ijlpr 2022; doi 10.22376/ijpbs/lpr.2022.12.6.L155-160

International Journal of Life science and Pharma Research

ISSN 2250-0480

Research Article

Antioxidant activity of Padina Gymnospora and Ulva Lactuca

Study On Phytochemical and Antioxidant Properties of Padina gymnospora and Ulva lactuca

N. Praba[1*] and Dr. S. Sumaya[2]

[1]Ph.D. Research Scholar, Department of Home Science and Research Centre Thassim Beevi Abdul Kader College for Women (Autonomous)*

[2]Head, Department of Home Science and Research Centre, Thassim Beevi Abdul Kader College for Women (Autonomous), Kilakarai, Ramanathapuram District, India

Abstract: Seaweed constitutes one of the commercially important living renewable resources in the marine ecosystem.Seaweeds have antimicrobial, antioxidant, and anti-inflammatory properties. The aim of the study is to estimate the phytochemicals and antioxidant activity of *Padina gymnospora* and *Ulva lactuca*. The antioxidant properties and phytochemical analysis of *Padina gymnospora* and *Ulva lactuca* collected from the Gulf of Mannar during the low tide period were analyzed.The study's objective is to analyze the phytochemical and antioxidant activities using methanol as solvent. Antioxidant activities were determined using 2,2-diphenyl-1-picryl-hydrazyl (DPPH) assay treated with methanol and Phytochemical analysis carried out by Folin-Ciocalteu reagent. Antioxidants and phytochemicals in seaweeds provide health-promoting effects and protect against non-communicable diseases such as Cancer, arthritis, diabetes, obesity, etc. The results reveal that antioxidant activities were higher in *Padina gymnospora* in methanol solvent than in *Ulva lactuca*. The percentage of inhibition of *Padina gymnospora* and *Ulva lactuca* was 61.5 and 64.5 respectively. IC50 value of *Padina gymnospora* and *Ulva lactuca* was 60.4 and 60.6 respectively.The preliminary phytochemical analysis of *Padina gymnospora* and *Ulva lactuca* was found to contain Alkaloids, Steroids, Terpenoids, Glycosides. To know the efficiency of green seaweed *Ulva* lactuca with brown seaweed *Padina* gymnospora the study was carried out. The highest flavonoid content was recorded in brown seaweed *Padina gymnospora* (25.5±0.02mg REg^{-1}) compared to*Ulva lactuca* (19.32±0.5mg REg^{-1}). The total phenolic content was highest in *Padina gymnospora* (3.7±0.06mg GAEg-1) whereas *Ulva lactuca* contained (2.81±0.3mg $GAEg^{-1}$). Therefore, the present work recommends consuming seaweeds in our regular diets to have a healthy lifestyle. From the current study of phytochemical and antioxidant rich seaweeds can be incorporated in the formulation of drugs used for treating various non- communicable diseases prevailing in the present era.

Keywords: Padina gymnospora, Ulva lactuca, Phytochemical, Antioxidants and Methanol

***Corresponding Author**

N. Praba , Ph.D. Research Scholar, Department of Home Science and Research Centre Thassim Beevi Abdul Kader College for Women (Autonomous).

Received On 21 July, 2022

Revised On 30 September, 2022

Accepted On 7 October, 2022

Published On 1 November, 2022

Funding This research did not receive any specific grant from any funding agencies in the public, commercial or not for profit sectors.

Citation N. Praba and Dr. S. Sumaya , Study On Phytochemical and Antioxidant Properties of Padina gymnospora and Ulva lactuca.(2022).Int. J. Life Sci. Pharma Res.12(6), L155-160 http://dx.doi.org/10.22376/ijpbs/lpr.2022.12.6.L155-160

1. INTRODUCTION

The Gulf of Mannar Biosphere Reserve (GOMBR) is a rich biodiversity area, as it possesses various natural resources such as corals, seaweeds, pearl oysters, turtles, crabs, lobsters, shrimps and fishes. The marine ecosystem consists of two major types of algae: the macroalgae(Seaweeds) and the microalgae (phytoplankton). Seaweeds are commercially important renewable marine living resources. A large variety of red and green seaweeds is found in subtropical and tropical waters, while brown seaweeds are widely common in cooler and temperate waters. Seaweeds are emerging tremendously in the biomedical field due to the presence of potent pharmacological substances with vast arrays of potential health benefits. The marine environment is described as a source of new chemical variety for drug discovery because many bioactive substances are isolated from marine organisms[1]. The presence of Higher levels of alkaloids, saponins, glycosides, tannins, terpenoids, steroids, flavonoids, and phenols are the phytochemicals found in the seaweeds are extensively used in pharma industries[2]. Polyphenols compounds are reported to have microbicidal activity against many pathogenic bacteria. The phytoconstituents of the seaweeds possess a good antioxidant potential and other important biogenic activities[3]. Bioactive compounds from seaweeds are medicinally used as healing agents in inflammation, burns, piles and as antidotes[4]. Seaweeds are richest producers of secondary metabolites compared to terrestrial plants [5]. Their phenolic compounds are one of the most effective antioxidants in marine algae[6]. These Seaweeds are a potential source of natural antioxidants[7]. Free radicals are produced during the metabolism of the mitochondria, inflammatory actions, phagocytosis, and physical exercises. Extrinsic factors like smoking, pesticides, and radiation accelerate the free radicals inside the cells and cause various stress-related issues[8]. Uninterrupted production of excess free radicals injures the biological materials such as the macro and micronutrients that lead to an imbalance of a healthy lifestyle. Therefore, it is necessary to protect the biological system from damage [9]. Oxidative stress is an important factor in treating diseases like cancer, acquired immune deficiency syndrome, arthritis, diabetes, obesity[10] are cured by regular consumption of seaweeds.The constituents of the seaweed possess good antioxidant potential and other important activities. The marine seaweeds have several chemical constituents of high therapeutic efficacy and they are producers of secondary metabolites compared to land plants used in the pharmaceutical industry. The compounds present in the seaweeds exhibit antibacterial, antimicrobial, antiviral and anti-inflammatory activities. Seaweeds possess medicinal properties and are useful in developing new medicines[11]. The multifaceted seaweed is used as food, fodder for animals, fertilizers, in paper and tyre industries. The present study is focused on brown and green seaweed- *Padina gymnospora* and *Ulva lactuca* respectively. *Padina gymnospora* belongs to the *Order-Dicyotales,* family *Phaeophyceae, Class- Phaeoyophyta. Padina gymnospora* the brown algae are a dark brown coloured, thalli fan shaped, greenish brown in upper portion and brown in the basal portion with coarse and leathery texture attached to the corals. *Ulva lactuca* belongs to *Order-Ulotrichales, Family –Ulvaceae, Class- Chlorophyta. Ulva lactuca* the green algae known as sea lettuce to establish the efficiency and the efficacy of the phytochemicals present, nutraceuticals, secondary metabolites available with *Ulva lactuca* in comparison with already known studies of *Padina gymnospora.,* the present study is to investigate the phytochemicals and antioxidant capacity of *Padina gymnospora* and *Ulva lactuca* edible seaweeds from the Gulf of Mannar coast was carried out.

2. MATERIALS AND METHODS

2.1 Collection of Seaweeds

Fresh seaweeds were collected in polythene bags containing seawater from the coastal line of Mandapam, Ramanathapuram district, India and transported to the laboratory. The algal species was identified at Central Marine Fisheries Research Institute, Mandapam, India. Then the seaweeds were thoroughly washed in running tap water to remove the marine debris. It was then washed with distilled water and allowed to dry under shade for 5 days, until the moisture contents are completely removed. The dried seaweed was ground into a coarse powder using an electrical mixie and stored in airtight containers at room temperature for further extraction method followed by Cyril *et al.*2017[12].

2.2 Identification

The study species *Padina gymnospora* and *Ulva lactuca* was recognized by Dr. P. Anantharaman, Professor, Department of Marine Science, CAS, Annamalai University, Parangipettai. The voucher specimen of the selected seaweed *Padinagymnospora*(IPA91012) and *Ulva lactuca*(IPA91014) have been deposited at Instituto Agronomico de Pernambuco Herbarium (IPA).

2.3 Preparation of Crude Extract

Finely ground seaweed powder extracted with methanol in the ratio of 1:10 (w/v) in a conical flask for 24 hours. The mixture was filtered in a separate container. The process was replicated two times with the same residue using fresh solvent. All the supernatant was collected together and the solvent was removed by rotary evaporator. The filtrate thus obtained was stored at 4°C for further studies.

2.4 Preliminary Phytochemical Screening

The phytochemicals such as alkaloids, steroids, saponin, terpenoids, glycosides, tannins present in the selected seaweeds of *Padina gymnospora* and *Ulva lactuca* was carried out using the standard method [13, -14]

2.5 Determination of Total Phenolic Compounds and Flavonoid Content

Total phenolic compounds (TPC) of seaweed extracts were determined by Folin-Ciocalteu reagent with slight modification of method followed by Siddhuraju P *et al.* (2007)[15]20µl of extracts were mixed with 100µµl of 1:10 Folin-Ciocalteu reagent followed by the addition of Na_2CO_3 (80µµl). After incubation at room temperature for 2 hours in the dark, the absorbance was recorded at 765 nm. Gallic acid was used as the standard reference. TPC was expressed as mg gallic acid equivalents per gram of dried extract (mg GAE g^{-1}) (gallic acid equivalent). Determination of the total phenols was carried out in triplicate, and the results were mean values of the triplicates. Flavonoid content (FC) (flavonoid content) of each extract was determined by colorimetric method described by Brighentee*t al.* (2007)[16]. 20µµl of each extract was separately mixed with 20µµl of 10 % aluminium chloride, 20µµl of 1 M potassium acetate and 180µµl of distilled water, and incubated at room temperature for 30 min. The absorbance of the

reaction was recorded at 415 nm. Flavonoid content was expressed as mg of rutin equivalents per gram of dried extract (mg RE g^{-1}) (rutin equivalent). Determination of the flavonoids was carried out in triplicate, and the results were mean values of the triplicates.

2.6 DPPH Radical Scavenging Assay

DPPH radical scavenging activity was estimated according to the method of Zhang *et al.* (2007) with slight modifications[17].The antioxidant activity of the methanol extract of *Padina gymnospora* and *Ulva lactuca* was measured on the basis of the scavenging activity of the stable 1, 1-diphenyl 2-picrylhydrazyl (DPPH) free radical. One mL of 0.1 mM DPPH solution in methanol was mixed with 1 mL of various concentrations (10 - 60 µµg/mL) of the extract. The mixture was then allowed to stand for 30 min incubation in the dark. Ascorbic acid was used as the reference standard. 1 mL of methanol and 1 mL DPPH solution were used as the control. The decrease in absorbance was measured using UV-Vis Spectrophotometer at 517 nm. The percentage of inhibition was calculated using the following formula:

Percentage of DPPH radical inhibition = Control – Sample x 100 /*Control*

3. STATISTICAL ANALYSIS

All the experiments were conducted in triplicates and data given in tables were average of the three replicates. All data were reported as mean triplicates.

4. RESULTS

4.1 Phytochemical Analysis

The phytochemicals of the methanol extract of *Padina gymnospora and Ulva lactuca* showed the presence of Alkaloids, Steroids, Glycosides, Saponins, Tannin,Terpenoids,,flavonoids. The presence of phytochemicals in the selected seaweeds are the secondary metabolites known for its medicinal effects in treating various defects in the body caused by the intrinsic and extrinsic factors. Regular consumption of the natural marine seaweeds can cure many vulnerable diseases in the near future.

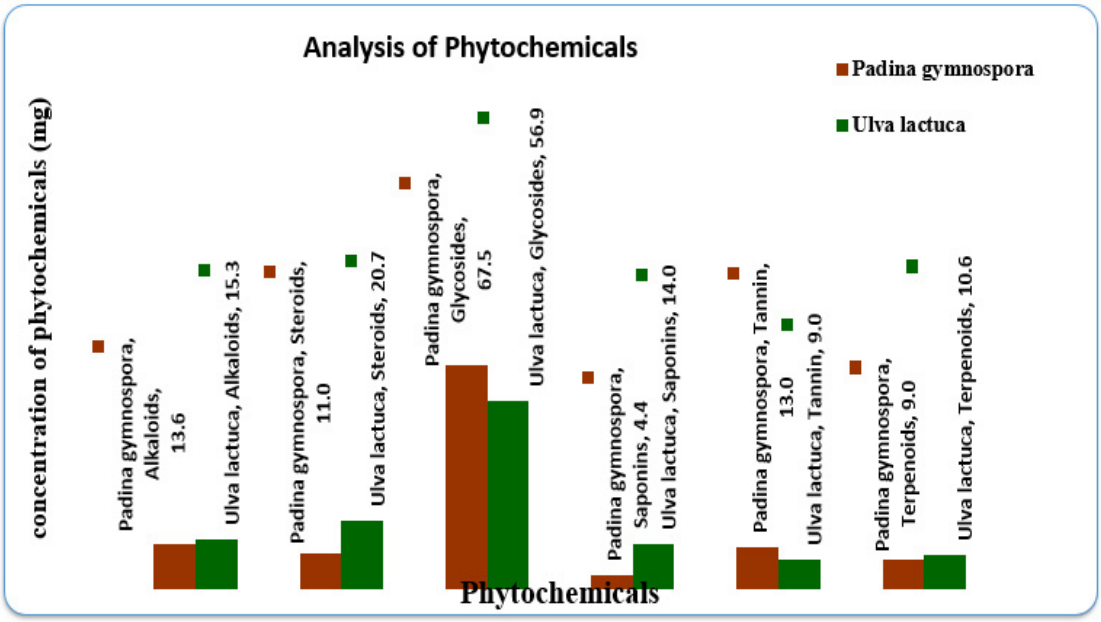

Fig 1: represents the Phytochemicals present in the seaweeds

The presence of flavonoids plays a major role in the human biological system. The flavonoids are found to possess anticancer, antiviral, antioxidant and anti-inflammatory properties. As part of a balanced diet, seaweeds should be consumed in our daily diets. The highest flavonoid content was recorded in brown seaweed *Padina gymnospora* (25.5±0.02mg REg^{-1}) compared to *Ulva lactuca* (19.32±0.5mg REg^{-1}). The presence of phenols is higher in *Padina gymnospora* than in *Ulva lactuca.* The total phenolic content was highest in *Padina gymnospora* (3.7±0.06mg $GAEg^{-1}$) whereas *Ulva lactuca* contained (2.81±0.3mg GAE g^{-1}) . The phytochemicals depend upon the types of extraction, seasons, place of collection, and species. The presence of phytochemical compounds and other secondary metabolites in the seaweeds has a greater impact in Pharmaceutical industries[18] for the manufacture of medicines. The phytoconstituents of the seaweed possess a good antioxidant potential and other major biogenic activities.

4.2 Antioxidant Properties

The extracts showed a high capacity for scavenging the free radicals by reducing the stable DPPH (1,1-diphenyl-2-picrylhydrazyl) radical to the yellow coloured 1,1-diphenyl-2-picrylhydrazine. The inhibitory effect of the seaweed extracts was quantity dependent in the range of the tested concentrations. Ascorbic acid was used as standard. The reducing capacity increased with the increasing concentration of the extracts.

Table1- DPPH Radical Scavenging Activity			
S.NO.	Concentration(µg/ml)	% inhibition	
		Padina gymnospora	*Ulva lactuca*
1	10	15.2	17.4
2	20	25.4	29.5
3	30	34.5	39.4
4	40	45.3	49.2
5	50	52.6	56.8
6	60	61.5	64.5

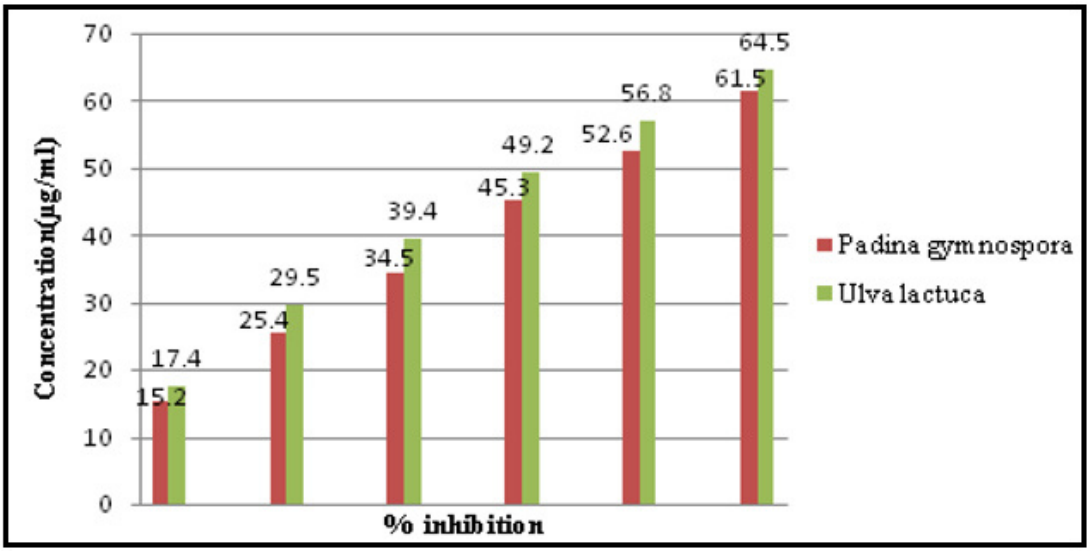

Fig 2: Percentage of DPPH scavenging activity of *Padina gymnospora* and *Ulva lactuca* in methanolic extract

The table1 shows the percentage of DPPH scavenging activity of *Padina gymnospora* and *Ulva lactuca* in methanolic extract. Brown seaweeds have higher antioxidant activity compared to red and green seaweeds. Lower the percentage of inhibition, higher the antioxidant activity. *Padina gymnospora* had maximum inhibition of 61.5 and *Ulva lactuca had 64.5*. From the Fig 4, it shows that *Padina gymnospora* are more scavengers than *Ulva lactuca.*

5. DISCUSSION

The biological active ingredients present in the seaweeds are responsible for various biological activities such as antibacterial, antifungal, anti- inflammatory, antioxidant [19]. This study analyzed the phytochemicals and antioxidant activity present in *Padina gymnospora* and *Ulva lactuca*. The methanolic extracts of seaweeds showed the presence of phytochemicals such as alkaloids, steroids, glycosides, saponin, terpenoids, tannins [20]. Cardiac glycosides, tannins, and Rutin are present abundantly in *Padina gymnospora* making the marine algae more important in the field of drug discovery and developing medicines in pharmaceutical industries.[21]. The presence of bioactive compounds such as fucoxanthin, fucoidan, pholorotannina, sulphated polysaccharides present abundantly in brown seaweeds than other seaweeds. These bioactive compounds are used in the Pharmaceutical industries in treating various diseases. Marine seaweeds have significant importance in therapeutic effects [22]. Terpenoids present in the seaweeds are cytotoxic and has antitumor activity [23]. Tannins are used in the preparation of drugs for the treatment of piles, inflammation [24]. Results of the phytochemical compounds may vary depending upon the solvent extraction, climatic conditions, and harvesting season[25]. The bioactive compounds present in the marine organisms play a major role in the discovery of medicines. Seaweeds are considered as the best source of drugs in the pharmaceutical industries [26]. The phytochemicals, flavonoids, and phenolic compounds are medicinally proven constituents which are widely present in marine organisms. In order to extract the bioactive compounds from the seaweed, naturally produced compounds from the seaweeds has less side effects [27]. Different solvents such as methanol, petroleum ether, chloroform and acetone were used in the past studies. The present study used methanol as the solvent as it gives more efficiency than other solvents [28]. The important role of antioxidants is to fight against the oxidative damage caused by the free radicals due to the external disturbances in the cells. Marine seaweeds can cure non- communicable diseases such as obesity, cancer, diabetes. Seaweeds can decrease the tumoriceffect[29]. Natural antioxidants are available in foods and synthetic antioxidants include Butylated Hydroxyanisole (BHA), Butylated Hydroxytoluene (BHT) is commercially available and cause side effects. Marine seaweeds are the best resource of natural antioxidants. Brown seaweeds have higher antioxidant activity compared to red and green seaweeds [30]. The most increased antioxidant activity was seen in *Padina gymnospora* during summer [31]. The Antioxidant activity of the present study with methanolic extract showed considerable activity under DPPH free radical scavenging activity, total phenol and flavonoid activity [32]. This antioxidant activity mainly depends on the season, environmental conditions and geographical location[33].

6. CONCLUSION

The present study findings reveal that methanol extraction is more capable of determining the seaweed's antioxidant and

phenolic contents. The higher antioxidant activity in seaweed plays a major role in treating various diseases. The phytochemicals present in seaweeds can be used in the pharmaceutical industry for the preparation of drugs.

7. ACKNOWLEDGEMENT

The author expresses their sincere gratitude to Dr Sumaya, Head, Department of Home Science and Research Centre for the valuable support rendered in conducting the research.

8. AUTHOR'S CONTRIBUTION STATEMENT

N.Praba carried out the experiment and wrote the manuscript , Dr Sumaya supervised in writing the manuscript.

9. CONFLICT OF INTERESTS

Conflict of interest declared none.

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