., 2018)(Samuel, Acharya and Rao, 2020)(Praveen et al., 2001)(Neelakantan et al., 2011)('Oligonucleotide therapy: An emerging focus area for drug delivery in chronic inflammatory respiratory diseases’, 2019)(Kumar et al., 2006). Tetrasporic plants are typically larger than oogonal and antheridial plants in the Mandapam region. Padina gymnospora is ecologically and biologically significant in Indian subcontinent. Padina gymnospora cultures provide food, development, and a hospitable atmosphere for other living things. Padina gymnospora is one of the most important species in preserving ecosystem equilibrium because of these characteristics. Padina gymnospora is a brown alga that has been investigated for its ecological importance and biochemical properties, including its high capacity to accumulate heavy metals. The fucans are thought to be one of the major polysaccharides involved in metal binding and precipitation in cell walls. Coastal waters have a lot of dissolved organic matter, and marine algae are a big source of it. Carbohydrates, polysaccharides, nitrogenous, and polyphenol products are various organic materials found in the seaweeds. A popular antioxidant assay is based on the scavenging of DPPH free radicals. For this assay, a variety of procedures have been used, resulting in a wide range of findings in the laboratory. In this study, DPPH assay has been used to determine the antioxidant levels of seaweed Padina gymnospora(Ramadan-Hassanien, 2008; Sirivibulkovit, Nouanthavong and Sameenoi, 2018; Vasantharaja et al., 2019; Chen, Liang and Han, 2020). Various studies has been conducted in our institution to develop knowledge on research activity(Pitchich Sivaperumal, Kamala and Rajaram, 2018; P. Sivaperumal, Kamala and Rajaram, 2018; Kamala et al., 2019; Jaisankar and Arivarasu, 2020; Karthik, Arivarasu and Rajeshkumar, 2020; Priya et al., 2020; Shankar et al., 2020; Shree et al., 2020). The main objective of this study is to evaluate the antioxidant activities of Padina the most Pura from the south coastal area of Tamil Nadu, India.

MATERIALS AND METHODS

Sample collection and preparation

The fresh seaweed sample Padina gymnospora was collected from Gulf of Mannar biosphere reserve, Tamilnadu. The samples were washed thoroughly with tap water then shade dried on table tissue paper for 4 weeks and ground into a fine power.

Preparation of extraction

10g of dried powdered seaweed sample was mixed with 100ml of methanol and incubated for 24 hours at ambient temperature. Then the mixture was filtered through whatman filter paper (No.4), the filtrate was centrifuged at 3000rpm for 10min, followed by filtration through 0.45µm syringe micro filter. At last, the solvent was evaporated via a vacuum rotary evaporator to obtain dried powder. Then the samples were stored in an aluminum container at 4ºC for further analysis.

Total antioxidant activity

Total antioxidant activity of the crude seaweed was determined by following method: 0.3 ml of sample was prepared in different concentrations (0.5 - 3mg/ml) with 3ml of reagent solution (0.6 M sulfuric acid, 28mM sodium phosphate and 4mM ammonium molybdate). Reaction mixture was incubated at 95ºC for 90 minutes in a water bath. Absorbance of all sample mixtures was measured at 695 nm. Total antioxidant activity has been expressed as the number of equivalents of ascorbic acid.

DPPH Assay

The antioxidant potential of seaweed extract was determined on the basis of their scavenging activity of the stable 1,1-diphenyl-2-picryl hydrazyl (DPPH) free radical. Different concentrations (0.5-3mg/ml) of samples were mixed with 2.9ml diphenylpicrylhydrazyl (DPPH) solution (120µM) in methanol and incubated in darkness at 37°C for 30 minutes. The absorbance was recorded at 517 nm. Inhibition of free radical by DPPH in percentage (I %) was calculated with the following equation:

\[
\text{Percentage of Inhibition (I %) } = \frac{(\text{A blank} - \text{A sample})}{\text{A blank}} \times 100
\]
Where, \( A_\text{blank} \) is the absorbance of the control reaction and \( A_\text{sample} \) is the absorbance of the test compound. The values of inhibition were calculated for the various concentrations of the sample. Ascorbic acid was used as positive control (Kamala et al., 2015) and all the tests were carried out in triplicate.

**Scavenging of hydrogen peroxide (H\(_2\)O\(_2\))**

Ability of the crude extract from the seaweed to scavenge H\(_2\)O\(_2\) was determined by following method: 40mM H\(_2\)O\(_2\) was prepared and the concentration was determined spectrophotometrically by measuring the absorption with the extraction coefficient for H\(_2\)O\(_2\) of 81 M\(^{-1}\) cm\(^{-1}\). Seaweed extract and the standard ascorbic acid (0.5, 1, 1.5, 2, 2.5 & 3 mg/ml) were added to 0.6 ml of 40mM H\(_2\)O\(_2\) solution and the absorbance of H\(_2\)O\(_2\) was determined at 230 nm after 10 min incubation against a blank solution, containing phosphate buffer without hydrogen peroxide. The percentage of scavenging of H\(_2\)O\(_2\) was calculated as follows:

\[
\text{Scavenging effect (\%) } = \frac{(A_\text{cont} - A_\text{test})}{A_\text{cont}} \times 100
\]

**RESULTS AND DISCUSSION**

![Total Antioxidant Equivalent Activity](chart1.png)

**Fig.1:** Bar graph represents the total antioxidant activity equivalent of Ascorbic acid. The X axis represents the various concentration levels in µg/ml and Y axis represents the total antioxidant activity, data implies as mean±SEM.

![DPPH Activity of P. gymnospora seaweed methanolic extract.](chart2.png)

**Fig.2:** Bar graph represents the antioxidant effect of the P. gymnospora seaweed methanolic extract. The X-axis represents various concentration levels and the Y-axis represents the percentage of the DPPH inhibition, data implies as mean±SEM.
In our study done by DPPH assay using methanolic extract of Padina gymnospora seaweed. There was a significant difference seen in antioxidant potential of herbal extract when compared to that of the standard. Contradicting results were observed in a previous in vitro study done by Leelavathi et al. The study showed ulva lactuca exhibited the highest levels of antioxidant and free radicals scavenging activities in petroleum ether extracts when compared to other seaweed samples. The limitations of the study include only antioxidant activity whereas antimicrobial, cytotoxicity, anti-inflammatory properties have not been studied. Bioactive compounds responsible for the antioxidant property have not been studied. Further scope of research includes formulations of the herbal extract into gel or mouthwash formulation and clinical trial evaluations could be done. (Pushpaanjali, Geetha and Lakshmi, 2020) (Aathira, Geetha and Lakshmi, 2020) (Baskar and Lakshmi, 2020) (Manya Suresh, 2020) (First Report on Marine Actinobacterial Diversity around Madras Atomic Power Station (MAPS), India, no date) (Physicochemical Profile of Acacia Catechu Bark Extract - An in Vitro Stud - International Journal of Pharmaceutical and Phytopharmacological Research, no date) (Lakshmi, 2021) (Awareness of Drug Abuse among Teenagers - International Journal of Pharmaceutical and Phytopharmacological Research, no date) (Mangal, Anitha and Lakshmi, 2018) (COX2 Inhibitory Activity of Abutilon Indicum - Pharmaceutical Research and Allied Sciences, no date) (Jibu, Geetha and Lakshmi, 2020) (Sindhu et al., 2020) (Nivethitha et al., 2020) (Mariona, Roy and Lakshmi, 2020)

At variant concentration levels, the herbal extract was compared with the standard. At 25µg/ml, Padina Gymnospora extract showed the TAA equivalent to Ascorbic Acid standard (31.00). At 50µg/ml, Padina gymnospora showed the TAA equivalent to Ascorbic Acid standard (60.00)). At 75µg/ml, Padina gymnospora showed the TAA equivalent to Ascorbic Acid standard (85.00). At 100µg/ml, Padina gymnospora showed the TAA equivalent to Ascorbic Acid standard (95.00). At 125µg/ml, Padina gymnospora showed the TAA equivalent to Ascorbic Acid standard (120.00). At 150µg/ml, Padina gymnospora showed the TAA equivalent to Ascorbic Acid standard (135.00). At 25µg/ml, DPPH showed less than 10% of inhibition whereas standard showed 40% of inhibition. At 50µg/ml concentration, DPPH showed 30% of inhibition whereas standard (ascorbic acid) showed 60% of inhibition. 80% and 60% of inhibition were seen in ascorbic acid and DPPH organic compounds in 75µg/ml. At 100g/ml concentration, DPPH showed 80% of inhibition whereas standard (ascorbic acid) exhibited 40% of inhibition. At 125µg/ml concentration level, DPPH showed nearly 60% of inhibition whereas standard (ascorbic acid) exhibited 85% of inhibition. At 150µg/ml concentration level, DPPH showed 90% of zone of inhibition whereas standard (ascorbic acid) exhibited 100% of zone of inhibition. % of Hydrogen peroxide inhibition at 25µg/ml showed a significant difference between hydrogen peroxide and standard. At 50µg/ml concentration level, H2O2 showed 20% of inhibition whereas standard (ascorbic acid) exhibited 50% of inhibition. At 75µg/ml concentration level, H2O2 showed 30% of inhibition whereas standard (ascorbic acid) exhibited 65% of inhibition. % of Hydrogen peroxide inhibition at 100g/ml has also shown a significant difference between hydrogen peroxide and standard (ascorbic acid). At 125µg/ml concentration level, H2O2 showed 45% of inhibition whereas standard (ascorbic acid) exhibited 85% of inhibition. At 150µg/ml concentration level, H2O2 showed 60% of inhibition whereas standard (ascorbic acid) exhibited 100% of inhibition.
**CONCLUSION**

Within the limitations of the study, antioxidant properties of various concentrations of methanolic extract of Padina gymnospora seaweed was studied. The result suggests this herbal extract as a good anti oxidant agent which requires further invivo studies.

**CONFLICT OF INTEREST**

There was no conflict of interest

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